

**MATERNAL SOCIAL CAPITAL AND CHILD
GROWTH:
DO GRANDMOTHERS HELP?**

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**A thesis submitted for the degree of
Doctor of Philosophy**

UCL

DECLARATION

I, Adriana del Pilar Vázquez Vázquez confirm that the work presented in this thesis is my own. Where information has been derived from other sources, I confirm that this has been indicated in the thesis.

Signature:

Date:

ACKNOWLEDGEMENT

For my favourite grandmothers, my mom and my 'mami'

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ABSTRACT

Evolutionary life history theory assumes that the resources available to an organism in any environment are finite and that each organism has been selected to allocate those resources in ways that maximize reproductive fitness. In humans, high levels of parental investment are required to raise offspring, because of their large, costly brains. The costs borne by each individual mother may be mitigated by obtaining social support from other family members. Maternal grandmothers could be a particularly reliable source of support because of the high degree of genetic relatedness and since they represent a valuable source of knowledge and resources, and can release mothers from the 'energetic demands' of childcare.

This thesis tested the over-arching hypothesis that women with grandmothers' support would have more favourable physiological and psychological characteristics that, in turn, would be associated with healthier growth patterns and lower levels of irritability in their children. To test this hypothesis, a cohort of 90 mother-infant dyads (52 with grandmothers' support, 38 without) was recruited in Merida, Mexico. Children were first-borns and aged 1.7-2.3 years. Anthropometric and body composition data and behavioural and physiological markers of stress (salivary cortisol) in both mother and child were assessed. Additional data on maternal resilience and self-efficacy and infant temperament were obtained.

In the whole population, mothers showed high levels of self-efficacy and resilience and perceived their children as having favourable temperament, suggesting that mothers with greater psychosocial capital have a positive impact on their child's development. However, there were no differences between the groups in stress markers or nutritional status among mothers or infants. Notably, mothers without grandmaternal support benefitted from higher levels of support from other female family members. In conclusion, mothers can find different sources of social support for rearing their children, and those lacking a grandmother are not at a systematic disadvantage.

IMPACT STATEMENT

Promoting healthy growth patterns in early life, primarily during the 'first thousand days of life', is a public health priority. Currently, there is a growing interest in the idea that health interventions could also target maternal behaviour, for instance through increasing women's support networks, to improve children's health outcomes. However, there are still few data testing the associations between maternal social support and growth patterns of their offspring.

To address this issue, my contribution links three areas: nutrition, public health and evolutionary anthropology, which could open up exciting new challenges and opportunities for academic enquiry and health policies.

On the one hand, my approach using life history theory and the study of energetic trade-offs at the individual and intergenerational level in individuals of our species is fundamental to evolutionary biology research. My findings may add valuable knowledge about the variability with which females 'strategically' manage energy investment in both reproduction and their own maintenance. The investment strategies that natural selection may have favoured in ancestral female humans are difficult to identify with confidence, however, at the same time they are central to one important question: how can human females produce costly, slow-maturing infants at short intervals in a wide range of habitats? An important evolutionary strategy is the development of the ability to establish and maintain social bonds (Hrdy, 2009). My methodological approach could provide an effective framework for the analysis of how and why such behavioural strategies evolved.

On the other hand, my approach treats maternal phenotype as a composite of many forms of 'capital', all of which may favour investment in offspring (Wells, 2010a). Maternal capital includes social traits, and I proposed this project to differentiate associations of this component of maternal capital with maternal and offspring nutritional status and behaviour patterns. This links with the aim of acquiring pilot data for future 'women's groups' interventions aiming to promote women's well-being and children's growth. In this sense, my research can have applied implications.

Finally, I also wanted to focus on a specific evolutionary approach by identifying grandmothers as critical for the survival of offspring in challenging environments (Sear and Mace, 2008; Hrdy, 2009). I am interested in the idea that postmenopausal lifespan could be considered as an adaptive strategy that has a beneficial impact on child nutritional conditions. With this project, I wanted to extend this idea by obtaining evidence that shows the possible beneficial impacts of this strategy on a mother's health and behaviour.

To improve public health efforts and promote healthy growth patterns among populations it is necessary to address the fact that early exposures are mediated by maternal resources. My study addressed this issue by focusing on the benefits of social support of mothers at a critical time in the life course of the next generation. This project could represent a pioneering epidemiological analysis of associations of maternal social capital and their well-being and that of their children.

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LIST OF ABBREVIATION

ATP III	Adult Treatment Panel III
BMI	Body mass index
CDC	Centre for Disease Control and Prevention
CI	Confidence interval
CONACYT	National Council of Science and Technology of Mexico
CONEVAL	National Council for the Evaluation of Social Development Policy of Mexico
DIF	National System for the Integral Development of the Family
ECBQ	Early Childhood Behaviour Questionnaire
ELISA	Enzyme-Linked Immunosorbent Assay
ENSANUT	National Survey of Health and Nutrition of Mexico
GBP	Pound Sterling
GM	Grandmother
IIF-AS	Iowa Infant Feeding Attitude Scale
INEGI	National Institute of Statistic and Geography
IQR	Interquartile range
ISSSTE	Institute of Security and Social Services of State Workers
ISSTY	Institute of Social Security of Workers of the State of Yucatan
Max	Maximum
Mex\$	Mexican pesos
Min	Minimum
NCEP	National Cholesterol Education Program
NEB	Never Exclusively Breastfed
OD	Absorbance
P25	25 TH percentile
P75	75 TH percentile
PES	Parental Evaluation Scale
PSS	Perceived Stress Scale
RESI-M	Scale of Resilience in Mexicans
SD or \pm	Standard deviation
Sedesol	Ministry of Social Development
TEM	Technical Error of Measurement
TBW	Total Body Water
WC	Waist circumference
WHO	World Health Organization
WHR	Waist-to-hip ratio

DO GRANDMOTHERS HELP?



On the left, a grandmother nurses her granddaughter. Katsuyama, Japan.
Author: Dr Masayuki Nakamichi.

CHAPTER 1

INTRODUCTION

1.1 Background

Child nutritional status in Mexico continues to be an important public health issue especially in the south region of the country. Yucatan has some of the highest rates of child undernutrition (stunting) and of overweight/obesity (OW/OB). According to the last National Nutritional Survey (ENSANUT), in 2012, the prevalence of low height-for-age in school-aged children (1-4 years) from urban communities was 15.8% and 15.1% for OW/OB (Instituto Nacional de Salud Pública, 2012).

There is evidence in the Latin-American population about how early growth retardation increases obesity risk and may permanently modify body fat throughout adolescence and into adulthood (Walker et al., 2002; Li et al., 2003; Varela-Silva et al., 2012; Wilson et al., 2012). In Mexico, there is evidence about the use of interventions for the prevention of undernutrition (Sepúlveda et al., 2006; Rivera et al., 2011) and some of these have been successful through the distribution of nutritional supplements, nevertheless, this not enough to modify the patterns of infant health.

A major challenge for public health efforts is to address the complexity resulting from the fact that early exposures are mediated by maternal phenotype and changes in hormone production. Hence, life history theory can contribute to explaining how growth retardation early in life has implications for body composition since different energy allocation first depends on maternal resources and primarily arises through hormonal pathways. Therefore, my focus on maternal phenotype and hormones is relevant to both components of malnutrition in the offspring.

Broadly, the fundamental tenet of life history theory states that increased allocation of energy into one biological function cannot be achieved without diverting energy from one or more other functions (Hill, 1993). Because available resources in any particular environment are finite, organisms have to face energetic trade-offs that can be thought of as strategies they use to adapt to their environment in order to maximize fitness (Hill, 1993). Broadly, humans must divide their resources across physiological maintenance, reproduction, growth and immune function (Stearns, 1992; McDade,

2003). However, given the high investment that one or both parents may make in their offspring (Trivers, 1972), it is relevant to take into account not only intra-individual trade-offs but also inter-generational trade-offs (Stearns, 1989). Females make a high investment during pregnancy, lactation, and childcare, and must develop strategies to bring their offspring to maturity (Mousseau and Fox, 1998; Reinhold, 2002; Grossman, 2006; Hrdy, 2009; Jasienska, 2009). One of those strategies is to acquire resources from other members of their social group to help meet the costs of reproduction.

The acquisition and transfer of energy resources can reflect the dynamics of social interactions in which the transfer of capital occurs between individuals. Therefore, cooperative breeding allows different members of a social group to contribute indirectly to reproductive effort, thereby supplementing maternal investment in offspring (Reiches et al., 2009).

Evidence has highlighted that the strategy of accumulating and using energy capital is sensitive to social signals (Wells, 2012) and for females, this could represent an advantageous strategy. For instance, associations with kin and non-kin individuals could increase the overall level of care for dependent young and/or decrease the cost of maternal investment. Moreover, social relationships may also provide a benign environment for raising and socializing offspring, which in consequence could enhance children's health and survival (Hrdy, 2009). Thus, the effective use of social support (social capital) can benefit both mothers and their offspring, with potential implications for life history trade-offs.

A growing body of research has demonstrated that cooperative relationships among females may help mothers to maximize their investment in their offspring at a lower cost to their own physiological condition (Wrangham, 1980; Silk, 2001; Gerlach and Bartmann, 2002; Silk et al., 2003, 2009, 2010; Hayes and Solomon, 2004). However, not all social relations can be entirely benign, so it is important to know which relationships are more advantageous for the mother-offspring dyad.

Evidence suggests that mammalian females in general gain fitness advantages from remaining in their natal ranges and associating with their relatives (Pope, 2000; Pomeroy et al., 2001; Wells, 2003c; Hrdy, 2009). This may be because they could feel more confident around kin and therefore be more liable to share the care of individual infants. In humans, according to several reviews, maternal grandmothers

could be the most reliable source of support to mothers for rearing their children (Sear and Mace, 2008; Hrdy, 2009; Meehan et al., 2014). On the one hand, they are on average genetically related to a degree of 50% with their daughters and 25% with their grandchildren (Hamilton, 1964). On the other hand, they could represent an important source of knowledge, and a potential financial and food provider while also releasing mothers from the 'energetic demands' of child care (Beise and Voland, 2002; Sear et al., 2002; Hawkes, 2003; Gibson and Mace, 2005).

In some populations, the presence of the maternal grandmother is correlated with the enhanced well-being of children (Sear and Mace, 2008; Hrdy, 2009), mediated by protecting them from the dangers associated with weaning (Beise and Voland, 2002; Sear et al., 2002), and by helping mothers with heavy domestic tasks (Gibson and Mace, 2005). This means that maternal investment can be extended to maternal grandmothers' investment (Trivers, 1972). Since grandmothers are often post-reproductive, their investment costs are typically lower, while the potential fitness benefits to themselves and those that they help are high (Trivers, 1972). Therefore, grandmothers may represent an 'extrasomatic' source of energy for their daughters.

Although a few studies found that grandmothers have a positive effect on growth in young children (Gibson and Mace, 2005; Meehan et al., 2014; Sheppard and Sear, 2016), there is still very little evidence on this topic. The proposed study will help to fill this gap.

Finally, there is substantial evidence that the greatest period of plasticity is during prenatal and early postnatal life (Smith et al., 1976; Lucas, 1991; Zhu et al., 2004) when the impact of the external environment on infants is buffered by maternal phenotype (Wells, 2003a). The association of grandmaternal support and maternal phenotype is important to consider in order to identify potential effects on children's growth at early stages of life and in addition, to identify potential effects on children's and mothers' behaviour. Mothers that have access to social support could be in a better condition to deal with stressful events, i.e. be more resilient, and hence have the psychological resources (psychological capital) to offer high quality care to their offspring. The present study aims to offer insight into potential effects of social support at both physiological and psychological levels.

I tested the overarching hypothesis that women with maternal grandmothers' support would have more favourable physiological and psychological characteristics that, in turn, would be associated with healthier body composition and lower levels of irritability in their children. To test this hypothesis, a cohort of 90 mother-infant dyads (52 with grandmothers' support) was recruited in Merida, Mexico. All children were first-borns (age 1.7-2.3 years). Anthropometric and body composition data (by deuterium isotope analysis) and behavioural and physiological markers of stress (salivary cortisol) in both mother and child were assessed. Data on other maternal social relationships was also obtained.

In summary, I proposed this project to test potential associations of maternal social capital with maternal and offspring health and behavior patterns. I am interested in the idea that postmenopausal lifespan could be considered a strategy to promote child nutritional status. I extended this idea by exploring a wider range of nutritional and metabolic traits in both mothers and their children and in contrast to most studies of grandmother support, I also took into account other social relationships, in order to address more comprehensively the mother's social network.

1.2 Overview of the thesis

Here I provide a general outline of the thesis:

Chapter 2: This chapter addresses the theoretical framework within which I situated my empirical analyses. Overall, I present a review of evolutionary and social theory and evidence that grandmothers promote parental investment. I used this framework to highlight new connections about how grandmaternal support can potentially drive hormonal changes with potential effects on mothers and children health outcomes.

Chapter 3: This chapter describes the methods and techniques of the study, including the study design and outcome measures. In addition, I provide an anthropological perspective on the research questions and hypothesis. The chapter also provides information about the pilot studies conducted before the data collection began. Finally, I provide a summary of the planned statistical analysis.

Chapter 4: The chapter provides an overall description of the recruitment results and the contextual framework for the research project by describing the living conditions

and socioeconomic status of the families as well as a description of the antenatal care and perinatal data of the mothers and infants.

Chapter 5: This chapter provides a description of the social capital of the mothers. The aim was to identify the members of the mother's personal social network, which may become available to the mother as a result of the history of these relationships. I obtained data about the quantity and quality of the mother's social relationships (family, friends, neighbours and co-workers). This chapter also presents data about the support provided by the maternal grandmother.

Chapter 6: This chapter addresses the primary question and hypothesis of the study and includes the main outcomes related to the nutritional status of the mother-infant dyads with a comparison between those mother-infant receiving support and care from the grandmother and those without this support.

Chapter 7: This chapter addresses the secondary questions and hypothesis of the study. This section includes the results for the physical and behavioural markers of stress in infants and women and a comparison between the grandmother support groups. Physical markers of stress were assessed by analysing both cortisol concentrations in saliva and behavioural markers, through the analysis of self-administered scales provided to the women. Moreover, levels of resilience and self-efficacy about motherhood were described and their associations with the behavioural marker of stress were explored. Finally, information about maternal responses to child behaviour was assessed.

Chapter 8: This chapter addresses an exploratory analysis of intergenerational influences and secular changes among the maternal grandmother-mother-infant triad. This additional analysis of the data obtained in this project aimed to explore associations in linear growth variables among three generations, and analyse the differences in the physical characteristics and environmental conditions experienced by the mothers and grandmothers. This analysis was relevant considering that the growth profile attained by a mother and her recent ancestors, such as the grandmother, may reflect the quality of the conditions experience both in utero and during postnatal growth.

Chapter 9: This final discussion chapter contains a summary of both primary and secondary outcome results. This section also includes the strengths and limitations of my research and the implications of this study. Finally, I provide a conclusion and suggestions for future research.

Further details are described in the introduction of each chapter.

CHAPTER 2

LITERATURE REVIEW

2.1 Introduction

To develop a theoretical framework within which to situate my empirical analyses, this chapter reviews several aspects of evolutionary and social theory. First, I developed an evolutionary framework linking life-history theory with parental investment and social capital. The human life history strategy, with relatively short birth intervals, poses a problem for women as they may need to take care of several dependants simultaneously. Nowadays, most anthropologists agree that females are capable of such reproduction in part due to the support they receive from other members of their social network.

Following this framework, I then present a review of human social capital. Humans are capable of developing the ability to establish and maintain social bonds with different members of their group. The way these bonds can be analysed is complex and subject to criticism, both conceptually and in terms of methodology. However, this overall approach encourages questions and reflections that can help develop studies to investigate the social context of health variability. ‘Who helps’ varies between populations and, hence, understanding the societal structure, norms of behaviour and gender dynamics are all relevant for understanding the dynamic between the social environment and health outcomes.

Because this study stems from the theoretical expectation that other family members can subsidize human care, I also included a review of the patterns of support from relatives, focusing on grandmothers as one important component of maternal social capital. Unusually, human females spend a considerable proportion of their lives in a nonreproductive state and may be available to help mothers in taking care of offspring at a relatively low cost, which could have a greater impact on their reproductive fitness. Evidence has suggested that grandmothers can promote parental investment, as demonstrated by grandchildren's outcomes.

However, the potential benefits of the grandmother for maternal outcomes have been less explored. This issue is worth considering, as during early infancy, the developmental trajectory of each offspring is shaped by signals relating to the

magnitude and quality of maternal capital. Hence, identifying the potential benefits that the mother could receive could help elucidate whether grandparental influences act partly through maternal health. Thus, the question should be, what does the grandmother contribute to the mother and as well as to the child?

Finally, since several hormones, such as insulin and cortisol, play an important role in coordinating life-history strategies, and since grandmother's investment can potentially impact life-history traits of the mother and offspring, I include a review of how social capital can potentially drive hormonal changes with potential effects on health outcomes.

2.2 Life history theory: a brief summary

Life history theory explains how during specific developmental periods, organisms differentially allocate limited resources to several life functions (physiological maintenance, reproduction, growth and immune function). In stochastic environments the available resources may be insufficient to meet all of these functions to an optimal degree, and organisms may never draw on more resources than they have available (Stearns, 1992; Hill and Kaplan, 1999; McDade, 2003).

Then, how should a finite set of resources be spent? And, which investment decisions are the best? To respond to these questions we need to consider that investment strategies depend on individual circumstances and specific information that is gathered from the living environment (Kaplan and Gangestad, 2005). For instance, evidence has shown that organisms that face a high risk of predation reduce their investment in growth and instead increase their allocation of resources to reproduction (Reznick and Endler, 1982). Moreover, it is known that the process of growth is nutritionally and energetically demanding, and is particularly sensitive to diverse ecological conditions, environmental pressures and the mother's capacity to provide resources (Wells and Cole, 2002; Wells, 2007; Pomeroy et al., 2012). Thus, organisms exposed to poor maternal resources during gestation must reorganize their investment to prioritise expensive tissues such as the brain (Aiello and Wheeler, 1995; Yajnik et al., 2003). In this context, the most important resource is energy, and life history theory is primarily a theory of energy allocation.

Selection will favour organisms' energy allocation strategies that result in fitness maximization. However, due to limited energy budgets, it is also to be expected that

selection will favour compensatory strategies that might involve significant cost (Metcalfe and Monaghan, 2001; Kaplan and Gangestad, 2005). This implies that investing more today means having fewer resources to invest tomorrow, which could have long-term consequences. For example, babies that experienced reduced maternal nutritional supply during pregnancy had to invest their scarce resources in the accumulation of body fat to promote survival (Yajnik et al., 2003). However, this phenotype may persist postnatally and predispose to an insulin-resistant stage and cardiovascular diseases, ultimately reducing long-term survival. Therefore, the 'best' or 'optimal' strategies will depend on the circumstances and resources currently available, but the consequences persist at later life stages.

Fundamentally, life history theory focus on three specific trades-offs that allow organisms to maximize their fitness: present versus future reproduction, the quantity versus quality of offspring, and mating versus parenting effort (Hill, 1993; Kaplan and Gangestad, 2005). Once an organism allocates energy to producing new individuals, additional trade-offs emerge. Now energy must be allocated not only to the individual's own life functions (*intraindividual* trade-offs) — for example between a female's reproductive effort and her subsequent survival — but also to the life functions of their offspring (*intergenerational* trade-off) — for instance, between a female's survival and the probability of her offspring surviving and breeding. This indicates that any change in an organism's allocation strategy at the intra-individual level is expected to have implications for their offspring, especially in those species where parental care and provisioning are important (Stearns, 1989).

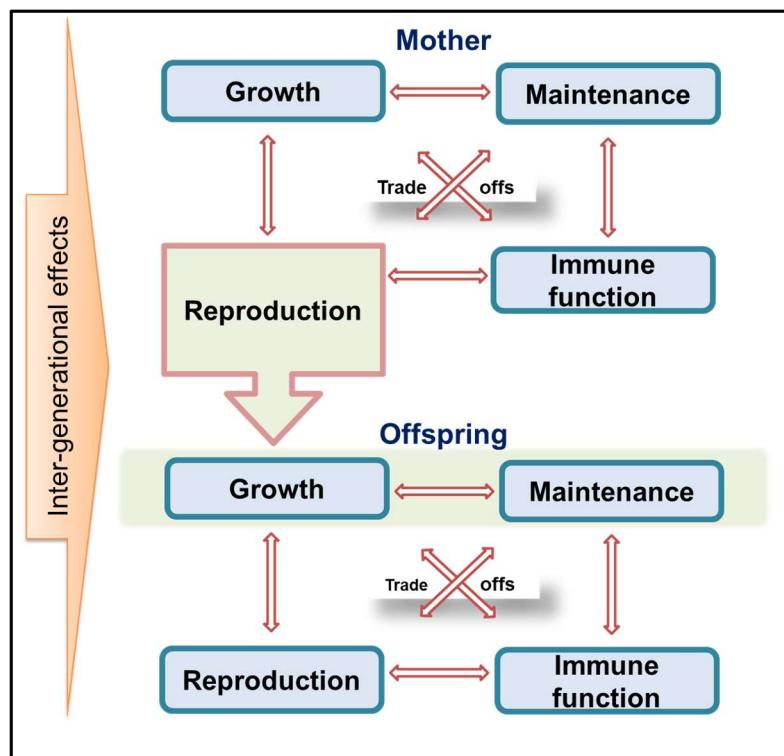
2.3 Parental investment

In evolutionary biology and behavioural ecology, parental investment is defined as any parental expenditure, whether in terms of food, shelter, time or care, that enhances the offspring's survival and their long-term ability to reproduce while decreasing the parents' ability to invest in other offspring (Trivers, 1972). It can be provided at any stage of the offspring's early life, for instance by egg guarding or incubation and placental nourishment during prenatal development, and through food provisioning and protection of offspring during postnatal development (Wolf et al., 1988; Keller and Chasiotis, 2007).

Due to sexual selection (Trivers, 1972), males and females tend to contribute unequally to parental investment (Kaplan and Gangestad, 2005). In approximately 95% of mammalian species, investment is greater for females (Clutton-Brock, 1991; Clutton-Brock and Vincent, 1991) due to the higher demands during pregnancy, lactation and because they provide directly or indirectly the majority of postnatal nutrition. In addition, in humans, parental investment is often culturally based, and women are assigned the role of the main caregiver, leading to greater responsibility for childcare (Mousseau and Fox, 1998; Reinholt, 2002; Grossman, 2006; Keller and Chasiotis, 2007; Jasienska, 2009). Hence, maternal investment strategies for reproductive success are critical, since the production of each offspring is energetically demanding (Figure 2.1).

For the offspring, allocation to maintenance and growth is sensitive to this maternal investment strategy during early stages of development, such as the first thousand days of life, generating life-long effects on health (Wells, 2019) (Figure 2.1).

Figure 2.1 Trade-offs in energy allocation



The energy budget of offspring is determined by the maternal intra-individual trade-offs. During the first thousand days of an offspring life, energy allocation to growth and maintenance is sensitive to the maternal allocation, generating life-long effects. (Figure taken and modified from: Wells 2019)

Male parental investment is increasingly recognised, although the direct effects of fathers' contributions are difficult to disentangle from the maternal contributions (Geary, 2007; Keller and Chasiotis, 2007). For instance, an analysis of mortality risk in early twentieth-century England and Wales suggested that child survival was strongly associated with the employment position of the father (Reid, 1997). Children of professional fathers had 54% lower mortality rate than children of unskilled labourers. After controlling for confounders (socioeconomic status, environmental settings and maternal age at birth), children of working women had 34% higher mortality rate than children of women who were not employed. Although the lower mortality rates could be attributed to male investment due to the high income provided to the family, it is also necessary to consider that women married to professional workers with sufficient income could often stay at home to breastfeed, which could lower infant mortality rate (Rollet, 1997). Male provisioning is not always necessary for offspring survival and its value depends on social and ecological conditions. However, when male investment occurs, its quality and quantity could affect the well-being of the offspring (Geary, 2007).

Parental investment is a complex process that affects not only the survival of offspring but also their productivity and health as adults and their ability to reproduce. The cumulative result of such investment over time is the acquisition of a sum of traits (capital) that are embodied in the soma (Kaplan et al., 2009; von Rueden et al., 2015). Hence, the quality of offspring is an important function of parental investment (Kaplan and Bock, 2001).

2.3.1 Parental investment on embodied capital

Under Kaplan's model, embodied capital theory generalizes existing life-history theory by treating life function processes (physiological maintenance, growth and development) as investments in 'stocks' of somatic or embodied capital. According to this model, embodied capital includes *physical traits*, such as muscle, digestive organs, brains and immune competence, and *functional traits*, such as strength, knowledge, skill, immune function and other qualities like social networks (Kaplan et al., 2003; Lancaster and Kaplan, 2010). According to this approach, the largest and most variable aspect of parental investment is often embodied capital, which is required to obtain resources in stochastic environments (Hill and Kaplan, 1999; Lancaster and Kaplan, 2010).

Reproduction is costly and provisioning can vary from a few weeks or months, such as in bird species or, to years after weaning as in humans. Humans are characterized by a long period of juvenile dependency and slow physical growth (Wood, 2017) requiring increased parental investment, especially from the mother. This means that in many species, mothers store capital in advance to fund reproduction.

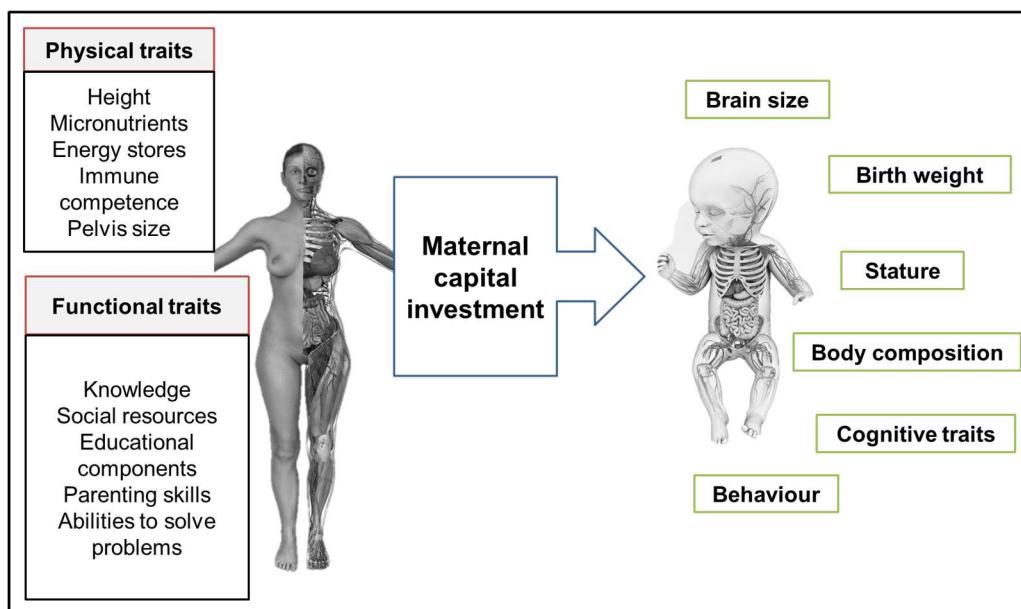
Mothers invest a substantial amount of resources in their offspring before they are even born. During pregnancy, maternal physiological condition is closely related to the growth and development of the foetus, especially of the brain. For instance, epidemiological studies showed the negative effects of foetal undernutrition on the growth of some abdominal organs such as kidney, spleen and liver. General malnutrition of the foetus due to maternal undernutrition or placental insufficiency and preferential blood flow to the brain and heart may deprive these organs (Latini et al., 2004). Then, after birth, the mother provides postnatal nutrition through breastfeeding. Breast milk is the primary source of nutrition for new-borns and provides antibodies, such as immunoglobulin-A, that protect them from infections (Keller and Chasiotis, 2007). The quantity and quality of breast milk depend on the mother's resources. For example, iodine and fatty acid profiles of breastmilk are influenced by the maternal diet (Milligan and Bazinet, 2008; Brough, 2014), and lactation efficiency may be influenced by physiological and behavioural communication between the mother and her infant. The pattern of demand signalled by the infant, and how the mother responds to this signal, could influence milk synthesis and composition (Wells, 2003b; Hinde, 2013). A recent randomized trial found that reducing maternal stress, through a relaxation intervention, influenced infant behaviour, breast-milk cortisol concentrations and milk volume, and subsequently infant weight gain (Shukri et al., 2019).

Since offspring are sensitive during the first thousand days to the magnitude of this investment, maternal effects contribute a major non-genetic influence on the phenotype of the offspring (Mousseau and Fox, 1998; Roff, 1998; Reinhold, 2002; Wolf and Wade, 2009). To explore the dynamics of maternal effects, Wells (2010a), building on the embodied capital model of Kaplan et al. (2003), developed the maternal capital hypothesis.

2.3.2 Maternal capital hypothesis

The maternal capital approach treats maternal phenotype as a composite of many forms of capital, all of which may favour investment in infants and children. These include physical traits such as height, micronutrient and energy stores, but also social traits (support networks and kin assistance) and educational components (Kaplan et al., 2003; Wells and Marphatia, 2018) (Figure 2.2). This hypothesis also acknowledges that mammalian offspring are not directly exposed to the external environment until birth, and in nutrition terms until weaning commences; rather, during early critical windows of development, offspring development is shaped by signals indexing the magnitude and quality of maternal capital. This makes maternal phenotype the main developmental influence on health (Wells, 2010a, 2019).

Figure 2.2 Components of maternal phenotype available for investment in the offspring



The first thousand days of an offspring's lives are shaped by the signals received about the magnitude and quality of maternal capital. The variability in maternal resources will be reflected in offspring phenotype. Adapted from: Wells 2016.

For instance, there is evidence that during exposure to famine during pregnancy, women experienced substantial weight loss (Stein et al., 1995), whereas the offspring only experienced a mean birth weight reduction of less than 10% (<300 g approximately) (Lumey et al., 1993). This means that the offspring was not exposed directly to the external ecological and social stresses of the famine, rather it responded to the variability in maternal capital (Wells, 2016). However, the timing of

exposure is an important factor to be considered when analysing the magnitude of these associations (Harding and Bocking, 2001; Altshuler et al., 2003).

Hence, if the mother has 'sufficient' capital to maintain her physical and physiological condition and respond favourably to her offspring's demands, this will be reflected in both the survival and optimal development of the offspring, as well as the mother's own survival and future reproduction (Hill, 1993). For example, fat storage accumulated during adolescence and pregnancy contributes to the energy demands of pregnancy and lactation (Lassek and Gaulin, 2006; Wells, 2010b). In well-nourished woman, the relatively larger lower-body fat stores provide sufficient energy to fund pre-and postnatal brain growth of the offspring, while a net gain in fat during each succeeding pregnancy allows the mother to recoup this physical capital for additional reproductive events (Lassek and Gaulin, 2006, 2008; Wells, 2010b). Nevertheless, it is not easy for mothers living in poor environments to maintain positive capital balance (Barbosa et al., 1997; Lassek and Gaulin, 2006; Jasienska, 2009). Low accumulation of fat stores, due to inadequate diets or a high burden of disease, may lead to deteriorating nutritional status of the mother across successive pregnancies and in consequence, constrain brain growth in the current and subsequent offspring (Lassek and Gaulin, 2006; Wells, 2010a). Thus, mothers who live in poor environmental conditions may be constrained in their ability to provide energy capital for their offspring, since they still require it for their own physiological maintenance.

Therefore, how can human females produce costly, slow-maturing infants at short intervals in a wide range of habitats and under variable environmental conditions? An important evolutionary strategy that is essential to consider regarding this question, is the development of the ability to establish and maintain social bonds with members of the social group (Capra, 1997; Hrdy, 2009). Maternal capital includes social traits (**Figure 2.2**), and I proposed this project to explore potential associations of this component of maternal capital with aspects of maternal and offspring health and behaviour.

An important aspect of the maternal capital approach is that mothers may interconvert different forms of capital, for example they may use social capital to acquire physical (eg energy stores) and psychological (eg resilience, self-efficacy and satisfaction) resources, and vice versa. This flexibility is likely to contribute to ensuring the nutritional supply of multiple offspring across a mother's reproductive career. For

instance, in cooperative breeding species such as humans, group members can lower the cost of mothering by providing protection, care and provisioning to offspring (Hrdy, 2017), which can allow the mother herself to invest less, and to reserve more resources for future reproduction. Moreover, it has been established that intimate and regular contact with relatives, neighbours, and friends enhances competence as a parent (Pridham and Chang, 1992; Mercer and Ferketich, 1994), and has a beneficial influence on the stress responses (Turner et al., 1999; Light et al., 2000, 2005; Ditzén et al., 2009). The physical and psychological resources obtained through social relationships could potentially improve the way in which the mother responds to the infant's signals (Wells, 2003b).

2.4 Social relationships as a source of capital

According to the previous review of evolutionary life history theory, resources in any given environment are finite. Hence, human females may have developed a wide range of strategies to help them meet the high costs of their large-brained offspring. One of these strategies is the ability to initiate and maintain meaningful social relationships through life. Social capital could be part of the current and potential resources that females can obtain from kin and non-kin that could mitigate the reproductive costs.

Importantly, social capital may play this role in species other than humans. Social capital, defined as an individual's or group's sympathy toward another that may produce a potential benefit, advantage, and preferential treatment (Robison et al., 2002), has been found to be beneficial for female fitness across diverse mammalian species.

2.4.1 What happens in non-human species?

The females of diverse species such as wolves, wild dogs, meerkats, bee-eaters, scrub jays, cichlid fish, papers wasps, monkeys and apes, make key contributions to a range of strategies that enable the rearing of offspring (Hrdy, 2009). The effective use of female social groups is one example of those strategies.

For instance, it has been reported that female baboons who spent more time grooming and associating with others, experienced greater longevity (Silk et al., 2010) and were more likely to rear their infants successfully (Silk et al., 2003, 2009) (**Figure 2.3**).

Figure 2.3 Female baboons associating with others



Female baboons groom one another to cement a close stable relationship

Credit: University of Pennsylvania

Available at: <https://phys.org/news/2016-07-well-connected-friends-benefits-female-baboons.html>

Likewise, experimental studies with female wood mice (*Apodemus sylvaticus*) and female prairie voles (*Microtus ochrogaster*) showed that litters of focal mothers in plurally breeding groups gained more weight than litters reared by solitary mothers (Gerlach and Bartmann, 2002; Hayes and Solomon, 2004). In addition, females in breeding units reduced their investment by significantly reducing nursing time (Gerlach and Bartmann, 2002) or by nursing fewer pups, presumably without adverse effect on pup growth (Hayes and Solomon, 2004) (**Figure 2.4**),

Figure 2.4 Social prairie voles (*Microtus ochrogaster*)



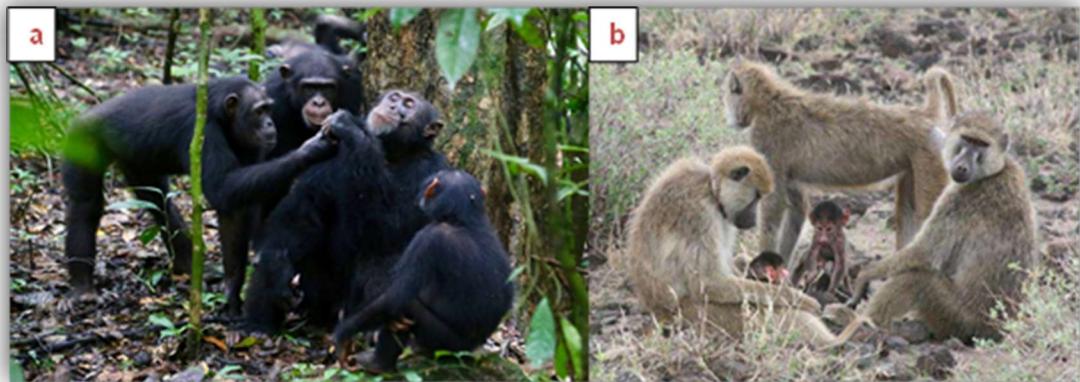
Credit: Karen Bales

Available at: <https://jeb.biologists.org/content/220/1/1>

Moreover, cooperative relationships among females may allow the sharing of information about food sites (Silk, 2001) and also provide opportunities for the exchange of caretaking responsibilities (Wrangham, 1980) (**Figure 2.5**). For instance, in hanuman langurs, offspring were observed to be carried by other females, which impacted the mother's energy budget by reducing the time to the next conception (Hrdy, 2004). Therefore, higher maternal investment in social relationships can increase a mother's reproductive success in species that live in social groups, and may help mothers to maximize their maternal investment at a lower cost to their own physiological condition.

A growing body of research has demonstrated that diverse mammalian species have the ability to establish and maintain social bonds (Gerlach and Bartmann, 2002; Silk et al., 2003, 2006, 2009; Hayes and Solomon, 2004; Hrdy, 2009; Langergraber et al., 2009). Observational evidence shows that female baboons (*Papio cynocephalus*) form very strong bonds with close maternal and paternal kin, especially with mothers, daughters and maternal and paternal sisters, independently of the differences in dominance rank between them (Silk et al., 2006).

Figure 2.5 Females sharing food and caretaking responsibilities



a) Chimpanzees share food with their friends

Credit: Liran Samuni, Taï Chimpanzee Project

Available at: <https://phys.org/news/2018-10-wild-chimpanzees-food-friends.html>

b) Female baboons and their infants in the Amboseli ecosystem in Kenya

Credit: Jeanne Altmann

Available at: <https://phys.org/news/2014-05-offspring-costly-phase-life-baboon.html>

Moreover, unrelated females in several primate species form similar bonds (Baldwin, 1985; Wallen and Tannenbaum, 1997; Langergraber et al., 2009). For example, it has been observed that rhesus monkeys establish social bonds with female peers to provide security (Wallen and Tannenbaum, 1997); and that female chimpanzee (*Pan troglodytes*) tend to form strong social bonds with females that are not closely related (Langergraber et al., 2009) (**Figure 2.6**).

Figure 2.6 Female bonobos interacting with other members of their group



Female bonobos at Wamba, Democratic Republic of Congo
Credit: Nahoko Tokuyama, Kioto University

Available at: https://www.kyoto-u.ac.jp/en/research/research_results/2016/160719_1.html

The above examples indicate that females have the ability to expand their social networks beyond kin relationships, which maximizes the probability that different group members (cooperative breeders) will protect them and their offspring (Taylor et al., 2000; Hrdy, 2009). This kind of social system, characterized by alloparental care, which is defined as 'the provisioning of care by individuals other than the young's biological parents' (Kenkel et al., 2017), is believed to be the strategy that permitted our ancestors to produce costly, slow-maturing infants at shorter intervals in a wide range of habitats (Hrdy, 2009). Larger brains may then promote alloparental care through a positive evolutionary feedback process, as across mammalian clades, there is a positive correlation between brain size and the amount of alloparents (Isler and van Schaik, 2012).

Alloparental studies conducted in rodents, such as prairie voles, has shown that among the behaviours displayed by the alloparental vole it is possible to observed pup retrieval, licking and grooming, and arched-back huddling (Kenkel et al., 2017), which, according to previous report (Roberts et al., 1998; Lonstein and De Vries, 2001), are not different from behaviours observed in parents. Kenkel et al (2017) stated that the purposes of these behaviours have been not elucidated to date; however, they speculate that could serve to keep the pup safe, warm and calm in the nest.

In addition, the reasons that motivate alloparental voles to care for a pup have also not been elucidated, so it is not possible to determine whether these behaviours are expressed because they provide a rewarding experience, or whether the distressed pup is perceived as a stimulus to be alleviated through the provision of care (Kenkel et al., 2017).

Nonetheless, the above findings are relevant, considering that other research in animals has shown the potential benefits of behaviours shown by non-biological parents. For instance, in cross-fostering experiments, pups reared by high licking and grooming rat dams exhibit high licking and grooming behaviour when they themselves become mothers, in contrast to pups reared by low licking and grooming dams (Francis et al., 1999). These positive behaviours are similar to those observed in young mammals who received more maternal grooming and contact (Champagne and Meaney, 2007; Champagne, 2008; Ross and Young, 2009).

Hence, investing in social relationships could be highly advantageous for females and their offspring. Non-maternal caregivers can provide security and resources to offspring and increase levels of care for dependent young, which may then decrease the direct costs of maternal investment.

Forming social bonds could be complex and according to my review, hormones play an important role in social behaviours. Several research studies across diverse mammalian species have shown that the hormone oxytocin plays a role in the process of bond formation, and functions to reduce stress, enhance social competence, initiate maternal behaviour, and promote social affiliation through life (Francis et al., 2000; Gimpl and Fahrenholz, 2001; Pedersen, 2004; Ross and Young, 2009; Waldherr et al., 2010). In rodents, endogenous oxytocin makes 'social approach' possible, as well as acting to reverse stress-induced social avoidance (Lukas et al., 2011). Similarly, male and female marmosets (*Callithrix penicillata*) who received intranasal oxytocin treatments showed enhanced initial partner-seeking behaviour and initiated more contact with their partners; conversely, it has been observed that oxytocin antagonist treatment decreased the frequency of social approach in primates (Smith et al., 2010). In this sense, oxytocin has a social role by suppressing the stress response and by facilitating social motivation and approach to novel others (Carter and Altemus, 1999; Heinrichs and Domes, 2008).

Living in groups, then, is an important evolutionary strategy for primates (Caporael, 1997). However, not all social relations may represent a reliable source of support, so it is important to know which relationships are more advantageous for females and their offspring. Evidence suggests that mammalian females gain fitness advantages from remaining in their natal ranges and associating with their relatives (Pope, 2000; Pomeroy et al., 2001; Wells, 2003c; Hrdy, 2009). This may be because females could feel more confident around relatives and therefore be more liable to share infants with other females, thus gaining alloparental support (Hrdy, 2009).

2.5 Social capital in human species

As in non-human animals, humans are also able to use their resources ('capital'), whether physical (e.g. height, lean mass, adiposity) or social (e.g. supportive relationships), to protect their offspring as well as themselves against uncertainty in the environment (Wells, 2010a). Children require investment from several adults to enhance survival in the early stages of childhood and maintain healthy development and mothers could bear some of the energetic costs of reproduction if they obtain support from other adults (relatives or non-relatives) (Sear et al., 2000, 2002, 2003; Aubel et al., 2004).

Hence, understanding the dynamics of social networks in human can help us to determine how resources provided by these networks can increase reproductive success in women.

2.5.1 Social capital: a brief review

Bourdieu defined social capital as 'the sum of the actual or potential resources linked to possession of a durable network of relations more or less institutionalized of mutual recognition' (1986, p 248). According to Coleman (Coleman, 1988), social capital is defined by its functions: 'they all consist of some aspect of a social structure, and they facilitate certain actions of individuals who are within the structure'. These functions include obligations, expectations, trust, and information flows. For Putnam (1993), social capital refers to 'connections among individuals – social networks and the norms of reciprocity and trustworthiness that arise from them'. Finally, the World Bank (Serageldin and Dasgupta, 2001) has conceptualized social capital as 'the institutions, relationships, and norms that shape the quality and quantity of a society's social interactions'.

Even though there are similarities among the several concepts coined, social capital can still be considered conceptually weak, given that economists use the word 'capital' to discuss financial resources and goods that make individuals more productive when they are used in combination with other forms of capital (Robison et al., 2002). Several criticisms have focused on the term 'capital' implying a delivered investment or an opportunity cost in the present that provides future benefits, something that social capital lacks (Robison et al., 2002).

However, social capital can imply a cost today that can produce potential benefits in the future and shares essential capital-like properties. First, social capital has a transformation capacity. For instance, social relationships can supply social need and validation (Robison et al., 2002). Positive communication with, and supportive feedback from, those you have a close relationship with can provide emotional resources, such as self-acceptance, that can be used to cope with stressful events. Second, social capital holds different degrees of durability (Robison et al., 2002). For example, social capital can reside in family members and friends. The first category is mostly durable whereas the second could be characterized by limited durability, depending on the context. Both relationships need an initial investment and probably depending on the costs and the benefits received, investments in maintenance could continue. Third, social capital could be flexible and allow access to a variety of services such as financial resources, transportation, companionship and knowledge/information. Fourth, social capital has a reliability dimension (Robison et al., 2002). For instance, relationships with family members could be highly reliable and their performance predictable. Fifth, social capital can be used to create new forms of social capital or strengthen the existing forms (Robison et al., 2002). Finally, it is important to consider that social capital can be inherited and that investment processes are important to maintain this capital (Robison et al., 2002). For example, when a child is born, the individuals of her/his parents' social networks, such as the family, immediately become part of her/his social capital. Later, as the child ages, the maintenance of this capital will depend on the investment he/she makes, e.g., the intensity of the communication and frequent interactions with networks.

Nevertheless, despite possible discrepancies in the way of defining and evaluating its properties, social capital always points to those elements that bring us closer as individuals, and to how this turns into opportunities for cooperative action and the well-being of a group.

2.5.2 Measuring social capital

To measure social capital, most studies consistently use three key indicators established by social capital theorists. First, *trust* seems to be one of the most relevant and is defined as 'a firm belief in the reliability, truth or strength of a person; a confident expectation; and a reliance on the truth of a statement without examination' (OED, 1994). Trust is a collective attribute and is applicable to the relations among people (dyads, groups, and collectivises). The two components of this indicator are *trust in others* and *trust in institutions* (government, police, etc.) (Lewis and Weigert, 1985; Kemenade, 2003).

The second indicator is *civic engagement*, which is measured by asking the individual or a group about their participation in a network or organization. It can relate to participation in community activities, such as sports leagues or parents associations, or political life, such as voting in elections (Kemenade, 2003).

Finally, the third indicator is *social networks*, which refer to 'the web of social relationships that surround individuals' (Heaney and Israel, 2008). They are formed by the individual's immediate environment, such as family, friends and neighbours, by secondary networks, which include relationships that an individual establishes in the workplace or during community activities, and formal networks, like health care professionals and human service workers (Kemenade, 2003; Heaney and Israel, 2008).

Moreover, social capital can be analysed using two components: a) a behavioural component, concerning what people do, such as being a member of networks (structural social capital), and b) a perceptual component, concerning what people think and feel, such as developing notions of trust, reciprocity and support (cognitive social capital) (Bain and Hicks, 1998).

One of the central functions of social networks is the provision of social support, which can be categorized in four types of supportive behaviours:

- a) *Emotional support* that may involve the supplying of trust, caring and empathy.
- b) *Instrumental support* which includes providing direct tangible services that assist an individual.
- c) *Information support* that involves the provision of knowledge and advice that could help a person to solve a problem.
- d) *Appraisal support*, which refers to the provision of information that a person could use for self-evaluation (Heaney and Israel, 2008).

Structural social capital can refer to ties between people who share similarities, or to linkages among people who are different, such as individuals from outside one's community (Harpham, 2006). All measures refer to the community; however, current research also views social capital as a property of individuals.

Individual social capital can be defined as 'the collection of resources owned by the members of an individual's personal social network, which may become available to the individual as a result of the history of these relationships' (Van der Gaag and Snijders, 2004). At the individual level, social capital can be measured by asking an individual about the quantity and quality of own networks (structural) and about whether they are trusting (cognitive) (Kemenade, 2003). This unit of observation of social capital is useful considering that the decision to invest in social relationships is first made by individuals (Glaeser, 2001).

Nevertheless, measuring social capital is not an easy task considering that a given study in any cultural setting needs to be sufficiently contextualized. It is necessary to understand the societal structure and organization, the norms of behaviour and gender dynamics, among other elements, in order to develop tools to measure social capital in specific settings.

For the present study, I wanted to focus on the association between individual social capital and health outcomes. In particular, I am interested in how women's social relationships may be associated with their health status and that of their children.

However, as I stated before, measuring social capital associations is complex and subject to criticism, both conceptually and in terms of methodology. I am not aiming to look for causality between maternal social capital and health conditions because I

am aware that health is related to multiple factors such as economic, political and cultural factors. Nonetheless, this approach encourages questions and reflections that can help develop studies to investigate the social context of health variability, and to improve health policies and interventions through the understanding of the dynamics and composition of the societies.

2.5.3 Maternal social capital. Establishing social bonds: With whom?

In the previous section, I presented evidence on social capital in animals that provides support for the hypothesis that social capital boosts female investment, with benefits for offspring. In humans, we can investigate these potential benefits considering that parents do not parent alone, and it is likely that those who make effective use of their social group may be more successful in maximizing offspring survival. For women, associations with relatives and non-relatives individuals could increase the overall level of care for dependent offspring and/or decrease the cost of maternal investment. Moreover, social relationships may also provide a benign environment for raising and socializing their offspring, which in consequence could enhance children's health and survival (Sear et al., 2000; Taylor et al., 2000; Sear and Mace, 2008; Hrdy, 2009). Thus, the effective use of their social support (maternal social capital) is potentially an advantageous strategy that can benefit both mothers and their children.

It has been identified that humans are cooperative breeders, which means that maternal care is often supplemented by other individuals that can improve offspring development, health and well-being (Hrdy, 2009). Such alloparents might include the father, grandparents, uncles, aunts and older children who, according to kin selection and parental investment theories, can increase their inclusive fitness by investing (time, energy and resources) on their kin (Hamilton, 1964). Hence, it is expected that the closer the relatives (ie higher proportion of shared genes), the greater the investment they should be willing to provide. However, not all kin are equally helpful (Sear and Mace, 2008), making it important to determine which supportive social relationships are more beneficial for the mother and their children across populations.

2.5.3.1 None-relatives

A. Friends

As a part of an individual's social network, friends could be supportive and contribute resources (Flaherty and Richman, 1989; Voorpostel and Van Der Lippe, 2007; Gameiro et al., 2010) through playing a unique role in fulfilling specific needs. For instance, friends can have the role of being companions (Wellman and Wortley, 1989) and confidants for discussion of personal issues, such as becoming a parent, (Tokuno, 1983). However, in some cases, friends are not considered as attachment figures (i.e. providers of reliable alliance and affection) (Carbery and Buhrmester, 1998).

Support and companionship from friends can vary according to the phase of life (single, married or married with children) of individuals (Fischer and Oliker, 1983; Carbery and Buhrmester, 1998). For example, in a cross-sectional study of undergraduate students, Carbery and Buhmester (1998) found that ratings of companionship and being a confidant (emotional support) with friends were higher for the single phase and significantly lower in the marital and parenthood phases. According to the researchers, their findings are consistent with the view that when individuals get married, the spouse 'replaces' friends as the primary source of intimacy and support.

Hence, it is probably expected that friends could provide support, but not regularly as might be expected and received from other groups of individuals, such as close relatives.

2.5.3.2 Relatives

A. Siblings

Siblings can be an important source of support, love and companionship, and help during stressful circumstances (Eriksen and Gerstel, 2002; Voorpostel and Van Der Lippe, 2007; Cicirelli, 2013; Riedmann 1992), and a high relationship quality and contact frequency between siblings has been related to increased exchange of emotional support (Voorpostel and Van Der Lippe, 2007). For instance, through the analysis of longitudinal data from the study of the 'Four stages of life' carried out by Lowenthal, Thurnher, and Chiriboga in 1975, Paul (1997) found that sibling interactions and the quality of their relationship affected their well-being. Conflict with siblings was associated with a more negative self-concept and with more

symptomatology, whereas positive interactions and more contact with siblings were related to less symptomatology and fewer feelings of loneliness.

However, support between siblings can be influenced by family structure and solidarity. For example, in a cross-sectional study in which the support of parents was considered to measure the support exchanged between siblings, results showed that when the relationship with the parents was poorer, more support was exchanged between siblings. It appears that siblings compensate for a poor relationship with a parent by turning to each other. The results of this study varied by gender, with women more likely to exchange emotional support with sisters and to increase support when they experience a more positive relationship with parents (Voorpostel and Blieszner, 2008).

It seems that support from siblings could be contingent on the relationship with the parent. According to my review, if siblings have a poor relationship with parents, their bond becomes closer, but what happens if they have a good relationship with them? It appears that support from siblings, as with friends, may not necessarily occur regularly.

B. Uncles and aunts

As part of the extended family, uncles and aunts could represent a source of instrumental, affective and emotional support for women and their children. For instance, never-married aunts and uncles may have the opportunity to take relevant and active roles in the life of their nieces and nephews. They could be willing and able to provide parenting to their siblings' offspring (Langer and Ribarich, 2007).

Because most of the literature reviewed noted that relationships between sisters tend to be closer than those between brothers (Rosas, 2000; Voorpostel and Blieszner, 2008), I wanted to focus on aunts as a possible reliable source of support for nieces and their children.

Kin keeping has been traditionally associated with feminine roles, and mothers, grandmothers and aunts are most likely to provide kin support (Leach and Braithwaite, 1996; Garey and Hansen, 1998). Moreover, in some circumstances, aunts can be seen as 'second mothers'. For instance, in kinship studies it has been found that in the absence of grandmothers, aunts often assume the role of foster caregivers for

children removed from their parents' custody (Davidson, 1997; Thorton 1991). Furthermore, a study focusing on exploring the perspectives on communication with aunts reported that nieces/nephews identified aunts as teachers, role models, confidant and advisors, and second mothers. Participants valued the emotional support provided by aunts, and described them as nurturing mothers, due to receiving guidance, direction, and encouragement from them, which appeared to positively influence the nieces/nephews' self-esteem (Ellingson and Sotirin, 2006).

Relationships are connected, and if we considered that aunts are related to nieces/nephews through their parents, the closer relationship between sisters (mothers and aunts) (Rosas, 2000; Voorpostel and Blieszner, 2008) could be transmitted to daughters/nieces, which could represent an important source of support, mainly when the mother is absent.

C. The partner

An important member of the mother's social network is the partner. The formation and maintenance of a pair-bonding relationship is a valuable example of cooperation between two individuals, especially in monogamous species (Soares et al., 2010). Cooperation between pair partners is particularly relevant for breeding success (Royle et al., 2012), emotional (M. Diamond, 2000; Misri et al., 2000; Grewen et al., 2005a; b; Light et al., 2005) and economic support (Townsend, 2010), and childcare assistance (Quinlan and Quinlan, 2008; Raley et al., 2012). However, the quality of the couple's relationship plays an essential role in health outcomes (Ebrahim et al., 1995; Coyne et al., 2001), with high levels of marital dissatisfaction associated with enhanced health risks (Baker et al., 2000; Orth-Gomér et al., 2000) and stress (Carels et al., 1998).

For women, a partner who seeks a close involvement in childcare (and probably housework) and not only seeks to fulfil financial provision could represent a valuable source of support that releases them from the energetic demands of motherhood. However, as I stated before, the quality of the relationship plays an important role hence, good communication between the couple and satisfaction with family task arrangements are relevant factors for maintaining marital quality, especially after the birth of children.

Nowadays, men have increased their participation in domestic work and invest more time in their children than before (O'Brien and Shemilt, 2003; Kan et al., 2011). These changes may be associated with the fact that currently, women have more participation in the labour market, and that couples with more egalitarian attitudes towards gender tend to have a more equitable division of domestic tasks than couples with more traditional gender visions (Greenstein, 1996; Pittman and Blanchard, 1996; Morrell et al., 2016). However, evidence suggests that women continue to do more domestic work than their partners (Bittman et al., 2003; Risman, 2011; Lyonette and Crompton, 2014) irrespective of their working hours and earnings (Lyonette and Crompton, 2014).

Moreover, fathers' contribution to infant survival is small. According to the review of Sear and Mace (2008), fathers frequently make no difference to child survival and the support provided by them varies substantially across populations in association with several factors, such as the availability of resources and degree of paternity certainty.

Women in pair-bonding relationships certainly seek and expect support from their partners and there is evidence of the importance of paternal investment among certain human groups (Hill et al., 1985; Hewlett, 1992; Hurtado and Hill, 1996). However, this support could vary substantially (Sear and Mace, 2008) and the specific contribution of the father cannot be guaranteed (Hrdy, 2009). Hence, what could be the solution for this 'lack of security' about partners support? Which other relatives could give more assistance to mothers and their children?

According to Hamilton (1964), it seems that the most likely candidates are close relatives of the child and the mother, such as grandparents. There is consistent evidence of beneficial effects of grandparents on the survival and health status of children (Sear and Mace, 2008). Moreover, according to the available evidence, support from maternal kin, especially grandmothers, could be more reliable (Sear et al., 2000, 2002; Gibson and Mace, 2005).

D. Grandparents

In recent years, parenthood has gained considerable attention. Studies have shown that by providing informal childcare support, grandparents facilitate working parents - particularly mothers – to accomplish a suitable work-life balance (Wheelock and Jones, 2002; Bishop et al., 2015; Gray 2005). However, which of the maternal

and paternal grandparents engages more in childcare and support could be influenced by the imbalance in paternity certainty, which triggers differential grandparental solicitude (Euler and Weitzel, 1996).

Several studies have demonstrated that grandparents assist their grandchildren in ways that may increase child survival and appear to support the paternity certainty factor, in that the maternal grandmother is the most likely to assist, followed by the maternal grandfather and the paternal grandmother and grandfather (Jamison et al., 2002; Sear et al., 2002; Hawkes et al 1989; Blurton-Jones et al 1989).

▪ **Grandfathers**

A recent study reported that the traditional understanding of grandfatherhood – i.e. distance and less involvement due to traditional male roles (Thompson et al., 1990; Roberto and Stroes, 1992; Hummel et al., 1995) – is changing, and a more involved type of grandfatherhood is emerging. There are several studies carried out in the United Kingdom that examined the ways in which grandfathers contribute to the care of their grandchildren (Wheelock and Jones, 2002; Mann and Leeson, 2010). Qualitative data have shown that although grandfathers perceived their contribution to child-caring as secondary in comparison to grandmothers, there are particular situations in which grandfathers become active and take on more primary roles with grandchildren.

For instance, the adoption of an active role could be related to grandmothers being engaged in full-time or part-time jobs so that it may be necessary to evenly distribute care duties (Mann and Leeson, 2010). These findings illustrate the dyadic nature of grandparenting, where grandfathers - mainly maternal grandfathers - performed caring duties as part of their partnership with the grandmother (Wheelock and Jones, 2002).

However, evidence of associations between the presence of grandfathers and child survival and nutritional status is scarce, and the findings give mixed messages. Gibson and Mace (2005) found no association between the presence of the maternal and paternal grandfathers and child height and weight. Moreover, grandfather's presence did not influence child survival, which could mean that they play a lesser role in determining child well-being (Gibson and Mace, 2005), which agrees with

studies that found that grandfathers seem to be less important to children (Sear et al., 2000, 2002; Sear and Mace, 2008).

Although more studies are needed to understand grandfather effects, until now, grandfathers' (maternal or paternal) contribution to infant survival appears to be small and variable, and it seems that an important factor in the establishment of their support is the presence of the grandmother-especially the maternal grandmother-, which still puts in doubt the reliability of their support.

- **Grandmothers**

Some authors have argued that investment from matrilineal kin has driven the evolution of human life history (Hawkes et al., 1998). Older women, such as grandmothers, could be responsible for caring for younger children while their mothers are engaged in subsistence task or taking care of another offspring, which could have a greater impact on their reproductive fitness by investing in their children and grandchildren rather than by continuing producing new offspring. Hence, to understand why elderly women are a common source of support for mothers across human societies, we need to understand why female humans typically live long enough to take on this role. Therefore, an evolutionary perspective on grandmothers is required.

The Grandmother hypothesis states that the long postmenopausal life span has been selected for specifically because of the support that older women provide to their children and grandchildren, which increases the reproductive output of their children, especially daughters (Hawkes, 2003; Sear et al 2000). However, there is still an ongoing debate as to whether menopause is an adaptation or by-product of something else (Kachel et al., 2011; Lahdenperä et al., 2012).

Even though postmenopausal longevity is not necessarily an adaptation, there is good evidence that older women do provide a significant amount of resources to grandchildren and have a beneficial impact on children survival (Hrdy, 2009; Hawkes et al 1997, Hurtado et al 1992, Kaplan et al 2000) and their nutritional status (Mace and Sear, 2005).

However, maternal and paternal grandmothers may not increase grandchildren's health conditions equally. In their review, Sear and Mace (2008) reported that the

majority of the cross-sectional studies found that the presence of the maternal grandmother is associated with higher child survival rates (Sear 2000; Sear et al., 2002; Beise, 2004; Ragsdale, 2004; Leonetti et al 2005) whereas the effect of the paternal grandmother's presence showed more variation (Gibson and Mace, 2005a; Sear, 2006; Mulder 2007).

Evidence has also shown that the presence of the maternal grandmother has been correlated with enhanced well-being of children by helping mothers in heavy domestic tasks (Gibson and Mace, 2005). Meanwhile, the presence of the paternal grandmother has been correlated with increased maternal fecundity, earlier reproduction, and shorter intervals between births (Hrdy, 2009). Such an increase in maternal fecundity may affect the condition of the mother over successive pregnancies and therefore increase child mortality (Sear and Mace, 2008). However, in populations in which patrilineal inheritance is important, the presence of the paternal grandmother was correlated with more help in keeping infants alive, even though the shorter birth intervals probably takes a toll on the mothers (Hrdy, 2009). According to Gibson and Mace (2005), paternal grandmothers are more likely to help women with activities from which they may gain a direct benefit.

Much of the literature considers that the greater investment of the maternal grandmother is due to the degree of genetic relatedness with the mother and child. However, it is also important to consider that a possible explanation for this variability could be related to the distinct roles that grandmothers play across populations according to cultural characteristics and beliefs, and to a range of factors such as residence patterns, local subsistence conditions, socioeconomic status, family composition and inheritance patterns, which are more important for settled populations (Hrdy, 2009).

Overall, maternal grandmothers appear to be more reliable helpers and have the strongest, most frequent and most positive effects on child survival and health than paternal grandmothers (Sear and Mace, 2008).

2.5.4 What about social support in Mexico?

According to my review, although there is evidence about the benefits of the support provided by social networks to mothers and children, there is little evidence about the patterns of support by social networks in the Mexican population. Moreover, I was not

able to find any study that specifically has the aim to find the effects of social support on children and women well-being.

First, I was able to identify that friends are some of the important social ties for Mexicans, however, support from friends depends on the level of intimacy and only when friendships are very close might the exchange of favours be expected (Lomnitz, 1994; Kana'laupuni et al., 2000; Rosas, 2000; Kana'laupuni et al., 2005). This is consistent with the perspective provided in **section 2.5.3 (A)**, indicating that support from friends could be not expected on a regular basis.

Then, there is evidence that in Mexico the relationship between siblings represents an important link in the daily life of individuals (Rosas, 2000; Palomar Lever and Cienfuegos Martínez, 2007) and for women, this bond is usually closer with sisters (Rosas, 2000). However, the potential benefits of this support are not clear. On the one hand, there is evidence suggesting that regardless of socioeconomic level, the main source of social, instrumental, and emotional support come from parents and siblings. On the other hand, studies have found that individuals with more socioeconomic disadvantage perceived less support from parents and siblings (Palomar Lever and Cienfuegos Martínez, 2007). Hence, the evidence suggests that the scarcity of resources could be one factor that limits the support between siblings. More studies are needed to find a clear pattern about siblings support and its benefits.

Regarding the potential benefits of the support provided by aunts, I was only able to find one study that mentioned the support of aunts under a specific circumstance, migration. In the case of Mexican migrants, women pointed out that their aunt was an important part of their social network when they relocated, tried to find a job and began to establish their new home (Bastida, 2001).

Concerning male provisioning and support, in Mexico, female paid work has favoured flexibility in the division of family work that has allowed an increase in male participation in household chores and childcare. Recent social research suggests that Mexican fathers are showing higher levels of participation in the care of their children and relatively more participation in home activities (Casique, 2008; Martinez-Salgado and Rojas 2016). However, according to the 2014 National Survey on Time Use (in Spanish: *Encuesta Nacional sobre Uso del Tiempo 2014-ENUT*. Available at: <https://www.inegi.org.mx/programas/enut/2014/>) (n= 19,000 households, considering urban-rural populations), women and men (from 12 years to older ages) invest

different amounts of time in childcare and domestic tasks. The results showed that women invest on average 24.9 hours per week in the care of household members between 0 and 14 years, while men only invest 11.5 hours. Regarding participation in domestic tasks, women invest on average 29.8 hrs per week in domestic activities and men, only 9.7 hrs. These results demonstrate, as in other studies carried out in Mexico (Casique, 2000, 2008), that although men are showing more participation, Mexican women continue having the major responsibility of housework and childcare.

Studies about grandfatherhood are even scarcer and the studies I identified were not focused on investigating the role of maternal and paternal grandfathers in the care of their grandchildren. However, the information provided can help us to identify some types of support provided by them. One of the studies focused on the perception of adolescents (n=177) about their relationship and bond with their grandfathers and grandmothers. Overall, 66% of the granddaughters and 46% of the grandsons reported having a better relationship with the maternal grandmother, which could be influenced by the frequency of contact with grandfathers. From this sample, 83% of the participants reported having more contact with the maternal grandfather and 47%, with the paternal grandfather. Moreover, there was a difference in the support received from the grandfathers. A higher percentage of granddaughters (23%) and grandsons (20%) reported that maternal grandfathers were more likely to give advice than paternal grandfathers ($\leq 7\%$ of grandchildren) (León et al., 2016). Additionally, in an intervention study carried out in four communities of Oaxaca, Mexico, it was observed that the transfer of knowledge of traditional production processes occurred mainly between fathers and sons, grandfathers and grandchildren, and uncles and nephews (De la Paz Hernández et al., 2005). Thus, grandfathers could be seen as an important source of knowledge for grandsons on specific activities.

Finally, although there is evidence about the preference for maternal grandmothers as caregivers in the Mexican population (Partidas, 2004), my review did not identify any study carried out that had the clear aim of measuring the impact of maternal grandmothers on their grandchildren's health conditions and, specifically, no evidence was found from Yucatan, Mexico.

I was only able to identify a few studies carried out in Yucatan that mentioned any role of the maternal grandmother in health-related practices. These studies were not focused on knowing the role of the grandmothers in the care of their daughters and grandchildren. However, among their results, they mentioned the grandmother as a

source of support when their daughters needed help in childcare and when mothers needed support in decision-making at home. Additionally, these studies reported that grandmothers acted as a source of information during pregnancy, delivery and first days of life of their grandchildren.

For instance, in the community of Uayalceh, Yucatan, where female migration is frequent, it was observed that maternal grandmothers take care of their grandchildren while the mother (single, divorced or in support of her husband) leaves the community for work reasons (Gutiérrez-Carbajal et al., 2019). In Tizimin, Yucatan, researchers observed that women construct their knowledge about reproduction (in terms of diet, pregnancy risks, diseases during pregnancy, delivery) through talks at health centres and through popular knowledge transmitted from mothers to daughters, and from grandmothers to granddaughters (Rodríguez-Angulo et al., 2012). Moreover, in other communities, it was found that grandmothers play an important role in the nutrition of their grandchildren. In Kiní, Yucatan, grandmothers recommended maternal care practices, such as washing the breasts with hot water and not consuming cold food, to ensure the production of breast-milk in the new mother. These care practices reflect the Mesoamerican hot-cold classification (Reyes-Gutierrez and Cervera-Montejano, 2013) typical of the Yucatecan culture. Finally, in a study carried out in Merida, Yucatan, it was found that some grandmothers had a negative impact on their grandchildren's feeding patterns, by suggesting the consumption of excessive amounts of food (Hernández-Escalante et al., 2015).

Although there is scarce evidence regarding the tendency for grandmother's support in Mexico, the little evidence available suggests that grandmothers – mainly maternal grandmothers - appear to be the more reliable and wanted helpers in comparison with others potential sources of support. In fact, according to preliminary data from a previous study carried out by the Department of Human Ecology (Cinvestav) in Merida, Yucatan, Mexico, from 2011 to 2015 (Intergenerational effects on growth in inhabitants of Merida, Yucatan; Project number: Conacyt 168047), approximately one-third of a sample of 230 mothers gained support from the maternal grandmother. This evidence, and the previous studies presented, gave me the starting point to consider in more detail the importance of grandmothers in this population.

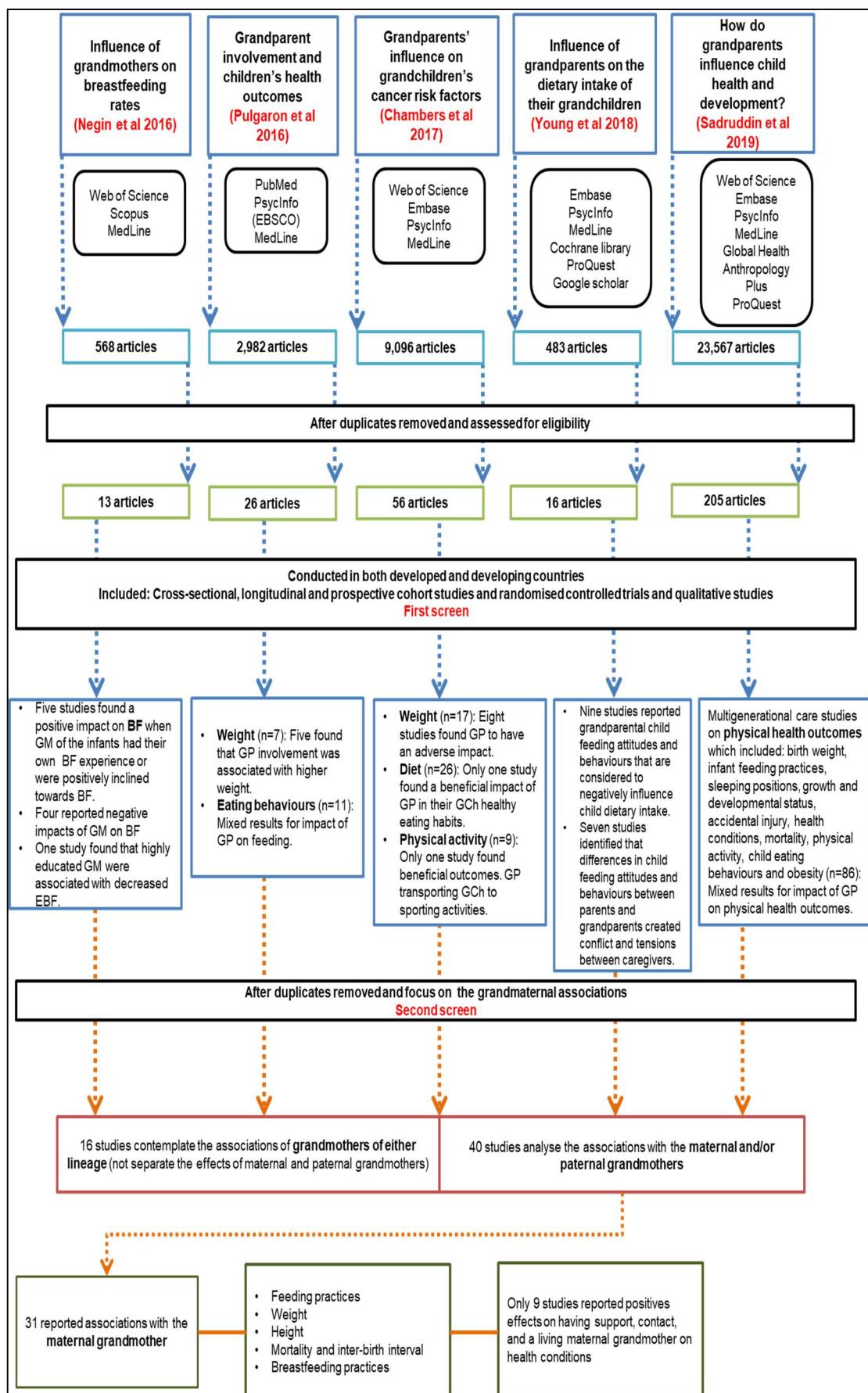
2.6 Studies on maternal grandmother effects

During my review, I was able to identify five systematic reviews focused on the links between grandparents and grandchildren's health (Negin et al., 2016; Pulgaron et al., 2016; Chambers et al., 2017; Young et al., 2018; Sadruddin et al., 2019). These reviews included different variables such as breastfeeding duration, child health outcomes, grandchildren's cancer risk factors and children's health and development. However, for many of the studies included in these reviews, grandparent involvement in children's health was a secondary outcome or was one of many potential predictors, which could be one of the reasons for the mixed results of the impact of grandparents (**Figure 2.7**).

However, in spite of the limitations, the studies helped me identify the areas in which grandparents might be a factor related to their grandchildren's health outcomes, such as growth, feeding practices and physical activity (**Figure 2.7**). Due to there being good evidence that maternal grandmothers have the most frequent and consistent effects on child survival and health (Sear and Mace, 2008), whereas there is less evidence of the effects of other grandparents, I focused on those studies that included the involvement of the maternal grandmother as a secondary outcome or potential predictor.

I first screened all the studies included in the systematics reviews (n=316) and focused on: a) studies in which grandparents and parents were caregivers and b) studies of physical health outcomes (birth weight, infant feeding practices, growth and developmental status, health conditions, mortality, physical activity, child eating behaviours and obesity). Once I identified the appropriate studies (n=185), I undertook a second screening, focusing on studies that reported grandmaternal effects. Based on this last criterion, I identified 56 studies of which 16 addressed the effects of grandmothers without differentiating the lineage and 40 considered associations differentiating between the maternal and paternal grandmother (**Figure 2.7**). From these 40 studies, 31 reported associations for the maternal grandmothers on different topics such as feeding practices, mortality rates and nutritional conditions (**Figure 2.7**).

Figure 2.7 Flow diagram of data review



BF: Breastfeeding; EBF: Exclusive breastfeeding; GP: Grandparents; GCh: Grandchildren

I identified mixed results in these studies. Nine studies reported positive effects of having support from, or contact with, a living maternal grandmother on health conditions (Mahoney and James, 2000; Jamison et al., 2002; Sear et al., 2002; Heath, 2003; Ragsdale, 2004; Gibson and Mace, 2005; Cunningham et al., 2010; Sheppard and Sear, 2016; Cisco, 2017), eight studies reported that grandmother's perceptions and knowledge related to feeding practices could influence, positively or negatively, the family's food choices and the grandchild's diet (Black et al., 2001; Johnson et al., 2010; Nunes et al., 2011; Dashti et al., 2014; Odom et al., 2014; Giugliani et al., 2019; Doung et al 2004; Eli et al 2015), ten studies found negative effects of having contact and/or a living maternal grandmother on health or feeding practices (Susin et al., 2005; Giugliani et al., 2008; Kohlhuber et al., 2008; Sear, 2008; Tanskanen, 2013; Bernie, 2014; Emmott and Mace, 2015; Speirs et al et al 2009; Liu et al 2013; Eli et al 2017) and four studies reported no effects on health outcomes (Kemkes-Grottenthaler, 2005; Modin and Fritzell, 2009; Soldateli et al., 2016; Perry, 2017).

The mixed results could be due to the diverse ways in which grandmothers are involved in the lives of their grandchildren, and variation in how this involvement was operationalized. Moreover, the lack of, or limited, attention to contextual variables that might influence the grandmother effect might also contribute to the heterogeneity of the results (Sadruddin et al., 2019).

In **Table 2.1** I present a summary of the 56 studies that examined grandmaternal effects on different children outcomes.

From the eight cases that addressed the effects of grandmothers on infant survival, 88% (n=7) analysed the effects of maternal grandmothers and 75% (n=6), the paternal effects. Maternal grandmothers were associated with improved child survival in 71% of cases (5 of 7 studies). Paternal grandmother effects were inconsistent and only in 17% of the cases (1 of 6 studies) was a positive impact on child survival reported.

Regarding children's nutritional status, 60% of cases (6 of 10 studies) separated maternal from paternal effects. Thirty three-per cent of the cases (2 of 6) reported a beneficial effect of maternal grandmothers on child height and 17% (1 of 10) a positive effect on birth-weight. Two cases (33%) did not find any association and one case (17%) reported that maternal grandmothers increased the risk of child overweight. Only three studies reported paternal effects, which were consistently negative.

Paternal grandmothers were associated with an increased risk of overweight and undernutrition.

Seventeen studies addressed the effects of grandmothers on feeding practices and physical activity. Only 41% (9 of 17 studies) of the cases separated maternal from paternal effects. Of the seven cases that analysed maternal effects, four studies reported negative influence of the grandmother on infant feeding, one study did not find an association and two studies pointed out the influence of grandmothers on food choices and that interventions that included the grandmothers could improve infant feeding practices.

Finally, concerning breastfeeding practices, of the 22 studies identified, 73% separated the effects of maternal and paternal grandmothers. Overall, more cases (11 of 16 studies) described the influence of maternal grandmothers on breastfeeding patterns. Their attitudes and preferences towards breastfeeding and their perceptions about this practice were associated (positively or negatively) with breastfeeding patterns. One study specifically reported that non-daily contact with the maternal GM was a protective factor for maintaining breastfeeding and another study pointed out that exclusive breastfeeding was less likely in the presence of higher maternal GM education. Studies that addressed paternal grandmothers' influence on breastfeeding were fewer (n=7), but 57% (4 of 7) reported a protective effect of the paternal grandmother on exclusive breastfeeding.

Table 2.1 Literature review

Outcome: Mortality

Author(s)	Year	Country	Design	Sample	Outcome	Impact of the GM
Jamison et al	2002	Japan	CS	2,381	Children mortality	The only grandparent whose presence exerted a consistently negative effect on the likelihood of a child's death was the maternal GM
Sear et al	2002	Gambia	L	2,294	Mortality	Having a living mother, maternal GM , or elder sister had a significant positive effect on the survival probabilities of children, whereas having a living father, paternal GM , grandfather, or elder brother had no effect
Heath	2003	USA	CS	283	Infant mortality	Presence of maternal GM , aunts, uncles, and paternal aunts were significantly associated with increased infant survivorship while grandfathers, paternal GM , and paternal uncles showed little effect
Gibson and Mace	2005	Ethiopia	CS	700	Mortality and growth	In general, grandparents and especially maternal GM exerted a positive influence on child survival.
Kemkes-Grottenthaler	2005	Germany	L	---	Infant mortality	The survival odds of infants were greatly improved by the existence of a paternal GM , while the paternal grandfather had a negative impact. Maternal grandparents appeared to be of no consequence
Sear	2008	Malawi	CS	509	Mortality	Weak evidence that paternal GM may be beneficial to a child's survival chances Maternal GM had a significant impact on child mortality, appearing to increase mortality rate
Ragsdale	2014	UK	CS	47	Mortality and inter-birth interval	Maternal GM survival was found to influence child survival both via maternal survival and independent of maternal survival
Dong et al	2017	China, Japan and Taiwan	CS	69,125	Children mortality	Effects of co-resident kin (such as the paternal GM) for child survival were null or inconsistent between populations

Table 2.1 Literature review

Outcome: Nutritional conditions

Author(s)	Year	Country	Design	Sample	Outcome	Impact of the GM
Sowan and Stember	2000	USA	L	630	Influence of the parental characteristics on the development of infant obesity	Grandmother cohabiting with the child was not a significant predictor of infant obesity
Gibson and Mace	2005	Ethiopia	CS	700	Mortality and growth	Maternal GM had a particularly beneficial effect on child height, but paternal GM less so
Sharna and Kanani	2006	India	C	70	Child caregiving and child's nutritional status among families where GM were present and where GM were absent	GM help enabled mother to practice more caregiving behaviours. However, children in both groups had low calorie intake and a high prevalence of undernutrition
Modin and Fritzell	2009	Sweden	CS	7,719	BMI	On the maternal side: the mother's and the grandfather's, but not the maternal GM , earnings were predictive of cognitive ability, whereas only the grandfather's earnings seemed to be important for BMI
Cunningham et al	2010	South Africa	E	9,0000	Importance of the mother's access to the father and grandparents of the child during pregnancy	Women whose mothers (maternal GM) were alive had heavier new-borns, but no additional benefit was observed from residing together. Co-residence with any grandparent was not associated with birth weight after controlling for the mother's partnership
Moschonis et al	2010	Athens	CS	729	Prevalence of overweight and obesity in children	The GM as the child's primary caregiver was significantly associated with childhood overweight and obesity
Tanskanen	2013	UK	C	9,000	Association between maternal and paternal GM childcare and early years overweight in the UK	Maternal and paternal GM seemed to increase the risk of child overweight equally
Shepard and Sear	2016	Guatemala	CS	6,262	Children height-for-age	Having a living paternal GM tended to be negatively associated with child height. By contrast, contact with maternal kin appeared broadly to be beneficial for child height, although these relationships are weaker
Perry	2017	Bangladesh	E	86	Height, weight and education	Multivariate analyses found that patrilocal residence , net of income effects, was associated with children having shorter stature and lower weight. Matrilocal residence was associated with better child educational attainment, net of income, as was having the maternal GM present in the home and/or serving as primary alloparental caregiver.
Schrijner and Smits	2018	31 sub-Saharan Africa countries	CS	344,748	Stunting	Children in households with a co-residing GM had significantly lower odds of being stunted, if the GM was in the 50–75 age range. When the GM was very young or very old, the likelihood of being stunted is higher

Table 2.1 Literature review

Outcome: Feeding practices and physical activity

Author(s)	Year	Country	Design	Sample	Outcome	Impact of the GM
Black et al	2001	USA	RCT	181	Infant feeding practices	The intervention improved the communication and negotiation between mother and maternal GM regarding infant feeding
Bezner Kerr	2008	Malawi	Q	---	GM role and views of child feeding practices and their influence on women's practices	Paternal GM had a powerful and multifaceted role within the extended family in terms of childcare. They often differed in their ideas about early child feeding from conventional Western medicine
Giugliani et al	2008	Brazil	L	220	Infant feeding practices	Introduction of water/herbal teas was associated with cohabitation with maternal GM
Lindsay et al	2009	USA	Q	31	Understand how social class, culture, and environment influence feeding practices	Adverse effect of GM on eating behaviours
Speirs et al	2009	USA	CS	62	Impact of grandmothers on their grandchildren's fruit and vegetable consumption	Maternal GM shaped their grandchildren's fruit and vegetable consumption by purchasing and providing food. Although GM reported liking fruits and vegetables they did not consume the daily recommendations, then the results suggest that GM could exert a negative influence over their grandchildren's eating habits due to this suboptimal consumption may transfer to their grandchildren
Titiloye and Brieger	2009	Nigeria	CS	486	Infant feeding practices	Mothers and GM employed different methods of feeding children, with mothers more likely to use a bottle and GM more likely to hand- or force-feed children
Johnson et al	2010	USA	Q	7	Matrilineal influence on family food choice	Matrilineal influence (including maternal GM) had an influence on family food choices. This finding may help to understand why sometimes it is difficult to change mothers' food habits.
Farrow	2014	UK	CS	100	Feeding practices	Grandparents (most of them GM) reported using negative feeding practices such as using food to regulate emotions and restricting food, but more positive practices such as providing a healthy food environment
Eli et al	2015	USA	Q	49 (27 GM)	Feeding practices	Mothers perceived that maternal GM provide high sugar products whereas GM perceived they provide a balance diet

Table 2.1 Literature review

Outcome: Feeding practices and physical activity

Author(s)	Year	Country	Design	Sample	Outcome	Impact of the GM
Kavle et al	2015	Egypt	Q	150 (40 GM)	Diet	Mothers were often persuaded by GM to give prelacteal liquids after birth. GM provided children with non-recommended food (cakes/biscuits)
Kobayashi et al	2015	Japan	CS	3,832	Dietary intakes	Young Japanese women and their mothers living with GM had significantly higher food and nutrients intakes than those living without GM
Mukuria et al	2016	Kenya	CS	509	Infant feeding practices	Engaging fathers and GM to improve their knowledge of optimal feeding practices and to encourage provision of social support to mothers could help improve some feeding practices
Soldateli et al	2016	Brazil	L	323	Infant feeding practices and eating behaviours	Cohabitation with the maternal GM had no effect on the quality of children's diet
Eli et al	2017	USA	Q	22 (11 GM)	Feeding practices (beverage consumption)	Some maternal GM spoiled their grandchildren with high sugar drinks
Karmacharya et al	2017	Nepal	CS	1,399	Infant and young child feeding practices	Among households with GM in residence, the odds of appropriate feeding practices were at least two times more likely in households where GM had correct knowledge compared with households where GM had incorrect knowledge
Xie et al	2018	USA	Q	53	Children's leisure time physical activity	Grandparents (mostly GM) had a very positive perception of physical activities for children and supported grandchildren's physical activities, directly (e.g., doing physical activities with grandchildren) and indirectly (e.g., making suggestions to parents)
Yue et al	2018	China	CS	1,382	Infant feeding practices	Grandmothers (paternal GM) engaged in poorer feeding practices than do mothers

Table 2.1 Literature review (Continuation)

Outcome: Breastfeeding

Author(s)	Year	Country	Design	Sample	Outcome	Impact of the GM
Li et al	1999	Thailand	CS	221	Breastfeeding practices (exclusive and partial) at 3 months postpartum	Increased the risk of mixed or formula feeding if GM (and others) was the main child caretaker
Mahoney and James	2000	USA	CS	66	Anticipated infant feeding practices (exclusive and partial) from 4 to 52 weeks after delivery	Direct association for encouragement from the woman's mother (maternal GM) to breastfeed and anticipated breastfeeding
Ingram et al	2003	UK	Q	30	Breastfeeding	The GM was generally enthusiastic about breastfeeding, so the antenatal intervention was designed to enhance GM knowledge and enable them to give accurate advice
Ludvigsson	2003	Bolivia	CS	502	Duration of exclusive breastfeeding and use of prelacteal food and/or colostrum	The attitudes of the maternal GM towards breastfeeding did not influence infant feeding patterns
Duong et al	2004	Vietnam	CS	463	Exclusive breastfeeding within the first week after delivery	Maternal GM's preference of breastfeeding could influence the initiation rate and breastfeeding patterns
Duong et al	2005	Vietnam	CS	463	Exclusive breastfeeding within the first week and 4 and 6 months postpartum	Mother-in-law (paternal GM) preference for breastfeeding was associated with feeding practices
Susin et al	2005	Brazil	PCS	601	Exclusive breastfeeding at 6 months after delivery	Abandonment of breastfeeding within the first six months was associated with maternal and paternal GM giving advice about other drinks. Non-daily contact with the maternal GM was a protective factor for maintaining breastfeeding until six months
Masvie	2006	India	Q	31	Grandmothers' perspective on breastfeeding	Paternal GM held colostrum in high regard, used no prelacteal and supported early initiation of breast feeding

Table 2.1 Literature review (Continuation)

Outcome: Breastfeeding

Author(s)	Year	Country	Design	Sample	Outcome	Impact of the GM
Santo et al	2007	Brazil	PCS	220	Exclusive breastfeeding at 6 months after delivery	A protective effect was present on exclusive breastfeeding when the mother cohabited with her mother-in-law (paternal GM)
Kohlhuber et al	2008	Germany	PCS	3,822	Breastfeeding practices at 2, 4 and 6 months postpartum	No initial breastfeeding was associated with maternal GM's negative attitude towards breastfeeding
Chen et al	2011	Taiwan	CS	210	Breastfeeding practices (exclusive, partial and non-breastfeeding) at 3 months postpartum	Breastfeeding among mothers-in-law (paternal GM) was positively associated with breastfeeding at 3 months postpartum
Nunes et al	2011	Brazil	RCT	323	Exclusive breastfeeding at 6 months after delivery	Counselling sessions on infant feeding in the first 4 months postpartum for mothers and maternal GM , when they cohabit, was an effective strategy to postpone the unnecessary introduction of other drinks on breast-fed infants
Liu et al	2013	China	CS	1,385	Breastfeeding practices (exclusive and partial) at 6 months postpartum	Exclusive breastfeeding was less likely in the presence of higher maternal GM education
Wasser et al	2013	USA	PCS	209	Non-maternal involvement in feeding during the first 2 years of life and	Non- maternal caregivers (including the GM) involvement was associated with lower likelihood of continued breastfeeding and higher likelihood of children consuming juice or whole fruit
Bernie	2014	UK	CS	9	Infant feeding practices. Breastfeeding.	Maternal GM was identified as important influences on some women, and, in particular, concerns were raised that GM sometimes undermined intentions to breastfeed by offering to bottle feed infants
Dashti et al	2014	Kuwait	CS	373	Breastfeeding practices (exclusive and partial) at 6 months postpartum	Maternal GM preference of breastfeeding was associated with breastfeeding practices
Odom et al	2014	USA	L	2,041	Breastfeeding initiation	Never breastfeeding was significantly associated with the following perceptions: maternal GM preferred only formula feeding and maternal GM had no opinion/didn't know their feeding preference

Table 2.1 Literature review (Continuation)

Outcome: Breastfeeding

Author(s)	Year	Country	Design	Sample	Outcome	Impact of the GM
Emmott and Mace	2015	UK	CS	18,827	Feeding practices	Frequent contact with maternal and paternal GM was associated with a lower likelihood of breastfeeding initiation and a higher risk of breastfeeding termination
Susiloretni et al	2015	Indonesia	DR	163	Determinants of exclusive breastfeeding	One factor which shortened breastfeeding duration was GM lack of support for EBF
Bai et al	2016	Hong Kong	L	1,172	Infant feeding practices	No direct relationship between the GM infant feeding preferences and breastfeeding cessation, co-residence with a mother-in-law was significantly associated with early cessation
Cisco	2017	USA	CS	594	Breastfeeding support	Mothers receive significant support, particularly from spouse and maternal GM
Giugliani et al	2019	Brazil	RCT	323	Infant feeding practices: Promote exclusive breastfeeding and reduce pacifier use in the first 6 months of life	The intervention had a significant impact on reducing pacifier use only in the group in which maternal GM were involved

This review allowed me to identify the areas in which maternal grandmothers can play an important role and to consider the methodological proposals that were necessary to include in the design of the present study, such as a clear operationalization of grandmothers' involvement. Moreover, this review drew my attention to the lack of studies carried out in Latin America, especially in Mexico, considering that there is evidence about grandmothers - mainly maternal grandmothers – being the preferred substitute for childcare (Partidas, 2004) and as the family members that have most contact and a closer relationship with grandchildren (León et al., 2016).

Moreover, I was not able to identify studies that addressed the effects of the maternal grandmother on their daughter's health conditions. It is clear that grandmothers can enhance mothers' fitness through improving children's survival and, as part of the maternal social network, have a central role as an advisor to mothers in topics such as breastfeeding and food practices. However, none of the studies considered if the presence and/or support of the grandmother could influence the mother's health conditions that, in turn, could be associated with child growth patterns.

There is evidence about the grandmother's contribution to keeping children alive and about their potential beneficial contribution to children's growth. However, are the grandmothers helping to keep children healthier? Moreover, are they contributing to their daughter's well-being?

To respond to these questions, I wanted to explore a wider range of nutritional and metabolic traits in both mothers and their children. Additionally, in contrast to most studies of grandmother support, I also wanted to take into account other social relationships, to address more comprehensively the mother's social network.

2.7 Potential grandmaternal benefits on maternal and infants health

As I stated in previous sections of this chapter, the literature has highlighted that the strategy of accumulating and using energy capital is sensitive to social signals (Wells, 2012) and for females, the effective use of social capital can benefit both mothers and their offspring. In this sense, according to my review, grandmaternal investment can potentially favour the accumulation of capital, with beneficial effects on life history traits.

In Table 2.2, I present, according to the results reported by previous studies, how maternal grandmother investment could be one important factor in driving life-history-trade-offs.

Table 2.2 Potential life-history trade-offs relative to grandmaternal investment

Population	Exposure	Outcome	Maternal GM investment on
Gambia Sear et al 2002	Presence of maternal GM	Higher infant/child survival	Maintenance and Immune function
Japan Jamison et al 2002			
USA Heath 2003			
Ethiopia Gibson and Mace 2005			
Malawi Sear 2008			
UK Ragsdale 2014			
Ethiopia Gibson and Mace 2005	Presence of maternal GM	Beneficial effects on child height	Growth
Guatemala Shepard and Sear 2016		Beneficial effects on birth weight	
South Africa Cunningham et al 2010			
Nepal Karmacharya et al 2017	Presence of maternal GM	Appropriate feeding practices	Growth, maintenance and immune function
USA Mahoney and James 2000	Presence of maternal GM	Increase exclusive breastfeeding practices	Growth and immune function
Brazil Nunes et al 2011			
Kuwait Dashti 2014			

For children, previous studies have highlighted the potential benefits of the presence of the maternal grandmother on their survival and growth. For mothers, it seems that this has been less explored and, according to my review, only one study reported findings of grandmaternal benefits on their daughter's health and how this association might mediate the beneficial effects of grandmothers on their grandchildren's growth. In their analysis using data from the Guatemalan Survey of Family Health, Shepard and Sear (2016) found that the slightly higher maternal BMI associated with contact with maternal grandmothers could probably explain the small beneficial effect of grandmothers on child height, suggesting grandparental influence acts partly through maternal health.

Interestingly, a study by Karmacharya et al (2017) in Nepal, reported that grandmothers' knowledge about child feeding practices operates via maternal knowledge, showing that knowledge resources provided by the grandmother can enhance the well-being of their grandchildren via maternal behaviour.

Therefore, considering that grandmaternal investment can have beneficial effects on life-history traits, and that hormones shapes life history strategies, could grandmaternal support drive hormonal changes that underlie health impacts?

2.8 Social support and stress

In humans, becoming a mother for the first time has been reported as a stressful experience due to women having to learn new maternal skills during this period (Hudson et al., 2001; Nelson, 2003), and infant caretaking tasks, such as feeding, sleeping, changing nappies (Kapp, 1998), and baby bathing (Beger and Loveland Cook, 1998). At the same time, women experience worries about infant well-being (Bennett, 1981) and also have to learn how to care for themselves. These challenges could represent an energetic burden to the mother because they could perceive themselves as having insufficient resources to handle the demands of motherhood and hence, this could negatively influence their capacity to respond to their children's needs.

Stressors, such as motherhood and family environment, income inequalities and work conditions and demands, have a major influence on health and its effects can vary according to the individual's vulnerability and resources. Some of the protective factors that have been identified are social support, self-esteem and optimism

(Schneiderman et al., 2005). In the literature, it has been suggested that social support can help to relieve the negative effects of stress and that the family can be the most important source of support promoting the skills needed to overcome adversity (Seccombe, 2000).

Although it is not clear how positive social interactions suppress the stress-responsive physiological systems or stimulate other internal regulatory systems involved in the attenuation of stress reactivity, it seems that one mechanism is through the suppression of cortisol concentrations during stress exposure (Heinrichs et al., 2003; Clodi et al., 2008; Ditzen et al., 2009).

The main biological system responsible for the stress response is the hypothalamic-pituitary-adrenal (HPA) axis, including both neuronal and endocrine structures (Martínez-Sanchís et al., 2007). Cortisol is a steroid hormone released from the adrenal glands that regulates several metabolic functions such as the metabolism of macronutrients - for instance, increasing blood glucose concentrations and synthesizing glucose from non-carbohydrate sources (amino and fatty acids) (Khani and Tayek, 2001). Moreover, it has several direct effects on brain functions and enhances the ability to adapt to stress (McEwen et al., 1997). Hence, glucocorticoids, like cortisol, are commonly used as biomarkers of stress responses, and their role in stress processes in both animals and humans is well accepted (Thurin and Baumann, 2003).

The stress response comprises a complex interaction between different mechanisms that are activated for a better adaptation to the environment, facing or avoiding unfavourable situations. However, chronic stress, in which chronically elevated cortisol together with other physiological adjustments are present, has been shown to have detrimental effects on numerous physiological, emotional and behavioural functions (Starkman et al., 1981; McEwen, 1998; Belanoff et al., 2001; Chu et al., 2001; Burke et al., 2005; Ventura et al., 2012). Physiological effects include resistance to the effects of insulin, promotion of obesity and osteoporosis, altered immune responses and increased blood pressure (Seeman et al., 1997; Lupien et al., 1998; McEwen, 1998; Dennison et al., 1999; Rosmond and Björntorp, 2000). Endogenously elevated cortisol has also been associated with alterations in emotion regulation (Starkman et al., 1981), psychotic symptoms (Chu et al 2001), clinical depression (Belanoff et al., 2001; Burke et al., 2005) and anxiety (Ventura et al., 2012). Moreover, cortisol has been implicated as a potential mediator of increased energy intake in

humans (Tataranni et al., 1996; Koo-Loeb et al., 2000), and in promoting the visceral accumulation of body fat (Björntorp, 1997; Fraser et al., 1999; Rosmond and Björntorp, 2000; Gluck et al., 2004; Wells, 2010b) by expressing lipoprotein-lipase (Björntorp, 1997). Therefore, the most relevant hormone here is cortisol, which shapes strategies in response to various types of stress and, at the same time, has a close relationship with high adiposity and inadequate behaviours, with negative implications in health conditions.

According to my review, it has been established that intimate and regular contact with relatives, neighbours, and friends enhances competence as a parent (Pridham and Chang, 1992; Mercer and Ferketich, 1994), and has a beneficial influence on the stress response (Turner et al., 1999; Light et al., 2000, 2005; Ditzen et al., 2009). Hence, it is possible to consider that the assistance and presence of maternal grandmothers might potentially suppress cortisol concentrations in their daughters during exposure to stresses related to motherhood and other factors, such as the work and family environment, income inequalities and working demands.

Moreover, from the studies reviewed, it is clear that grandmother's investment could be through direct childcare and knowledge provision; however, it is important to consider that they can also transmit confidence and encouragement to their daughters, which can be beneficial for maternal stress relief. For instance, mothers from north Texas who participated in focus groups in which the topic was grandmother breastfeeding support, pointed out that they do not expect grandmothers to be experts on breastfeeding practices, rather they want loving encouragement from them when they experience difficulties (Grassley and Eschiti, 2008).

Considering that social capital can potentially suppress cortisol concentrations and that cortisol has been associated with emotion regulation, maintaining secure social relationships and the resulting effects on stress responsiveness could be a central factor in shaping maternal behaviour, with physiological and behavioural effects on children. For instance, a cross-sectional analysis revealed that children's cortisol levels at 4.5 years of age were higher among those exposed to concurrent maternal stress than among children exposed to moderate or lower levels of stress (Essex et al., 2002). In another study, Ashman et al. (2002) found that children with elevated levels of internalizing symptoms (e.g. overanxious and depressive disorders) whose mothers had a history of depression, showed elevated baseline cortisol levels.

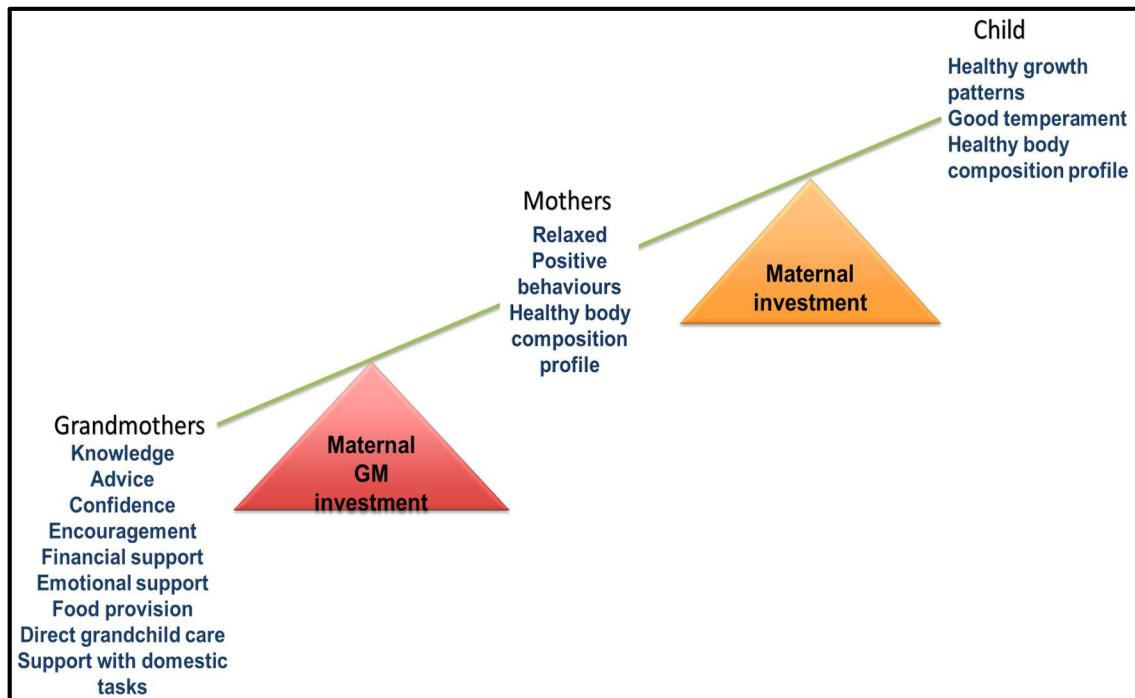
Evidence also has indicated that children whose mothers experienced more months of depression in the first two years of their lives exhibited higher baseline salivary cortisol levels at age 7 years, which suggests that younger children might be more prone to experience cortisol elevations in response to maternal anxiety (Ponzi et al., 2016).

Moreover, together, elevated cortisol concentrations and negative behaviours could lead to increased energy intake (Tataranni et al., 1996; Koo-Loeb et al., 2000) promoting the visceral accumulation of body fat (Björntorp, 1997; Fraser et al., 1999; Rosmond and Björntorp, 2000; Gluck et al., 2004; Wells, 2010b). Adipose tissue functions as a key endocrine organ and it has been stated that its accumulation in and around the abdomen is a key cause of metabolic abnormalities (Matsuzawa, 2006) associated with several diseases that share inflammation as a common link, such as hypertension, cardiovascular disease, type 2 diabetes, some types of cancer and the metabolic syndrome (Florez et al., 2006). Regarding behaviours related to increase energy intake, infants and younger children do not have complete autonomy over their own eating behaviours and therefore, their habits are influenced by maternal decisions. Having a mother that experiences daily stressors could lead to poor dietary habits in children that could continue into adulthood and increase the risk of chronic diseases.

Therefore, having grandmaternal support could have important effects on both behaviours and body composition through the mediation of stress responses (**Figure 2.8**). Through this mechanism, grandmaternal support could potentially reduce chronic disease risk in both, mothers and grandchildren.

Based on this review, I designed my study through an evolutionary lens to explore social capital effects on maternal and children health outcomes, taking into account one of the hormones known to mediate life history strategy.

Figure 2.8 Potential benefits of grandmaternal investment on mothers and infants health conditions



Grandmaternal investment can enhance their daughters' response to stress and improve behaviours, with a beneficial effect on their body composition. In turn, this healthy maternal profile could have a positive impact on their children's growth and temperament.

CHAPTER 3

METHODS AND PROCEDURES

3.1 Introduction

This chapter aims to describe in detail the methods used during data collection, data handling and data analysis. The chapter provides a section about the ethical considerations of the research and information about the pilot studies applied before the data collection began. A detailed description of the interview applied to the women is also contained in this chapter, as well as the description of the materials used to collect biological samples (saliva and urine). Following that, I describe the feedback to participants and data handling and analysis.

3.2 Ethical considerations

Ethical clearance was obtained from the University College London Ethics Committee in the UK on October 17, 2016, and by the Comité de Bioética para el Estudio con Seres Humanos (Bioethics Committee for the Study of Human Beings) in Mexico on May 03, 2017. Additionally, I obtained authorization from the Ministry of Education, the Ministry of Health, and the National System for the Integral Development of the Family (DIF), the Institute of Security and Social Services of State Workers (ISSSTE) and the Institute of Social Security of Workers of the State of Yucatan (ISSTEY) and the Ministry of Social Development (Sedesol) to recruit participants in the centres of child care.

Recruitment is described in section 3.4 of this chapter and was targeted at the mothers. It was clearly explained to the women that their participation was strictly voluntary and that they could withdraw from the study at any time. Participant information sheets were given to both the mothers and the grandmothers and the study was fully explained orally by the local research team. Moreover, I explained to the mothers that the biological samples collected would be stored in Cinvestav until their analysis and that all the information provided would be strictly confidential and would be used for research purposes only. Mothers and grandmothers were asked to discuss with their husbands their decision to participate in the study. Any question about the study was answered and all women signed consent forms after the study was explained, completely understood, and they had agreed to participate.

For the anthropometric measurements, all participants were asked to wear shorts and sleeveless shirts. They were asked to pull up their shirts for some measurements, such as waist circumference or skinfolds. No participant had an issue with this. All measurements were made, ensuring the privacy of the participants, by a female researcher with a female recorder.

Finally, during the interview, only the participant and the interviewer were present at the time of the interview application. All the women were informed that they could refuse to answer any question if they felt uncomfortable. Mothers and grandmothers were interviewed separately. However, in some cases grandmothers helped the mothers to answer some questions related to their childhood.

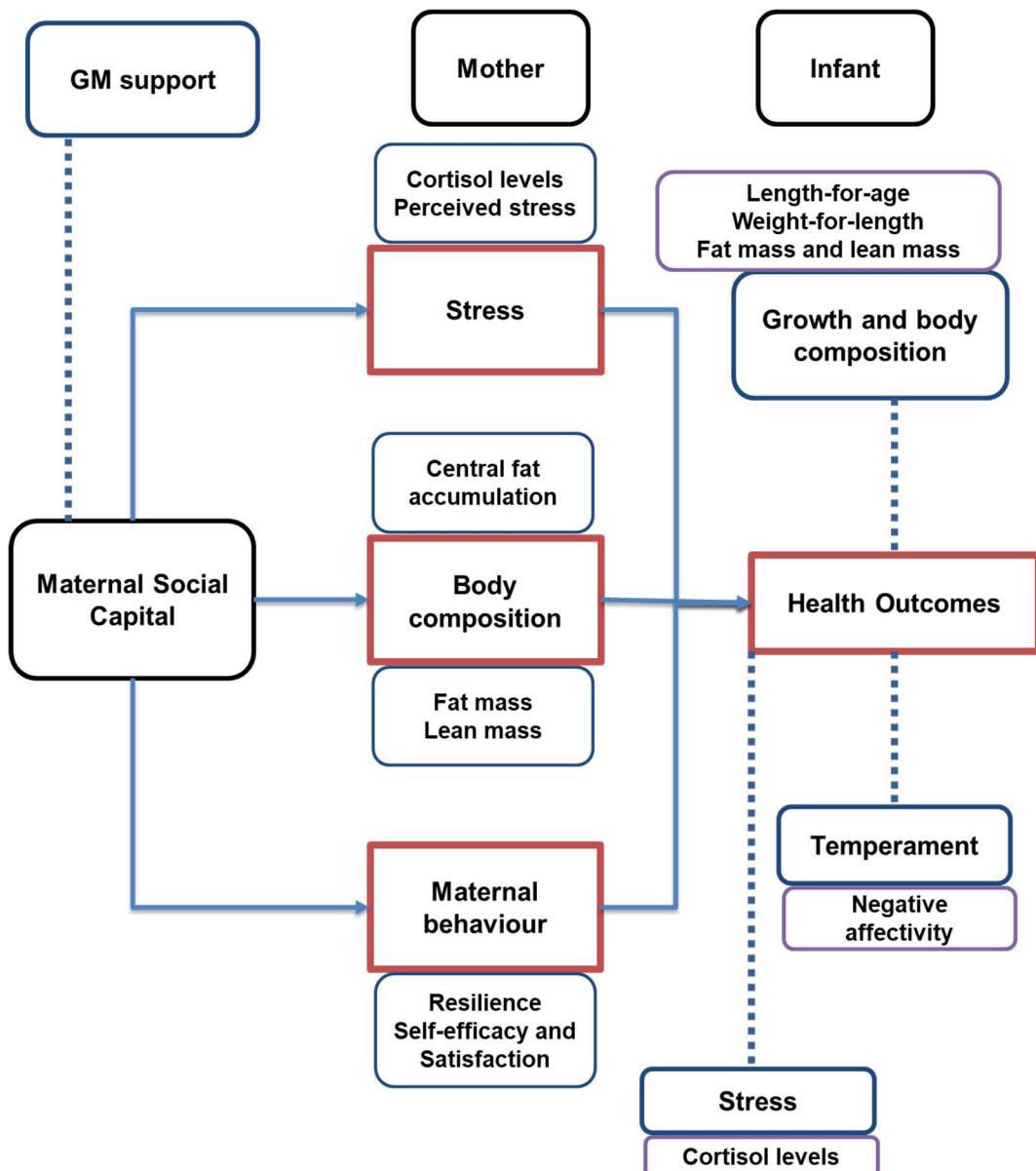
3.3 Study design

This was a cross-sectional study carried out in Merida, Yucatan, Mexico with the support of the Somatology Laboratory of the Human Ecology Department of the Centre of Research and Advanced Studies (Cinvestav-Merida) in charge of Dr. Federico Dickinson (FD). The pilot and training phases ran between January and May 2017, and the data were collected from June 2017 to July 2018.

The local field team included FD, Adriana Vázquez (AV), Graciela Valentín (GV), Hidekel Chan (HC), Carolina Batún (CB), Diletta Cipriani (DC) and Brenda Hernández (BH). FD and I (AV) led the investigation, while HC, CB, and GV were the primary members of the field team. DC and BH were in the field for two months and assisted with recruitment and information sessions.

The aim of this research was provide a better understanding of the association between maternal social capital and children's health by obtaining data related to the supportive social relationships of the mother, and in particular about the support of the maternal grandmother. I tested the over-arching hypothesis that women with grandmothers' support would have more favourable physiological and psychological characteristics that, in turn, would be associated with healthier growth patterns and lower levels of irritability in their children ([Figure 3.1](#)).

Figure 3.1 Cross-sectional study design



To test this hypothesis I recruited a cohort of 90 mother-infant dyads (52 with grandmothers' support, 38 without) in Merida, Mexico, in order to compare markers of maternal nutritional status and stress, and child's growth, body composition and behaviour between these two groups. Children were first-borns and aged 1.7-2.3 years.

3.4 Research questions and hypothesis

To meet the aim of the study, I developed primary and secondary research questions and hypotheses.

The primary research question was:

What is the relationship between maternal grandmother's help and children's growth? Evolutionary anthropologists have now produced several reviews and studies which show that the presence and help of grandparents are positively correlated with children's survival (Mace and Sear, 2005; Sear and Mace, 2008). One study among Aka foragers has extended these findings to children's growth (Meehan et al., 2014). However, it is clear that context matters for grandparental investment and behaviour (Sear, 2016). Broadly, maternal grandmothers appear to invest more and to be more beneficial to children compared to paternal grandmothers (Euler and Michalski, 2007; Sear and Mace, 2008; Sear and Coall, 2011), though not all studies agree (Pashos, 2000; King et al., 2003; Euler and Michalski, 2007). In Yucatan, it seems that maternal grandmothers are the preferred substitute for childcare and are a source of knowledge and support for their daughters (Rodríguez-Angulo et al., 2012; Reyes-Gutierrez and Cervera-Montejano, 2013; Hernández-Escalante et al., 2015; Gutiérrez-Carbajal et al., 2019). Hence, I expected that grandparents, principally maternal grandmothers, are still an important source of help. My primary hypothesis was therefore that children whose mothers receive support from the grandmother would have healthiest profiles of linear growth (i.e. greater height) and body composition (i.e. higher lean mass).

Secondary research questions were:

1. What is the relationship between the grandmother's support and maternal stress and body composition?

The presence of social support has been associated with decreased stress responsiveness (Heinrichs et al., 2003; Sims et al., 2006; Brent et al., 2011), through the suppression of cortisol concentrations during stress exposure (Heinrichs et al., 2003; Clodi et al., 2008; Ditzén et al., 2009). At the same time, social support may be associated with lower central fat accumulation by reducing chronic cortisol secretion. Becoming a mother for the first time has been reported as a stressful experience in some societies and handling 2 year-old children could be difficult considering that at this age, children are reaffirming their independence (i.e. what they like to do or not),

so grandmaternal support could be beneficial to the mothers during this new experience. My hypothesis was that women who received help from their mothers would be less stressed, have lower levels of cortisol and lower central fat accumulation.

2. What is the association between the grandmother's help and the mother's behaviour?

Previous studies have shown that individuals who have more maternal contact are better equipped to handle stress and give more optimal parenting to their own infants (Champagne, 2008; Ross and Young, 2009). My hypothesis was that women helped by their mothers would have better psychological skills to cope with the challenges of maternity. I expected that these women would show higher levels of resilience and satisfaction and self-efficacy about motherhood, which are skills constructed inside the family environment, especially through the relationship with their mother.

The association of grandmaternal support with maternal phenotype is important to consider in order to identify potential associations with children's growth during early stages of life and in addition, to identify potential effects on children's and mothers' behaviour. Mothers that have access to social support could have greater capacity to deal with stressful events, i.e. be more resilient, and hence, have the psychological resources (psychological capital) to offer high-quality care to their offspring. The present study aimed to offer insight into potential effects at both physiological and psychological levels.

3. What is the association between the grandmother's help and children's temperament?

Care-taker behaviour can influence infant personality (Crockenberg and Acredolo, 1983), and vice versa (Buss and Plomin, 1975; Rothbart and Derryberry, 1981; Derryberry and Rothbart, 1984). Therefore, the impact of grandmaternal support might potentially be associated with the infant's temperament. I hypothesized that children who received care from their grandmothers would be less stressed and have lower levels of cortisol.

Table 3.1 provides a summary of the study questions and hypotheses, outcomes measures and research tools used in this study.

Table 3.1 Hypotheses, outcome measures and research tools using in the study

No	Hypothesis	Outcome measures	Research tools
I	Primary hypothesis: children whose mothers receive support from the grandmother are taller and have higher lean mass	Infant length and body composition	Anthropometry: infant weight and length scales Ratio-mass spectrometry
II	Secondary hypothesis:		Outcome measures
i	Women who received help from their mothers: a) are less stressed b) have lower levels of cortisol c) have lower fat mass and higher lean mass	Maternal stress scores using a self-administered scale Saliva cortisol levels Body composition	Perceived Stress Scale (PSS) ELISA Kits Isotope-ratio mass spectrometry
ii	Women helped by their mothers: a) show higher levels of resilience b) show higher levels of satisfaction and self-efficacy about motherhood	Maternal resilience scores using a self-administered scale Maternal satisfaction and self-efficacy scores using a self-administered scale	Measurement Scale of Resilience in Mexicans (RESI-M) Parental Evaluation Scale (PES)
iii	Infants who receive care from their grandmothers: a) are less stressed b) have lower levels of cortisol	Infant temperament using a self-administered questionnaire filled by the mother Saliva cortisol levels	Early Childhood Behavioural Questionnaire (ECBQ) ELISA Kits

3.5 Location of research: Merida, Yucatan, Mexico

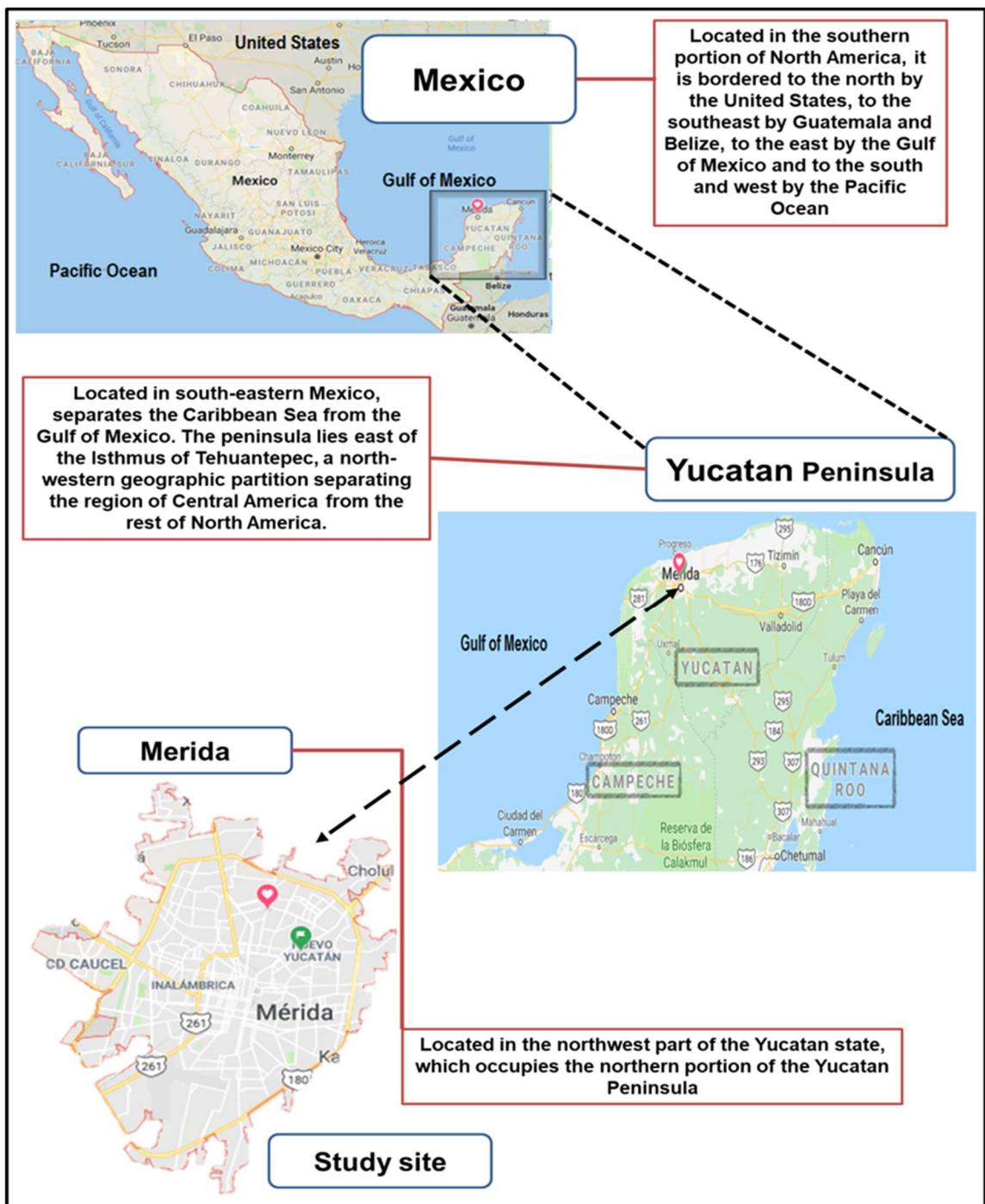
Merida is the capital and the largest city of the Yucatan Peninsula occupying a total area of 883 km². It is located in the northwest part of the state about 35 km from the Mexican Gulf (**Figure 3.2**). Currently, Merida is one of the most important cities in the region, with an outstanding availability of services, including education and health, and is a major tourist destination. According to the Mexican 2015 Census, Merida is the municipality with the largest proportion of the population in the state with 892,363 inhabitants (42.6% of the state population). The percentage of indigenous people was approximately 48.3% of the total population. Most of them were of Mayan ethnicity (INEGI 2016, Available at: <https://www.inegi.org.mx/>).

Another important characteristic is that Merida is a socioeconomically segregated city. The northern area is occupied by the population with the highest levels of income and, in general, has better infrastructure and services in terms of education and health. In contrast, the south is occupied by the population with the lowest income levels, most of them Maya immigrants from rural villages, and lacks proper services and infrastructure (Fuentes, 2005). In economic terms, the daily average income per workday (8 hours per day) is Mex\$102.68 (4.28 GBP) according to the National Minimum Wages Commission.

Eighty-five percent of the Yucatecan state territory is characterized by a sub-humid warm climate and the remaining 14%, located in the northern part of the state, has a dry or semi-dry climate. The average annual temperature is between 26°C and 28°C, the maximum average temperature is around 36°C (mostly during May) and the minimum average temperature is 16°C (in January). The average state rainfall is 1,100 mm per year and the rainy season is during the summer between June and October (INEGI. Available at: <http://cuentame.inegi.org.mx/monografias/informacion/yuc/territorio/clima.aspx?tema=me&e=31>).

Merida experiences a tropical wet and dry climate (Peel et al., 2007). The climate is hot and its humidity is moderate to high depending on the time of year. The average annual high temperature is 33°C, ranging from 28°C in January to 36°C in May, but temperatures often rise above 38°C during this period. Low temperatures range between 18°C in January to 23°C in May and June.

Figure 3.2 Map of Mexico showing the study location area



Source: Images were obtained from Google Maps

3.5.1 Economy

The Yucatecan economy is currently more diversified than it was for most of the twentieth century. Despite the scarcity of soil in the territory, Yucatan has traditionally had a relatively important activity in crop agriculture since pre-Hispanic time (Varguez Pasos, 1981).

The *milpa*¹, known as a productive ecosystem, has been part of the food subsistence system of the inhabitants of the region (Warman, 1985). The main foods cultivated through this system (corn, beans and squash) are an important part of the diet of the local population.

There are other crops that complement the traditional diet of the Yucatecan population and that are exported internationally. Citrus fruit such as oranges, lemons and grapefruits, are the main activity, especially in the south of the state (Varguez Pasos, 1981).

Livestock is an economic activity rooted in Yucatecan culture and of economic relevance for the population. Pig farming has become a dynamic sector that exports the majority of its production to other regions or abroad. Poultry farming is another activity that contributes to strengthening the Yucatecan agricultural sector (Varguez Pasos, 1981).

Due to the shortage of raw materials, Yucatan is not specialized in the industrial area and it is tourism and commercial activities that contribute most to the economy of the state. Tourism in Yucatan represents an important activity and is one of the most dynamic in the country (INEGI. Available at: <https://www.inegi.org.mx/>).

¹ It is a Mesoamerican agroecosystem whose main productive components are corn, beans and squash. In some regions, chilli is also cultivated in the milpa. "Making milpa" means to carry out the entire production process, from the selection of the land to the harvest.

3.5.2 Culture

1.4 million (65.4%) of Yucatecan inhabitants recognize themselves as indigenous (most of them Maya), regardless of whether or not they speak an indigenous language. The number of women considered indigenous (696,900) is greater than that of men (674,700) (INEGI 2015, data available at: <https://www.inegi.org.mx/>). Mérida has the highest percentage of indigenous population within any large city in Mexico. Forty-eight percent of the population recognize themselves as indigenous and 10.20% speak an indigenous language (INEGI 2015, data available at: <https://www.inegi.org.mx/>).

The Spanish language spoken in Yucatan is identified as different in comparison to the Spanish spoken over Mexico as a whole. It is heavily influenced by the Maya language, which has a lot of "sh" sounds (represented by the letter "X" in the Mayan language). However, even though many people speak the Mayan language, there is much stigma associated with it (Lozano et al., 2014).

Nowadays, the culture can be observed every day in the way that people dress, speak and celebrate traditions (Casares et al., 1998). One example of its traditions is the *Hanal Pixán* (Food of the dead), a Mayan/Catholic 'day of the dead' celebration. It is celebrated on November 1 and 2 (one day for adults, and one for children) and is commemorated through the elaboration of altars (*ofrendas* in Spanish) dedicated to dead relatives. The catholic influence can be seen in the use of crucifixes mingled with skull decorations and food offerings (Casares et al., 1998).

According to the results of the last census, by 2010 80% (1 554 805 inhabitants) of the Yucatecan population reported following the Catholic religion (INEGI 2010, data available at: <https://www.inegi.org.mx/>). The remaining 20% indicated following other religions such as Protestant or Christian.

3.5.3 Education and marital characteristics of the population

By 2010, in Yucatan, the average attainment of education among the population of those aged ≥ 15 years was 8 years. Recently, According to official reports (INEGI 2016, data available at: <https://www.inegi.org.mx/>), the average duration of education among Yucatecan women and men in 2015 was 8.6 and 9 years respectively, which represents an increase over recent years.

Due to an increase in the coverage of basic education to eradicate illiteracy among the population, the illiteracy rate decreased by nearly 5% between 2000 and 2015. In 2000, 12.3% of people in this age group could not read or write whereas in 2015, this indicator was reduced to 7.4%.

In relation to the marital characteristics of the Yucatecan population, by 2015, the population that was married predominated (46.6%), following by single people (33.5%) and couples living together (not married) (9.9%) and finally by people divorced/separated/widowed (9.9%). In Merida, a similar pattern was observed with 43% of the population being married, 35% single, 10.1% living with the partner and 12% divorced/separated/widow.

Although in the Intercensal survey carried out in 2015 there is no information about whether the marriages were religious, during the census of 2010, 60% of the population declared a religious marriage.

3.5.4 Health of the population

In 2016, life expectancy in Yucatan was 73.1 years for men and 78.2 years for women (INEGI. Data available at: <http://www3.inegi.org.mx/sistemas/temas/default.aspx?s=est&c=17484>). Among the 5 main causes of mortality in the state are heart disease, diabetes mellitus, cancer (stomach, liver and prostate), liver disease (mainly due to alcoholism) and cerebrovascular disease.

Within Mexico, Yucatan has some of the highest rates of child undernutrition (stunting) and overweight/obesity (OW/OB). According to the National Survey of Health and Nutrition (Ensanut), in 2012 the prevalence of low height-for-age in children aged 1-4 years from urban and rural communities was 15% and 20%, respectively, and for OW/OB was 15.1% and 12.3%, respectively (Instituto Nacional de Salud Pública, 2012).

Among school-age children (5-11 years), the prevalence of overweight and obesity was 22.4% and 22.7%, respectively. The prevalence of overweight in urban locations was 22.1% and for rural areas was 24.3%. The sum of both nutritional conditions (OW/OB) was higher for boys (46.5%) than for girls (43.8%) (Instituto Nacional de Salud Pública, 2012).

In adolescents (12-19 years), the prevalence of OW/OB was 43.2% and was relatively higher for women (44.1%) than for men (42.7%). The prevalence of OW/OB in this group by location was 42% in urban areas and 52% in rural settings (Instituto Nacional de Salud Pública, 2012).

In adults (≥ 20 years), the prevalence of overweight and obesity in women was 82.0% and in men 78.6%. The prevalence of obesity was higher in women (46.2%) than in men (43.3%), while the prevalence of overweight was similar in both sexes (35.3% for men and 35.8% for women). No analysis was carried out by location (urban/rural) (Instituto Nacional de Salud Pública, 2012).

3.6 Participants

I studied 1.7-to-2.3 years old infants, their mothers and, where relevant for one of my subsamples, the maternal grandmothers. I focused on this child age because it is a critical period in which individuals have a physiological sensitivity to the environment, especially to nutritional effects, and because, in terms of temperament, they can show difficult behaviours that can be associated with their health profile. Those effects may track on into later life. Therefore, it is relevant to study the physical growth and nutritional status at an early age as two key predictors of later health and well-being.

I conducted this study focusing on grandmothers' support as a possible factor that could explain variability in health and nutritional status in their daughters and grandchildren. Knowing from previous research that parents with children under age 5 years may receive more household help than parents with older children (Snopkowski and Sear, 2015), it seems relevant to know if this support could provide health benefits to the mother-infant dyad.

The inclusion criteria included:

- i. Infants between 1.7 and 2.3 years old living in Merida. Additionally, it was required that the infant was the first-born child. First-time mothers could benefit more from the support provided by the grandmothers than women who have experience because of previous children.
- ii. Women living in Merida. It was required that the mother was married or living with the child's biological father. Although partner relationships are not stress-

free (Carels et al., 1998), cooperation between marital partners is particularly relevant for child care assistance (Raley et al., 2012) so levels of maternal investment and stress between single and married women could be different. Moreover, the degree of relatedness influences investment, with biological fathers investing more in their children than step-fathers in partners' children (Salmon and Malcolm, 2011).

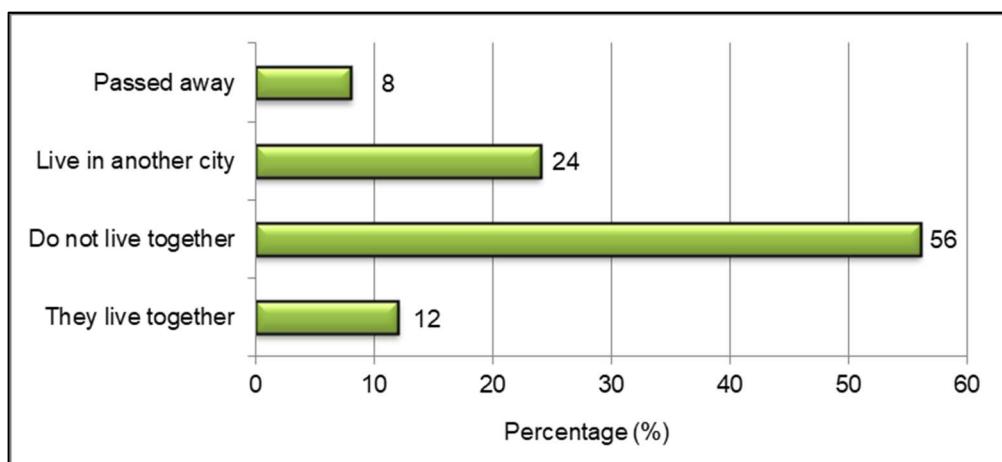
- iii. Women who were not pregnant at the time of the study.
- iv. Women without a diagnosis of diabetes or high blood pressure during pregnancy/delivery, due to possible effects on the infants' growth.
- v. Women without a diagnosis of depression and/or anxiety, because of the possibility of being under pharmacological treatment that could affect the hormonal analysis. Moreover, these conditions could presumably influence the mother's interaction with the infant.
- vi. Where appropriate, maternal grandmothers living in Merida that supported their daughters in the care of their grandchild.

3.7 Information about contact with maternal grandmothers

To identify what kind of information would be required to evaluate potential grandmothers' participation, I decided to apply a questionnaire of contact frequency of the maternal grandmother with non-participant mothers who had infants close to the target age and who were not first-time mothers. The information obtained was about the frequency of support given by the grandmother and the frequency of contact between the mother and the grandmother. I am aware that to obtain more accurate information about the frequency of support from the grandmother in the group of interest, I needed to include women and children who meet the established criteria. However, for this activity, I was not able to find volunteer participants who met the criteria.

Fifty questionnaires were delivered to volunteer women on March 30, 2017. Twenty-five (50%) were answered and returned. First, I asked the mothers if their own mother was alive. Then, if the answer was yes, I asked the mothers where the grandmother was living. According to their responses I found four groups of women (**Figure 3.3**).

Figure 3.3 Results of the questionnaire of frequent contact with the GM



The women that lived with the grandmothers indicated that they received support from them. The average number of days per week that the grandmothers took care of grandchildren was six days per week and the hours per day varied over a range of 4 to 12 hours. All the women indicated that they had contact every day with their mothers and spent time together. Those who reported that the grandmother lived in another city did not receive direct support from them. However, they communicated typically every day, or at least one or two times per week, through phone calls or messages.

The challenge was to analyse the data of those women whose mothers lived in Merida but not in the same house. From this group, seventy-eight percent of the women reported receiving support from the grandmother. The average number of days that the grandmother took care of the child varied between one and six days per week and the hours per day varied between 2 and 12 hrs. The remaining percentage of women reported that they did not receive support from the grandmother unless there was an emergency (for example, if the child was sick and the mother had to attend work). In those cases, they stated that this kind of problem had a frequency of less than once per month. Those who reported receiving support indicated that they communicated and spent time with their mothers almost every day. Women who reported not receiving support stated that they visited their mothers once or two times per week or per month and communicated with them almost every day, or at least one or two times per month.

According to the previous analysis, I decided that participation of the grandmothers would depend on the frequency of support in the care of their grandchildren. This decision was because due to the availability of technological resources such as mobile phones, women can be in regular contact with their mothers despite physical distance. In this sense, I included in the study those grandmothers who provided direct childcare support and had physical contact with the mother-infant dyad at least two times per week for ≥ 3 hrs per day.

3.8 Sample size

The number of participants per group was calculated through Lehr's Equation:

$$N = 16 * (SD^2 / D^2)$$

N=number per group

SD= standard deviation

D= difference between groups

A sample of 32 mother-infant dyads and 32 mother-infant-grandmother triads is required to detect a difference of 0.66 standard deviations between groups in growth outcomes with 80% power, $p<0.05$. The final sample size was 38 dyads mother-infant and 52 triads' mother-infant-grandmother.

I focused on growth outcomes considering that there is evidence suggesting that growth parameters (such as height-for-age) at 2 years are good predictors for long-term health outcomes (Victora et al., 2008).

3.9 Participants recruitment

Because the target population of this research project was infants and their mothers, centres of childcare were my first option to find potential participants that met the inclusion criteria. I located 149 centres around the city. As a first step, I submitted the research protocol to several governmental institutions that were in charge of these centres, such as the Ministry of Education of the State of Yucatan (SEGEY) and the Ministry of Social Development (Sedesol), to get approval to carry out the study. However, I obtained authorization to recruit participants only in 28 centres.

In addition, I made use of my social and work networks to recruit participants and therefore, achieve the objectives set in the project. These networks included:

- i. Local universities. I was able to advertise the project in five local universities and gave several oral presentations of the research to nutrition students to ask for support. Those students that decided to be part of the recruitment period were able to learn all the techniques I used in the field.
- ii. Radio. FD and I did an interview on a local radio channel named Radio Universidad which was aimed at the entire Merida population. The interview was carried out on September 6, 2017, and the aim was to explain the project and make an open invitation to participate for any women who fulfilled the specific criteria.
- iii. Facebook. I advertised the project on the Cinvestav' Facebook page to recruit participants.
- iv. Friends. I asked my friends to advertise the project on their Facebook pages. In addition, they recruited their friends that met the inclusion criteria.
- v. Health centres. I requested authorization to recruit women and children at the Merida's Children Hospital (Hospital de la Amistad Corea-México).

Once I received the relevant authorizations, I visited each centre and explained to each director the purposes of the project. All the directors asked me for a copy of the authorization document to corroborate the authenticity of the approval granted to work in the centres. The reason for this petition was to ensure the security of the children. After one or two weeks, the directors gave me information about the number of infants that filled the age criteria and were the first-born child. Then, I invited their mothers to an informative meeting. I designed the formal invitations which were delivered to the potential participants by the teachers from the centre a week before the meeting.

During the meetings, I explained in detail the purpose of the research and asked attendants for their support (**Figure 3.4**). I provided information sheets of the project to all the parents who attended to the meetings. Those interested in participating filled out the recruitment questionnaire.

Figure 3.4 Informative meeting with the parents



The time period between the first contact with the person in charge of the centre and the informative meeting with parents was between 2 and 3 weeks.

For the other recruitment methods, I used different approaches. At the hospital, I approached women either face-to-face or by distributing information sheets. If the potential participant was interested, I asked her the questions on the recruitment sheet to determine if she met the eligibility criteria. Then, I scheduled a first appointment to explain to their partners and to their mothers if relevant the purposes of the project.

Through Facebook, radio and local universities, women contacted me directly through a phone call or by email. Once I made the first contact with the interested women, I explained the research to them in detail and asked them the questions on the recruitment sheet.

During recruitment, women asked specific questions related to their participation. All questions were clearly answered aiming to reassure the mothers. In some cases, it was necessary to talk individually to each mother to create an atmosphere of trust and to encourage them to take part in the study. Moreover, in some cases it was necessary to do a home visit to explain the project to the mothers' partner to ensure her participation.

All the participants were aware that, to be part of the study, it was necessary that they satisfied the inclusion criteria and, where necessary, that the grandmother would agree to participate. So once I had the recruitment sheet of each participant, I verified if the woman met the criteria and then contacted them via phone call. Scheduling the appointments was not easy because many mothers were working, so they might only be contacted after several phone calls. In some cases it took me between one to three weeks to get in contact with them. At the time of the phone call, mothers decided the day and time for the home visits and most of the appointments were scheduled for the next three to seven days after the call.

In those cases in which grandmothers' participation was necessary, I first asked the women to explain the project to their mothers and promote their participation. Then, the grandmothers were recruited during a face-to-face visit at their homes, in which I explained them the aim of the study and gave them information sheets to read and keep.

The recruitment period lasted 11 months (June 2017 to April 2018). I had to visit the 28 centres of childcare and the hospital several times to try to recruit new participants. Likewise, it was necessary to return several times to the other recruitment networks to find new participants.

More detail about the recruitment process and the inclusion criteria applied in the screening phases before and after the informative meetings with the parents can be seen in **Table 3.2**.

Table 3.2 Overview of the recruitment process

First screening*	Inclusion criteria	Exclusion criteria
Infants	Infants age: ≤ 2.3 years [†] Only child	Infants age: ≥ 2.3 years Has siblings
Second screening[‡]		
Mother	Living in Merida Non-pregnant Married or living with the child's biological father Without a diagnosis of diabetes or high blood pressure during pregnancy/delivery, and depression and/or anxiety	Not living in Merida (i.e. living in villages outside the city) Pregnant or under treatment to get pregnant Married or living with a partner who is not the biological father of the child Diagnosed with diabetes or high blood pressure during pregnancy/delivery Diagnosed with depression or anxiety Under pharmacological treatment for depression/anxiety
Infants	Interested in participate in home visit sessions Free from serious illness that could affect their growth	Not interested in participating in home visits sessions Had illness that could affect growth
Grandmothers	Maternal grandmother Biological mother of the women Living in Merida Supporting their daughters with infant care Interested in participate in home visit sessions	No maternal grandmother No biological mother of the women Not living in Merida (i.e. living in villages outside the city) Not supporting their daughters with infant care Not interested in participating in home visits sessions

*First screening was carried out during the first visit to the childcare centres. Parents of the infants that met the criteria were invited to the informative meetings.

† Parents whose children were aged < 1.7 years were invited to the informative meetings. In those cases in which the parents were interested in participating, I waited for the child to reach 1.7 years.

‡Second screening was carried out after the informative meetings using the information provided by the parents. If the parents met the criteria for participation, I called them to inform about the study and schedule the first home visit.

Note: Through the other recruitment methods, I used a similar procedure. First, I asked the parents about the age of the child and if it was the only child in the family. If the child met the first criteria, I asked the questions related to the second screening and informed them if they met the criteria for participation.

3.10 Data collection

At least two home visits were undertaken with the mother and her child to obtain all the necessary data, and one visit to obtain the grandmother's data.

The first visit with the mother and infant involved the following activities:

- i. Anthropometric measurements of the child and mother, following methods and techniques described in **section 3.10.1**.
- ii. A questionnaire, described in **section 3.10.2**, designed to be answered by the infants' mothers.
- iii. Giving three self-administered assessment scales and three questionnaires about different topics for the women to complete. These instruments are described in **section 3.10.3**.

The first visit with the maternal grandmother involved:

- i. Anthropometric measurements, following methods and techniques described in **section 3.10.1**.
- ii. A questionnaire, described in **section 3.10.2**, designed to be answered by the grandmother.

The second home visit involved the following activities:

- i. Body composition assessment through the measurement of total body water (TBW) using isotope measurements. It was necessary to obtain saliva samples from the mother and urine samples from the child before and after they received an oral dose of deuterium oxide ($^2\text{H}_2\text{O}$) diluted in drinking water/juice/milk. The method used is explained in **section 3.11** and the techniques to obtain the samples, in **section 3.11.3**.

- ii. Collecting saliva samples in both mother and child to measure saliva cortisol levels. Details about the technique and methods used for the saliva collection are explained in **section 3.12.1**.
- iii. Collecting the self-administered scales and the questionnaires.

More detail about the activities carried out in the home visits can be seen in **Figure 3.5** and **Figure 3.6**.

Although the visits were planned to ensure that the mothers and grandmothers invested as little time as possible, there were cases in which I needed three to five visits to collect all the data. In those cases in which the grandmother's participation was necessary, I encouraged the mothers and grandmothers to schedule the visits together. However, it was not always possible due to their weekly activities.

Figure 3.5 Data collection procedure during home visits to the dyad mother-infant

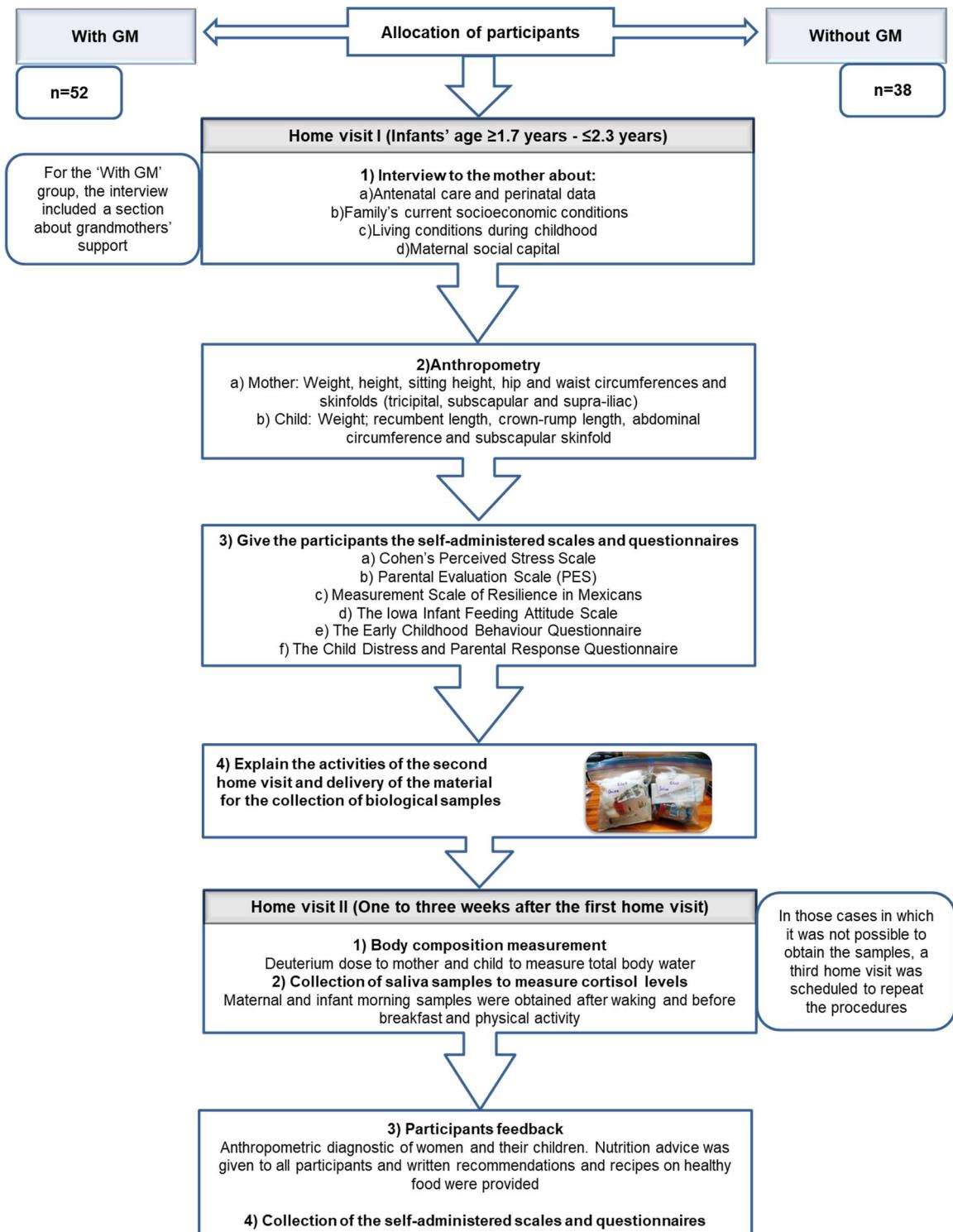
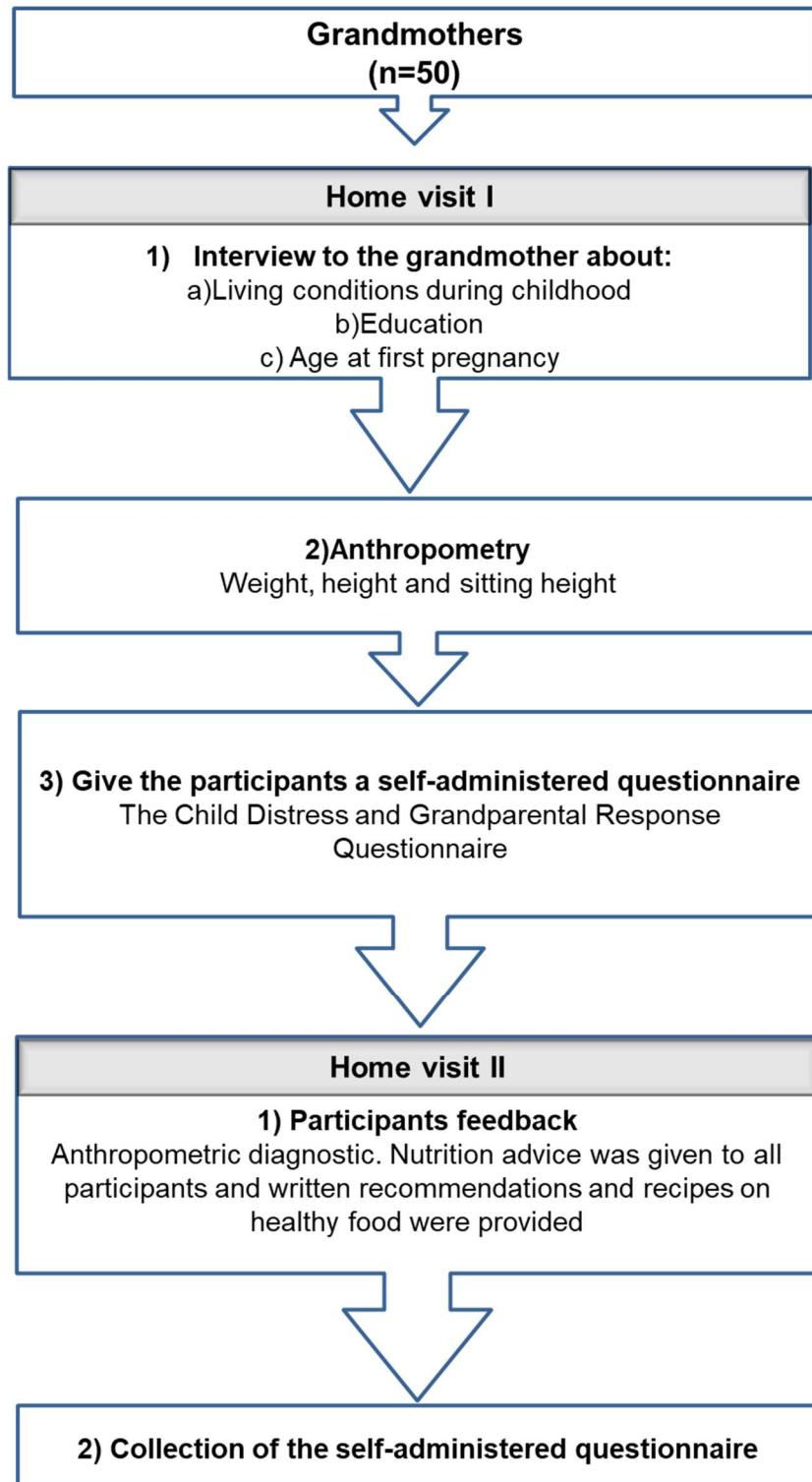


Figure 3.6 Data collection procedure during home visits to the grandmother



3.10.1 Anthropometry

3.10.1.1 Training

GV, one of the assistants that supported me during fieldwork, and I took the anthropometric measurements (**Table 3.3**).

Anthropometric standardisation was undertaken at Cinvestav-Merida between January and March 2017. GV and I did the standardisation. We have extensive experience in taking anthropometric measurements in research. We worked together in three previous studies carried out in Merida (Human ecology of the migration in Yucatan-Conacyt 59994-H, Nutritional Status and Health Outcomes in a Dual-Burden Population of Maya in Yucatan-Wenner Gren Foundation #ICRG-93 and Intergenerational effects on growth in inhabitants of Merida, Yucatan-Conacyt 168047) which involved the measurement of more than 1,400 individuals (children, adolescents and adult women).

Table 3.3 Anthropometric measurements taken in infants and women

Measurements	Infant	Mother	Grandmother
Weight	✓	✓	✓
Height	✗	✓	✓
Recumbent length	✓	✗	✗
Sitting height	✗	✓	✓
Crown-rump length	✓	✗	✗
Hip circumference	✗	✓	✗
Abdominal circumference	✓	✗	✗
Waist circumference	✗	✓	✗
Tricipital skinfold	✗	✓	✗
Subscapular skinfold	✓	✓	✗
Supra-iliac skinfold	✗	✓	✗

We measured 12 non-participant adult women and calculated the Technical Error of Measurement (TEM). For intra and inter-observer TEM involving two measurers, the equation used was:

$$TEM = \sqrt{\left(\sum D^2 \right) / 2N}$$

Where D is the difference between measurements and N is the number of individuals measured. The absolute TEM was transformed into relative TEM in order to obtain the error expressed as a percentage corresponding to the total average of the variable to be analysed. Intra-evaluator and inter-evaluator relative TEM were less than 1% for height, sitting height, and waist and hip circumferences (**Table 3.4**). This indicated acceptable variability in the accuracy of these measurements. Therefore, both were able to take these measurements (**Figure 3.7**). However, as GV had work commitments at Cinvestav at the time of our appointments with the mothers, most of the measurements were taken by myself (197/229).

Table 3.4 TEM calculation involving two measurers

Measurement	TEM (%)
Height	0.29
Sitting height	0.24
Waist circumference	0.83
Hip circumference	0.29
Tricipital skinfold	0.09
Subscapular skinfold	0.09
Supra-iliac skinfold	0.16

Figure 3.7 Anthropometric standardisation in women



In the case of skinfolds, since these can be associated with large measurement error so that interpretation is problematic (Ulijaszek and Kerr, 1999), we decided that a single person (myself) would do all the skinfolds measurements in women and children (**Figure 3.8**).

Figure 3.8 Anthropometric standardisation in infants



The standardization with infants (1.8 to 2.3 yr. old) experienced difficulties as the six non-participant children did not accept being measured several times by several people. Therefore, we decided that only one person (myself) would do all the measurements in the children. Each measurement was taken three times due to infants not being able to adopt a single position for a long period of time.

3.10.1.2 Methods and techniques

All the anthropometric measurements were taken following the manual reference of Lohman et al. (1988). Measurements were taken on the left side of the body and recorded by an assistant. All participants were measured while wearing thin cloth shorts and shirts provided by the research team.

A. Weight

Weight was measured using a Seca® scale, model 881, with 0.05 kg precision and a weight capacity of 200 kg. The scale was calibrated with a standard of known weight before the start of each anthropometric measurement. Each participant was asked to stand in the middle of the scale in bare feet, looking straight ahead and holding this position (**Figure 3.9**). In those cases in which the infant was reluctant to stand in the scale, I weighed the mother and the child together and subtracted from the result the mother's weight.

Figure 3.9 Infant weight measurement during fieldwork



B. Height and recumbent length

In women, height was taken using a moveable Martin type anthropometer, with the woman's head in the Frankfort horizontal plane and ensuring that the subject stood straight. In those cases in which the participant's knees angled in and touched each other when the legs were straightened, the feet were separated so that the medial borders of the knees were in contact but not overlapping (**Figure 3.10**).

In infants, recumbent length was measured with a Rollameter100 (Harlow Healthcare Rollameter100), covering the range 0-100cm. The measurer and the assistant took care to keep the child's head in the correct position and to maintain the child's body aligned at right angles (**Figure 3.10**).

Figure 3.10 Maternal height and infant length measurements



C. Sitting height and crown-rump length

Sitting height was taken using a moveable Martin type anthropometer and the women were asked to sit on a table, located on a flat surface, with the legs hanging freely and the hands resting on the thighs with their head in the Frankfort horizontal plane. In infants, crown-rump length was measured with the Rollameter100. The measurer and the assistant kept the child's head in the correct position and maintained the body aligned at the right angles. One measurer raised the child's legs so that the thighs were at an angle of 90° and moved the instrument against the buttocks (**Figure 3.11**).

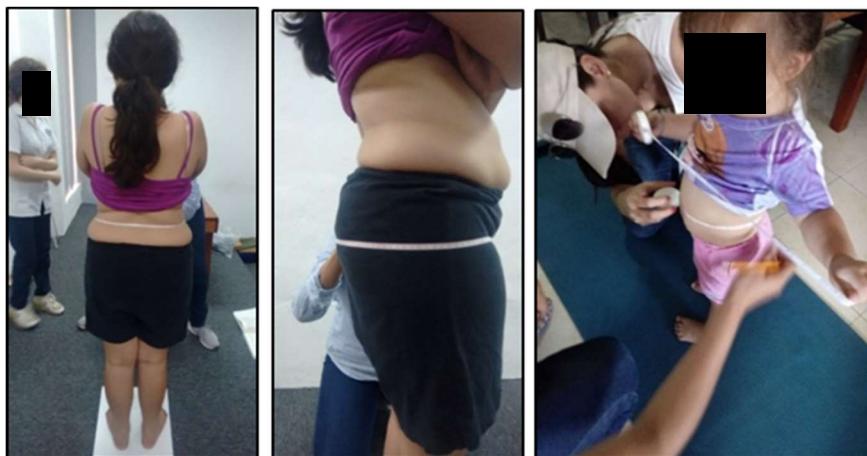
Figure 3.11 Infants' crown-rump length measurement during fieldwork



D. Circumferences

Waist, hip, and abdominal circumferences were measured using a non-stretchable fiberglass tape Seca®. All the maternal circumferences were measured with the subject standing. The waist was measured at the 'natural waist', halfway between the 10th rib and iliac crest and the hip at the maximum extension of the buttocks (Figure 3.12). Abdominal circumference was measured at the level of the greatest anterior extension of the abdomen in a horizontal plane (Figure 3.12).

Figure 3.12 Infants' and women circumferences measurements



E. Skinfolds

In infants, subscapular skinfold was measured in triplicate using a Holtain calliper (range 0mm to 46mm, dial graduation of 0.2mm), while in mothers, triceps, subscapular and supra-iliac skinfolds were measured with a Harpenden calliper (range 0mm to 80mm, dial graduation of 0.2mm). Callipers were zeroed before each use. For all skinfolds, an eyeliner pencil was used to mark the points to measure. The measurer held the calliper with the right hand and the thumb and index fingers of the left hand were used to grasp and elevate a fold of skin and subcutaneous fat approximately 1 cm proximal to the marked measurement point. The calliper was placed on the skinfold at 90° to the long axis and the measurement was taken four seconds after the pressure of the calliper was released.

For triceps, the posterior half upper arm length was identified and marked and for the subscapular, the inferior angle of the left scapula. The measurement was made diagonally inclined at approximately 45° to the horizontal plane. In some cases, specifically in obese women, it was necessary to ask the participants to bend the elbow and place the arm behind the back. Finally, for the supra-iliac skinfold, an oblique skinfold was grasped just posterior to the midaxillary line following the natural cleavage lines of the skin approximately at 45° just over the iliac crest.

3.10.2 Interview

3.10.2.1 Interview development

The interview was designed to be answered by the infants' mothers and grandmothers. The questions for the mothers were focused on four categories: Antenatal care and perinatal data, the family's current socioeconomic conditions, the mothers' living conditions during childhood, and maternal social capital. The first category focused on antenatal and postnatal events, such as age at pregnancy, length of gestation, antenatal care and type of delivery, birth weight, birth length, breastfeeding duration and timing of weaning. Secondly, the next section was focused on the current socioeconomic conditions of the family and included questions about family size, number of rooms used for sleep, type of building materials of the household, household assets and access to basic services such as piped water and electricity. In addition, this section included questions about the level of education, total years of study and the occupation of the infant's parents.

The third section of the interview aimed to obtain information about the mothers' living conditions during her childhood. This included questions such as number of siblings, birth order, family size, number of rooms for sleep, household services and assets and parental occupation.

The last section was focused on obtaining information about the social capital of the mother. This included questions about social support from the family and friends and networking activities such as belonging to religious, neighbour and/or social groups. In addition, it included questions about notions of trust and reciprocity and finally, specific questions about the help and support provided by her mother, i.e., the maternal grandmother, including how the mother felt about this support.

A second interview was carried out with the maternal grandmother, and was designed to obtain information about the grandmother's living conditions during her own childhood (place of birth, family size, number of rooms used to sleep, household services and assets, and parental occupation), her schooling and her age at first pregnancy.

Regarding the recall questions relating to the mothers' and grandmothers' childhood, I am aware that long-term memory might not be reliable. However, I tried to be more accurate by specifying a childhood period that the women were being asked to recall. My experience in the field suggests that including a specific childhood period helped to the women to recall most of the information especially that addressed ownership of household assets and access to household services, such as sanitation facilities.

All the questions included in each section of both interviews were based on the literature review and the long-term experience of the research team within Merida, Yucatan.

3.10.2.2 Interview procedure

A total of four interviewers collected data during fieldwork. GV, HC and I had conducted interviews as part of previous studies, and conducted the interviews of the pilot study of this research. Researcher CB did not have experience conducting interviews. However, her familiarisation with these interviews consisted of observing interviews during data collection and conducting interviews under my supervision.

On average, interviews with the mothers lasted between 40 and 50 minutes and between 30 and 40 minutes with the grandmothers. We did 90 interviews with mothers and 50 interviews with the grandmothers. Most of the interviews were conducted by HC (51/140) (**Figure 3.13**) and myself (70/140).

Figure 3.13 Interview during fieldwork



3.10.3 Self-administered scales and questionnaires

During the first home visit I left three self-administered scales on maternal psychological state and three questionnaires focusing on the mother's perception of, and experience with, her infant, which the mother filled in at her convenience.

The self-administered scales were:

A. Cohen's Perceived Stress Scale (PSS)

PSS is a psychological self-report instrument that consists of 14 items for measuring the perception of stress on a scale of five, from zero (never) to four (very often) (Cohen et al., 1983) on the basis of their occurrence during one month prior to the study. Seven out of the fourteen items of PSS-14 are considered negative (1, 2, 3, 8, 11, 12, 14) and the remaining seven as positive (4, 5, 6, 7, 9, 10, 13), representing perceived helplessness and self-efficacy, respectively. Total scores for PSS-14 range from 0 to 56 with a higher score indicating greater stress. It has been shown to be reliable and has been validated and used extensively globally, including in three national surveys in the US (Cohen et al., 1983; Cohen and Janicki-Deverts, 2012). This questionnaire has also been translated and validated for the Mexican population (Ramírez and Hernández, 2007).

B. Measurement Scale of Resilience in Mexicans (RESI-M)

The RESI-M is a self-report instrument constructed from two scales: The Connor-Davidson Resilience Scale (CD-RISC) (Connor and Davidson, 2003), and The Resilience Scale for Adults (RSA) (Friborg et al., 2003).

The RESI-M comprises 43 items with four answer options that assess five factors:

- a. Strength and self-confidence (questions 1 to 19): clarity in the objectives, effort to achieve them, self-confidence, optimism, and tenacity.
- b. Social competence (questions 20 to 27): competence of individuals to relate to others and make friends, make people laugh and enjoy a conversation.
- c. Family support (questions 28 to 33): support provided by the family, loyalty among family members and the time-shared with them.
- d. Social support (questions 34 to 38): support from friends in difficult times, people willing to help and encouragement to take care for others.
- e. Structure (questions 39 to 43): ability of people to organize activities, as well as having rules and activities even in difficult times.

The scores on the factors are obtained by the sum of items and a high score reflects strength in each of them (Palomar Lever and Gómez Valdez, 2010).

C. Parental Evaluation Scale (PES)

The Parental Evaluation Scale is a self-administered measure to assess satisfaction and self-efficacy feelings about motherhood in women with children aged 0-2 years (Farkas-Klein, 2008). The scale is composed of 10 items rated from zero (totally disagree) to 10 (totally agree). The final score is obtained by the sum scores divided by 10, with a higher score indicating a greater perception of self-efficacy and maternal satisfaction. Internal consistency of the scale was moderate (Cronbach's $\alpha = 0.67$) (Farkas-Klein, 2008). The original version of this scale is in Spanish (Farkas-Klein, 2008).

The questionnaires were:

A. The Iowa Infant Feeding Attitude Scale (IIF-AS)

The scale, comprising 17 items, can be utilized to measure maternal attitudes toward infant feeding methods (e.g., breastfeeding, formula feeding) (Mora et al., 1999). The scale was designed to cover various dimensions of infant feeding. For example, questions were written concerning the costs of infant feeding, nutrition and infant bonding. Respondents are asked to indicate the extent to which they agree with each statement, on a five-point Likert scale ranging from 1 (strongly disagree) to 5 (strongly agree). These scores are then computed so that a high score reflects a preference for breastfeeding (Mora et al., 1999). This questionnaire has a Spanish validation version (Jácome and Jiménez, 2014), and a translated and validated version for the Mexican population (Aguilar-Navarro et al., 2016).

B. The Early Childhood Behaviour Questionnaire (ECBQ)

The Early Childhood Behaviour Questionnaire has been designed to assess temperament in children between the ages of 18 and 36 months (Putnam et al., 2006). This instrument comprises 201 items that assess the following 18 domains of temperament:

1. Activity Level/Energy: Level (rate and intensity) of gross motor activity, including rate and extent of locomotion.
2. Attentional Focusing: Sustained duration of orienting on an object of attention; resisting distraction.
3. Attentional Shifting: The ability to transfer attentional focus from one activity/task to another.
4. Cuddliness: Child's expression of enjoyment in and molding of the body to being held by a caregiver.
5. Discomfort: Amount of negative affect related to sensory qualities of stimulation, including intensity, rate or complexity of light, sound, texture.
6. Fear: Negative affect, including unease, worry, or nervousness related to anticipated pain or distress and/or potentially threatening situations; startle to sudden events.
7. Frustration: Negative affect related to interruption of ongoing tasks or goal blocking.
8. High-intensity Pleasure: Pleasure or enjoyment related to situations involving high stimulus intensity, rate, complexity and novelty.
9. Impulsivity: Speed of response initiation.

10. Inhibitory Control: The capacity to stop, moderate, or refrain from behaviour under instruction.
11. Low-intensity Pleasure: Pleasure or enjoyment related to situations involving low stimulus intensity, rate, complexity, novelty and incongruity.
12. Motor Activation: Repetitive small-motor movements; fidgeting.
13. Perceptual Sensitivity: Detection of slight, low intensity stimuli from the external environment.
14. Positive Anticipation: Excitement about expected pleasurable activities.
15. Sadness: Tearfulness or lowered mood related to exposure to personal suffering, disappointment, object loss, loss of approval, or response to other's suffering.
16. Shyness: Slow or inhibited approach and/or discomfort in social situations involving novelty or uncertainty.
17. Sociability: Seeking and taking pleasure in interactions with others.
18. Soothability: Rate of recovery from peak distress, excitement, or general arousal.

The previous domains form three broad factors: Surgency (Activity Level, High Intensity Pleasure, Impulsivity, Positive Anticipation, and Sociability), Negative Affectivity (Fear, Sadness, Discomfort, Frustration, Motor Activation, Unsoothability, Shyness, Perceptual Sensitivity) and Regulatory Capacity/Effortful Control (Attention Shifting, Attention Focusing, Inhibitory Control, Cuddliness, Low Intensity Pleasure). Scale scores are calculated as the average of ratings for all completed items, with high scale scores corresponding to high levels of the temperament dimension. This questionnaire has been translated and validated for the Mexican population by Vianey-Mendez in 2010 (available at <https://research.bowdoin.edu/rothbart-temperament-questionnaires/>).

Scores were computed in SPSS by the following method:

- 1) Items indicated with an R on the items-by-scale list are reverse-scored. This was done by subtracting the numerical response given by the mother from 6. Thus, a response of five becomes four, four become two, three becomes three and two remains four and one becomes five.
- 2) I summed the scores for items receiving a numerical response (I did not include items marked "does not apply" or items receiving no response). For example, given a sum of 50 for a scale of 12 items, with one item receiving no response, two items marked "does not apply," and 9 items receiving a numerical response, the sum of 50 would be divided by 9 to yield a mean of 5.56 for the scale score.

C. The Child Distress and Parental Response Questionnaire

It has been suggested that infants perceived as difficult are fed more to quieten them, which could be related to subsequent fatness (Carey, 1985; Barr et al., 1989; Wells et al., 1997; Wells, 2003b; Braet et al., 2007; Faith and Hittner, 2010; Stifter et al., 2011; Wasser et al., 2011; McMeekin et al., 2013; Miller et al., 2016). Evidence has suggested that parents' practice of using food to soothe infant distress could be related to child weight status. Moreover, one study found that mothers who used food to soothe rated themselves lower in parenting self-efficacy and their children higher in temperamental negativity (Stifter et al., 2011).

This questionnaire was designed to obtain from the mothers and grandmothers information about how often the child was distressed by different perceived causes (frustration, tiredness, discomfort or hungry) and how they responded to relieve the distress (Wells, 1998). Women were asked if they used foods to relieve the stress and the type of foods used to this. The caregivers' response to child behaviour may have important consequences for the development of young children eating patterns.

3.10.3.1 Piloting of self-administered scales and questionnaires

Although all the instruments were appropriate for use in the Mexican population, it was necessary to organize a pilot study to ensure that the women were able to understand the questions being asked. First, I submitted, on February 20, 2017, the pilot study to the Ministry of Education to get approval to carry out the piloting activities at a childcare centre. I received the approval on March 16, 2017, and started the activities on March 24, 2017.

As a first step, I decided to work with a psychologist and two teachers from this centre. The main reason for including them was that they are in frequent contact with mothers. Hence, they are very familiar with the common language used by this group. I gave them the questionnaires and they returned them to me on March 27, 2017, in a feedback session. Then, I reworded and reformatted the instruments based on feedback and impressions from these participants.

Later, on March 30, 2017, the updated instruments were given to 15 non-participant mothers from this centre. Eleven participants returned the questionnaires on April 7, 2017. The comments and observations made to each question were discussed and afterward, I was able to refine and reword all the instruments.

A summary of all the instruments used for recruitment and data collection are provided in **Table 3.5**.

Table 3.5 List of questionnaires that were used in the study

No	Questionnaires	Type of questionnaire*	Stage	Method of conduct**
1	Screening questionnaire		Recruitment	
2	Mothers questionnaire	Developed by researchers	First home visit	Administered
3	Grandmothers questionnaire		First home visit	
4	Cohen's Perceived Stress Scale			
5	Measurement Scale of Resilience in Mexicans	Validated		
6	The Iowa Infant Feeding Attitude Scale			
7	Early Childhood Behaviour Questionnaire		Deliver to the participant women in the first home visit to be completed at their convenience	Self-administered
8	Parental Evaluation Scale	Adapted		
9	The Child Distress and Parental/Grandparental Response Questionnaire ^{&}	Translated		

***Developed by researchers:** Questionnaires were elaborated by the researcher in charge of the project and reviewed by the research team. The questions included were elaborated according to the literature review and previous researcher experience; **Validated:** Translated and validated for the Mexican population; **Adapted:** The original version of this questionnaire is in Spanish and was validated in the Chilean population (mothers with infants between 0 to 2 years). The scale was tested in the pilot study of the current project and changes were made due to differences between Mexican and Chilean Spanish; **Translated:** Developed by Jonathan Wells (1998) and translated by the researcher for application.

****Administered:** Led by the researcher; **Self-administered:** Completed by the mother/grandmother.

[&]Grandmothers only completed the 'Child Distress and Grandparental Response Questionnaire'

3.11 Measuring body composition

3.11.1 Total body water (TBW): deuterium isotope analysis

Body composition was assessed through the measurement of TBW using isotope measurements. Depending on their weight, mothers and children received an oral dose of deuterium oxide ($^2\text{H}_2\text{O}$) diluted in drinking water/juice/milk. The enrichment of deuterium in urine and saliva was measured using isotope-ratio mass spectrometry.

Although saliva samples are the best option, for small children saliva sampling was problematic. Therefore, I collected urine samples before and 6 hours after the drink. The time of urination was taken as the midpoint between the last time the cotton wool was dry and, checking every 30 minutes, the first time it was wet. For mothers, saliva samples were taken before and 5 hours after the drink at least 30 minutes after the last ingestion of food or drink. The equilibration period is longer for urine (4-6hrs) than saliva (3-4hrs) (Schoeller and van Santen, 1982), although the saliva equilibration period is extended in the obese (Bray et al., 2001; Haroun et al., 2005).

Drinks taken during the equilibration period were accounted for as they can dilute the isotopes and lead to a lower concentration and therefore, overestimation of TBW.

3.11.2 Dose preparation and administration

I received training for the preparation and administration of the doses during October and November 2016 at Great Ormond Street Institute of Child Health. Subsequently, in May 2017, I received additional training and specific instructions, via Skype, from my supervisor on how to calculate the amount of dose consumed by the participants.

A. For the preparation of doses:

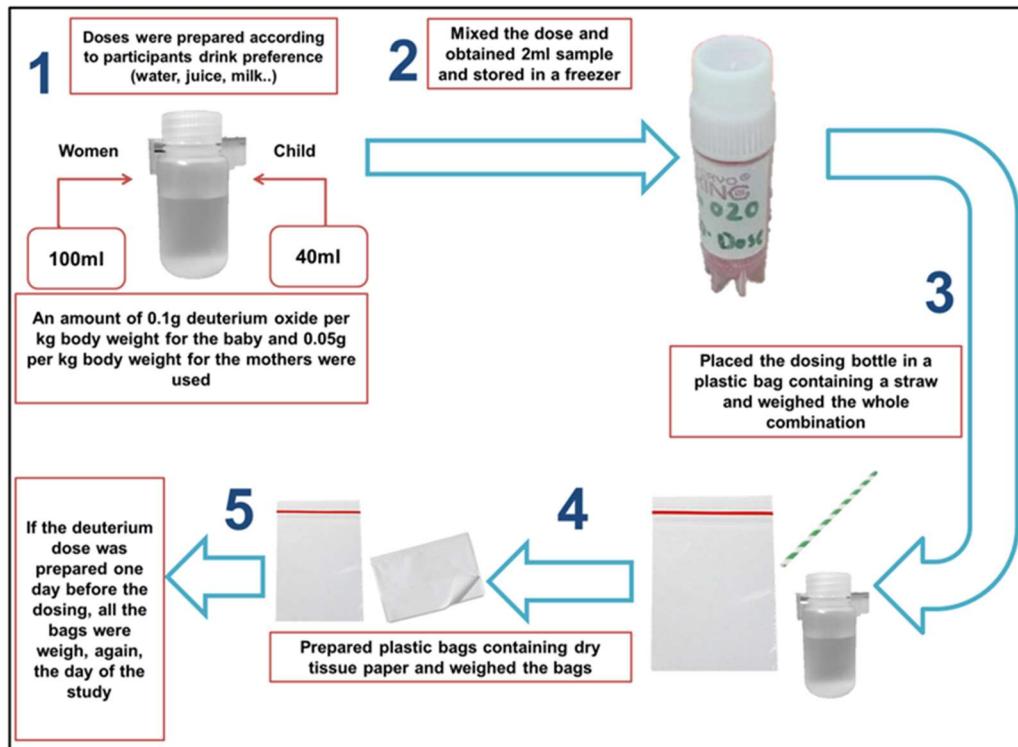
- i. I prepared the deuterium dose according to the child's and mother's weight and according to the participants' drink preference. I used an amount of 0.1g deuterium oxide per kg body weight for the baby and 0.05g per kg body weight for the mothers, and used a nominal amount of 40ml of fresh bottled water/juice/milk for the baby and 100ml of fresh bottled water for the mothers.

- ii. I extracted the amount of deuterium using a sterile syringe and filtered it with a 0.2 micro-litre micropore filter.

- iii. I mixed the dose for one minute and used a transfer pipette to obtain a 2ml sample from the middle of the bottle. Then, I placed the sample in a plain 2ml sample tube and stored the dose tube in the freezer (-80 °C).
- iv. I placed the dosing bottle in a plastic bag containing a straw and weighed the whole combination. I ensured that the dosing bottle was in the middle of the scale. If the deuterium dose was prepared one day before the dosing, I weighed it again, the day of the study.
- v. Finally, I prepared plastic bags containing dry tissue paper (to be used to mop up spillage when I gave the dose to the child). I weighed and recorded the weight of the bags.

A summary of the doses preparation is provided in **Figure 3.14**.

Figure 3.14 Procedure for preparation of doses



B. The day of the administration of the doses:

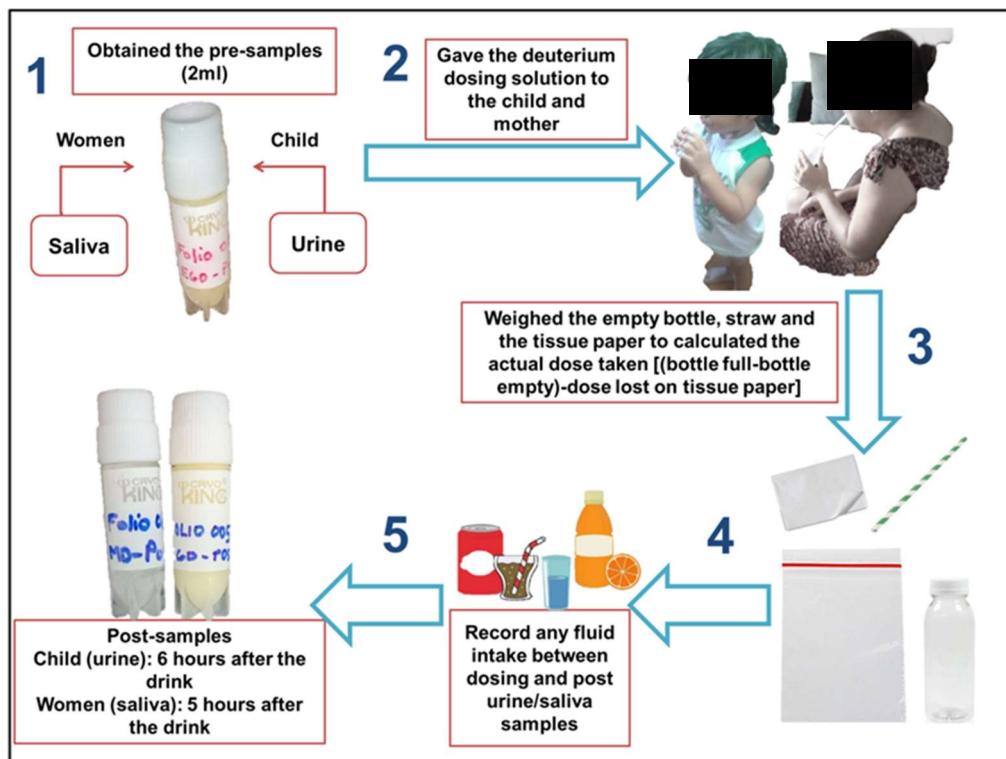
- I. After obtaining the pre-dose samples, I gave the deuterium dosing solution to the child (**Figure 3.15**) and the mother ensuring that as much as possible was swallowed with minimal spillage.
- II. I used the dry tissue to wipe any spillage of the dose and put the wet tissue paper back in the same plastic bag and sealed it immediately to prevent evaporation. Immediately, I weighed this bag containing the damp tissues.
- III. I put the empty bottle and the straw in the same plastic bag and sealed it as soon as possible and re-weighed it.
- IV. I calculated the actual dose taken by the babies and the mothers from the dose given (bottle full-bottle empty) minus the dose lost on tissue paper.
- V. I recorded the time the dose was given and indicated to the mother the time at which post-dose sample needed to be taken.
- VI. I asked the mother to record any fluid intake between dosing and post urine/saliva samples.
- VII. I asked the mother to record the time she obtained the post urine/saliva samples.

A summary of the administration procedure is provided in **Figure 3.16**.

Figure 3.15 Doses administration during fieldwork



Figure 3.16 Procedure for doses administration



In those cases where the child wanted to use a favourite bottle/cup to drink the dose (Figure 3.17):

- I. I poured the dose from the bottle into the child's bottle.
- II. After the child had drunk the dose, I put both the empty bottle and the child's bottle/cup back into the bag in which I brought the dose in, and weighed it.
- III. Then, I dried the child's bottle/cup and weighed it alone without any bag.
- IV. Now, the dose taken was calculated taking into consideration the weight of the child's bottle/cup.

Figure 3.17 Dose administration using the child's own cup



All the weighing was done with an OHAUS® CL201 Compact Scale. This scale has a capacity of 200g, readability of 0.1g, repeatability (standard deviation) of 0.1g and a linearity of $\pm 0.1\text{g}$.

3.11.3 Collection of biological samples for body composition analysis

A. Urine samples

The main method used to obtain the urine sample from infants was putting cotton wool inside a dry diaper and then compressing the wet wool with a 20 ml syringe to obtain the sample (Figure 3.18). However, because some children were in the 'potty training' stage and some children did not want to wear their diaper with the cotton inside, I had to include the following methods:

- I. Ask the child to urinate inside a sterilized plastic cup and then transfer the urine with a pipet into a tube.
- II. Use a special plastic bag to collect the urine. It was a sterilized plastic bag with an adhesive tape on one end, made to fit over the infant's genital area.

All urine samples were stored in a portable freezer with cool bags during transportation in the field and then, in a freezer at the Cinvestav laboratory at -80°C .

Figure 3.18 Method used for infants' urine collection



In those cases where the mother decided to help me obtain the urine and saliva samples, I gave them packages with the necessary material (Figure 3.19). To make sure that mothers followed the instructions to obtain the samples I: a) explained to them the methods to obtain the samples at the end of the first home visit, b) left them the written instructions and c) called them via phone one day before the second visit to explain, again, the instructions.

In addition, during the day of the dosing, I was in frequent contact with the mother via text messages or phone calls to remind them about the instructions and the time at which they had to obtain the post-samples.

Figure 3.19 Material given to the mothers during fieldwork



B. Saliva samples

To obtain the saliva samples from women it was planned to use a Salivette® or a saliva collection aid for passive drool. However, when I initiated the process to purchase these devices, I was informed that the Mexican Ministry of Health was not accepting the entrance of these materials to the country, since they were considered biohazard materials. In consequence, I started the piloting of two methods using alternative devices:

- I. Passive drool. I used a sterilized plastic cup to collect the saliva and then a pipette to transfer the saliva to the tubes.
- II. Cotton wool. As with the infants, I asked the mothers to put cotton wool inside their mouths and then, I used a 20ml syringe to compress the wet cotton and obtain the sample.

According to the six non-participant women that helped me in the piloting, the first option was messy and took a lot of time whereas the second method, although it was unpleasant to have the cotton in the mouth, was easier and faster. Because I wanted to avoid that a method would discourage some women from obtaining their samples, I decided to present both techniques to the participants and let them choose the most comfortable method for them.

3.12 Measuring stress in the mother and the infant: cortisol levels in saliva

3.12.1 Collection of biological samples

A. Saliva samples: Piloting and collection

The main concern around the research team was to apply the best method to obtain the infant's saliva samples. Therefore, I decided to pilot different methods in non-participant children that attended the centre of childcare where I piloted other instruments.

I did three visits to this centre during April 2017 and worked with nine non-participant infants. The methods I decided to pilot were (**Figure 3.20**):

- I. Cotton wool with a thread tied around and hanging out of the infant's mouth to prevent swallowing. Then, the wet cotton wool was introduced to a 20ml syringe and compressed to obtain the sample. Not all infants accepted the cotton wool in their mouths and among those who accepted, I was not able to obtain the complete sample because they did not salivate enough. Hence, I had to repeat the process several times to fill the tube. However, I observed that children got tired of repeating this activity.
- II. Spit into a sterilized plastic cup. I could not get the sample since the children refused to spit inside the cup.
- III. Drippers to obtain the saliva direct from the infant's mouth. I did not obtain the sample because although the children accepted the droppers in their mouths, they sucked the dropper releasing the saliva.
- IV. Pacifiers. I tried to use pacifiers to get infants to salivate. However, not all children agreed to use the pacifier and those who accepted and managed to salivate refused to spit inside the cup or to use the dropper to obtain the saliva.
- V. Spoons. The aim was to collect the saliva with a small spoon, but the children did not accept the teaspoon being introduced into their mouth.

Figure 3.20 Material used during pilot study for saliva collection



From the five methods listed above, I decided to use the first one in the field (**Figure 3.21**). Although it was challenging to obtain the complete sample, I considered that this was the best method that brought me closer to the research goals. In addition, I decided to use cotton wool without a thread because, during the piloting, the children pulled the thread several times, making it difficult to obtain the sample.

Additionally, I had an interview with the paediatrician from this centre and with one of the teachers in charge of the infants. During my interview with the paediatrician, I explained to her the aims of the study and the difficulties I experienced during the piloting of the methods to obtain the saliva samples. I asked her, according to her experience, about the possibility to ask mothers their help to obtain the samples. She told me that it was a better approach since children feel more comfortable with their mothers.

On the other hand, during my interview with the teacher, she explained two factors that made it difficult to obtain the saliva samples. First, that I was a strange person to the children and second, that I was asking them to perform an activity that at school and at home they are asked not to do (spit). Moreover, she commented that it was expected that some children were reluctant to do the activities because, at two years of age, the children are reaffirming what they like to do or not.

Therefore, I decided to involve the mothers in the saliva collection and to establish a rapport with the children during the first home visit to facilitate the sample collection during the second visit. In those cases in which the child was very reluctant to use the cotton wool, even with the mother's help, I decided to not obtain the samples. At the end of the study, I was not able to obtain the saliva samples of nine infants.

Figure 3.21 Use of cotton wool for infants' saliva collection



To obtain the women's saliva samples I used the methods described in **section 3.10.3 (B)**.

Each participant (mother and child) provided one saliva sample. Morning samples were taken after waking and before breakfast and physical activity since these activities may compromise the assay and cortisol values (Kirschbaum and Hellhammer, 1989; Mello, 2010; Sarkar et al., 2013). In addition, participant women were not breastfeeding at the time of the study to avoid variation in cortisol levels by breastfeeding status (Neelon et al., 2015). An important point to be considered is the possible impact of the female menstrual cycle on the cortisol awakening response (Wolfram et al., 2011). The only information I obtained was that women were not menstruating at the time of the saliva sampling.

For all the saliva samples, both in infants and women, I was very careful to avoid air bubbles in the samples and to obtain the 2 ml of saliva. All the samples were stored in a portable freezer with cool bags during transportation in the field and then, stored in a freezer at the Cinvestav laboratory at -80°C.

3.12.2 Analysis to determine cortisol concentrations in saliva

I obtained 2-mL of saliva from both mother (n= 80) and child (n=71). Saliva samples were labelled and immediately placed in portable freezer with cool bags upon collection. Then, the samples were stored at the Ecotoxicology Laboratory at Cinvestav. In February 2019, frozen samples were shipped to the Haematology

Laboratory of the Centre of Regional Research 'Dr. Hideyo Noguchi' in Merida for analysis. Dr. Alejandra Núñez de la Mora, a researcher from the Institute of Psychological Research in Xalapa, Veracruz, Mexico, supported me during the analysis.

Salivary cortisol levels were determined by enzyme-linked immunosorbent assay (ELISA) using DRG® Salivary Cortisol ELISA kits. Saliva samples were defrosted at room temperature on the day of assay. Once thawed, 200 μ L was aliquoted in Eppendorf tubes (**Figure 3.22**) and then vortexed and spun at 3200 rpm for 15 min in a centrifuge (Thermo Scientific Sorvall Legend Micro 21R).

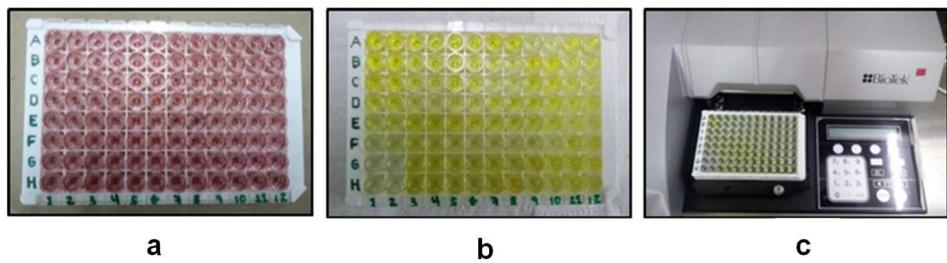
Figure 3.22 Preparation of the samples at the laboratory for ELISA analysis



Reagents and serial dilutions of the cortisol standard were prepared as per kit instructions. First, 100 μ L of standards, controls, and samples to be analysed were pipetted in duplicate into appropriate wells across a 96 well microtiter plate. Then, 200 μ L of enzyme conjugate was pipetted into each well (**Figure 3.23a**). The microtiter plate was incubated for 60 minutes at room temperature on a shaker at 300 rpm. Subsequently, the plate was washed five times with diluted wash solution (400 μ L per well) and 200 μ L of substrate solution was added to each well and incubated for 30 minutes at room temperature. Finally, to stop the enzymatic reaction, 100 μ L of stop solution was added to each well (**Figure 3.23b**).

To determine the absorbance (OD) of each well, the plate was read in a Biotek microtiter plate reader at 450 nm (**Figure 3.23c**) using Gen 5 Software Version 2.0. Salivary cortisol concentrations were calculated using MyAssays Free Cortisol in Saliva ELISA software (available at <https://www.myassays.com/free-cortisol-in-saliva-elisa.assay>). The mean intra-assay CV was 3.3% and the mean inter-assay CVs were 1.2% for low controls and 6.6% for high controls.

Figure 3.23 ELISA procedure



3.13 Feedback to participants

During the recruitment meetings, women were informed that during the second home visits they would receive their anthropometric measurements of themselves and those of their children. In Mexico, the nutritionist is trained and legally qualified to give a nutritional diagnosis. All of the research team, except FD and GV, were nutritionists. This allowed quality nutritional information to be given to the mothers by trained personnel.

The nutritionists were able to tell the mothers if the children were growing well (based on length-for-age and weight-for-length), and mothers and grandmothers received their weight diagnosis and recommendations about healthy weight. In addition, nutrition advice was given to all participants and written recommendations and recipes on healthy food were provided.

3.14 Data handling

3.14.1 Data input and cleaning

After completing the interview applied in the first home visit, each questionnaire was reviewed by the interviewer to identify any mistakes or missing data. In those cases

in which missing data or mistakes were found, the research team obtained the correct information during the second home visit. Then, the complete interviews were scanned and stored at Cinvestav. For security reasons, the data set was only stored in a computer without internet access and in two external memory disks. Only the research team had access to the computer and the external memory disks.

All interview and anthropometry data were transferred from paper documents to a spreadsheet in Access by HC, GV and myself, and then proofread, twice, by CB and myself. Then, the data was exported to Microsoft Office Excel 2010 and Stata 14 for analyses.

Dataset cleaning consisted of verifying the matching between the dataset and questionnaires. CB and I did this activity during the last four weeks of fieldwork. Twenty-five mistakes in data entry of 7127 observations (0.35%) were found in anthropometry, and no mistake was found in the deuterium dose data. We found 116 mistakes in data entry of 57,797 (0.20%) observations in all data obtained through the interview and the self-administered instruments filled by the participants. All the errors found in the database were corrected.

3.14.2 Derived variables

A. Anthropometry

Anthropometric measurements were converted to Z-score values using the zanthro command in Stata. For height/recumbent length, weight-for-length and BMI I used the 2006 WHO Growth Standards. For adult women I assumed that their height remains constant after the age of 19 years, and this age was used for the estimation of Z-scores. Early and recent studies on secular changes in height in the Mexican and Yucatecan population support this statement (McCullough and McCullough, 1984; Siniarska and Wolanski, 1999; Malina et al., 2004). For trunk and leg length, I used the 1990 British Growth References (Dangour et al., 2002).

The trunk length/leg length ratio (TL/LL), relative leg length index (LL*100/height) and relative trunk length index (TL*100/height) were also calculated for all participants. Leg length was calculated indirectly by subtracting sitting height from total height.

Infants were classified as stunted if below -2 SD of the reference for length-for-age. Weight-for-length was used to determine under- and over-nutrition in children. Risk

for overweight and obesity was categorised when children were above +1 SD and +2 SD of reference respectively.

Women were classified as stunted if they were below -2 SD of the WHO reference for height-for-age. I used values of waist circumference to determine the risk of abdominal obesity in mothers. I chose the cut-off point of 88 cm proposed by The National Cholesterol Education Program (NCEP) Adult Treatment Panel III (ATP III). This cut-off point is the most commonly used by the Mexican Health authorities. Waist-to-hip ratio ((waist circumference / hip circumference)*100) is another important indicator for health risk. I applied the cut-off point of ≥ 0.85 proposed by the WHO to determine risk of developing metabolic complications in mothers.

BMI is a risk indicator of disease; as BMI increases so does the risk for some diseases. Some common conditions related to overweight and obesity include cardiovascular diseases, high blood pressure, osteoarthritis, some cancers, diabetes and premature death (Weisell, 2002; Meigs et al., 2006; Baker et al., 2007). For adult women, overweight was defined as a BMI-for-age value over +1 SD and obesity as a BMI-for-age value over +2 SD according to 2006 WHO Growth Standards.

The conventional BMI cut-offs commonly used by the Mexican Health authorities are established in the Official Mexican Norm 008 (NOM-008-SSA3-2017) for the treatment of overweight and obesity (**Table 3.6**).

Table 3.6 BMI cut-offs point established by the NOM 008 according to age

Age (years)	Indicator	Reference	Cut-off points	
0-2	Weight-for-length	WHO	Overweight >2 and 3 SD	Obesity >+3SD
20-59	BMI according to height	---	Not stunted: 25.0 to 29.9 kg/m ² Stunted: 23 to 24.9 kg/m ²	Not stunted: ≥30 kg/m ² Stunted: ≥25 kg/m ²
≥60	BMI	WHO	28 to 31.9 kg/m ²	≥32 kg/m ²

B. Socioeconomic data

The number of people at home was divided by the number of rooms used for sleeping, to give an index of 'overcrowding' experienced by the infants, and by the women during their own childhood. According to the National Council for the Evaluation of Social Development Policy of Mexico (CONEVAL), overcrowding occurs when the number of persons per room is ≥ 2.5 (information available at: <http://www.coneval.gob.mx/contenido/prensa/6102.pdf>).

3.15 Data Analysis

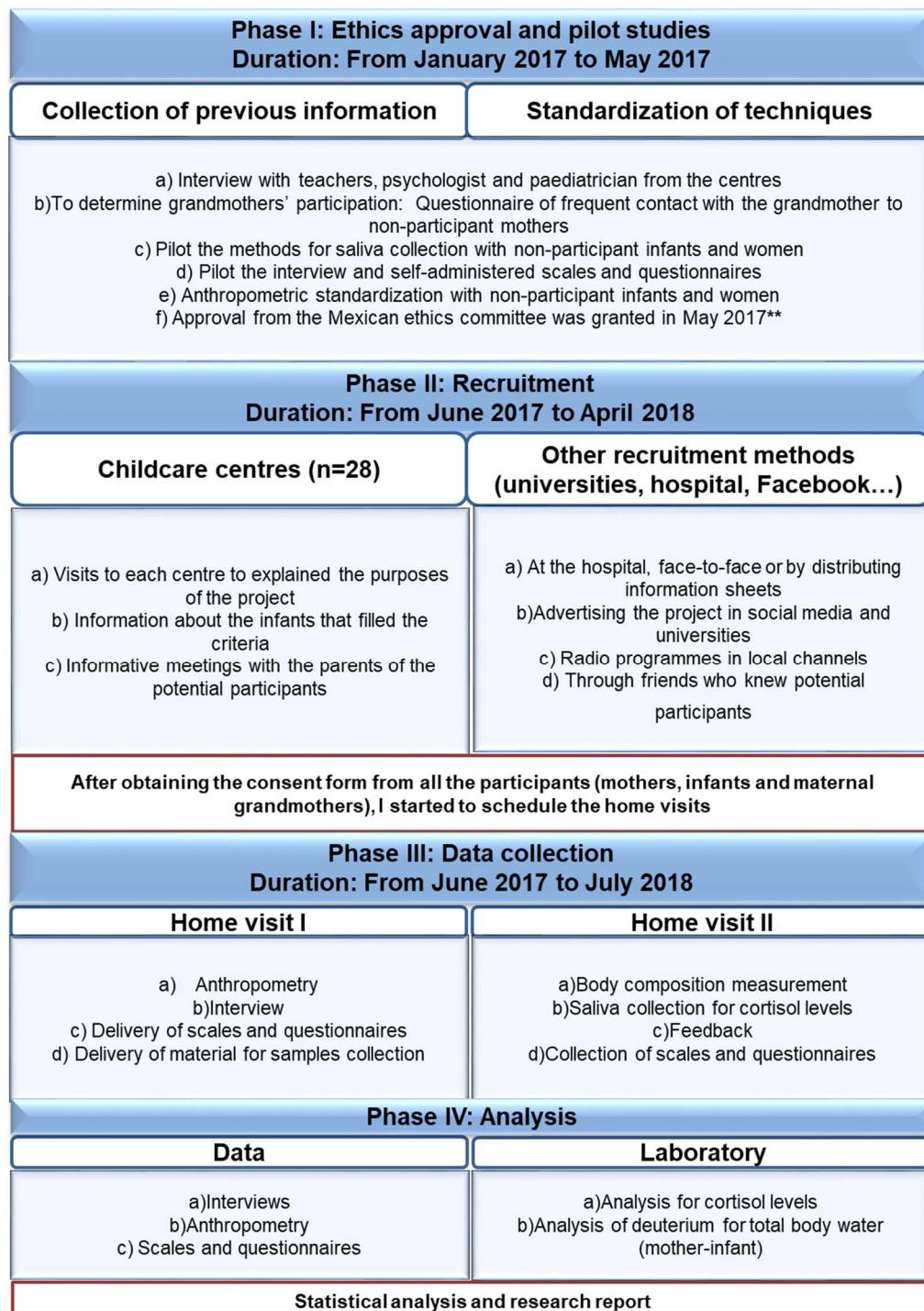
In this section, I describe the tests applied for the analysis of the data collected during the fieldwork. First, it is important to point out that the statistical tests were chosen depending on the data distribution and the type of variables. I applied tests of normality to determine the goodness of fit of the distributions, and obtained histograms to graphically verify data distribution. Overall, central tendency descriptive statistics and measures of variability, frequencies and percentages were obtained to describe the data, and Chi-Square tests, T-tests and Wilcoxon rank sum tests were used to assess differences between groups (e.g. by age-group, sex, grandmaternal group). All statistical analyses were performed using the Stata/IC 14.2 for Windows statistics package (StataCorp LP, 2015) and the significance level was set at $\alpha=0.05$.

Although multiple testing was done to test multiple outcomes within the primary and secondary hypothesis, the p-value was not adjusted for multiplicity since the comparisons responded to the same question in different ways (ie whether GM support was associated with a difference in the outcomes), and all the comparisons pointed to the same conclusion. Further details and description of specific statistical analysis are described in the following chapters.

3.16 Research flow chart

A flow chart providing an overview of the study design and the overall data and sample collection procedures is shown below (Figure 3.24).

Figure 3.24 Overview of the study



**Recruitment and data collection in Merida did not start until the approval of the Mexican ethical committee was obtained. This process lasted four months (January to May 2017).

CHAPTER 4

RECRUITMENT RESULTS AND BACKGROUND CHARACTERISTICS OF THE SAMPLE

4.1 Introduction

This chapter provides an overall description of recruitment, such as the number of participants recruited through the childcare centres, Facebook, radio, universities and health centres (n=90 mother-infant dyads and 52 maternal grandmothers). This general description includes the data obtain from each mother-child dyad and grandmother-mother-child triad and the age distribution of the participants.

The chapter also provides a contextual framework for the research project by describing the living conditions and socioeconomic status of the families as well as a description of the antenatal care and perinatal data of the mothers and infants. For this description, I analysed two sections of the interview applied to the participant women. The first section addressed antenatal and postnatal events, such as the age at pregnancy, length of gestation, antenatal care and type of delivery, birth weight, birth length, breastfeeding duration and age of weaning. The second section addressed the current socioeconomic conditions of the family and included questions about family size, number of rooms used for sleep, type of building materials of the household, household assets and access to basic services such as piped water and electricity.

The study sample was categorised into two groups: 'with GM support' and 'without GM support', and their results are compared. The 'with GM support group' consisted of the mothers and infants who received childcare support and had physical contact with the maternal grandmother at least two times per week for ≥ 3 hrs per day, and the 'without GM support group' by the mother-infant dyads that did not received direct support nor had frequent physical contact with the grandmother because she had passed away, lived in another city or was not available to provide this support.

Finally, it is relevant to mention that the no-grandmother group included women with support from the paternal and the maternal grandmother, but below the threshold established in the criteria of the study. In **Table 4.1**, I present the data provided by women from both groups about the frequency of support received by the grandmothers.

Table 4.1 Frequency of support provided by the grandmothers

	Days per week	Hours per day	Other
With GM (n=52)	2-7	3-12	---
Without GM (n=38)			
No support (n=32)	0	0	---
Only in emergencies (n=3)	0	0	1 day per month only if the child get sick
Other caregiver (paternal GM) (n=3)	1-2	2-4	---

It is clear that the groups are very different in terms of grandmaternal support, which indicates that overlap between grandmaternal groups is highly unlikely to have contributed to the results presented in the following sections.

The main aims of this chapter were: a) to compare my overall study sample with the general Yucatecan population and b) to identify differences between the groups.

4.2 Statistical analysis

All participant women (n=90) completed the socioeconomic and health background sections of the interview. Some data was presented as frequency and/or percentages, while univariate analyses (Chi-square/Fisher Exact test) were used to compare groups. Continuous data were tested first for normality distribution using histograms, Q-Q plots and statistical tests such as the Kolmogorov-Smirnov test and Shapiro-Wilk test. For normally distributed data, mean \pm standard deviation (SD) was presented along with the statistical results (T-test) for group comparisons. In those cases in which non-parametric test (Mann-Whitney) was used, the median \pm interquartile range (IQR) was presented.

4.3 Recruitment and data collection

Fieldwork started the first week of June 2017 after I had obtained the authorizations to visit the childcare centres to recruit participants. The overall periods of recruitment and data collection were simultaneous and a total of 28 childcare centres were visited, but potential participants were only found in 21. Overall, 183 mother-infant dyads were identified as potential participants through the information provided from the centres. On average, 73% (n=134) of the mothers attended the recruitment sessions. From

the mothers who attended, 104 met the criteria, but only 65% (n=68) accepted to take part in the study.

Given the considerable number of mothers that did not attend the meetings (27%), other sessions were organized and carried out on different days and times to try to increase the sample size. However, none of the mothers invited to the new sessions attended. Although the mothers did not give the reason why they did not attend the new sessions, the experience of the research team on previous projects suggests it was probably the type of occupation of these mothers that impeded them participating, due to the possibility of monetary or work implications.

Through other modes of recruitment (Facebook, radio, universities and health centres), I was able to recruit 22 more dyads. In July 2018, fieldwork concluded with 90 mother-infant dyads. After allocation of the participants to the appropriate group, the samples were as follows: 52 mother-child-grandmother triads and 38 mother-child dyads. However, during the data collection, not all the participants were able to provide all the required information (**Table 4.2**). In all cases, I analysed all the available data.

Table 4.2 Data obtained from the participants

	Infant (n=90)	Mother (n=90)	Grandmother (n=52)
Anthropometry	89	90	48
Questionnaire	---	90	50
Self-administered questionnaires and scales	---	80	45
Saliva samples for cortisol levels	71	80	---
Saliva and urine samples for total body water calculation	80	80	---

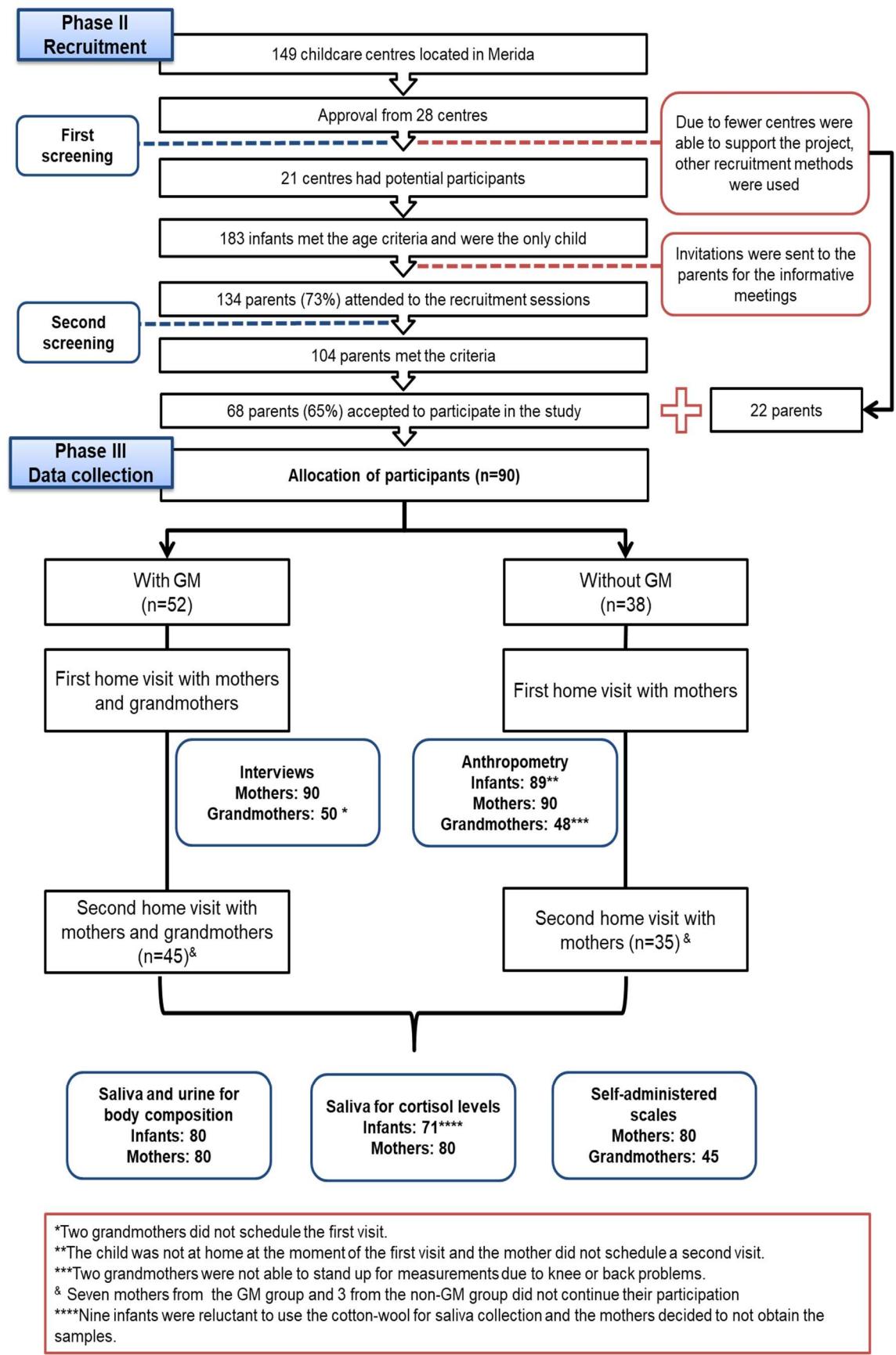
Overall, mothers that decided not to continue with the second part of the study (n=10) did not give their reasons for leaving the project. After several phone calls (mean of 10 calls) to schedule the second visit and without receiving any response, the research team decided to drop the case. This resulted in a lack of biological samples and self-administered questionnaires in 10 cases and in a lack of information on the grandmother in two cases.

Women that discontinued their participation were younger (median 26.7 vs 30.7 years) and less educated (median 15 vs 17 years) than women who completed the study ($p<0.05$).

In relation to the saliva samples that I was not able to collect from children ($n=9$), the decision to not obtain the samples came from the mother. In these cases, the children were very reluctant to use the cotton-wool or any other device to obtain the samples. I observed that the persistent crying and negative attitude of these children started to stress the mothers so, to try to avoid them dropping out of the study entirely, I asked them if they wanted to schedule another day to try to obtain the sample. The nine mothers decided not to schedule for another day and continued participating only in the activities related to body composition measurement.

More detail about the recruitment process and the overall data obtained from the participants is given in **Figure 4.1**.

Figure 4.1 Flow diagram of the recruitment and data collection processes and results



4.4 Age and sex distribution of the participants

In the entire sample of children, mean age was 1.9 years (± 0.2 , range from 1.7 to 2.3 years). The sample contained 43 boys and 46 girls. The distribution of age by infant sex is shown in **Table 4.3**. There were no significant differences in age between boys and girls ($z = -0.95$, $p=0.34$, Wilcoxon rank sum).

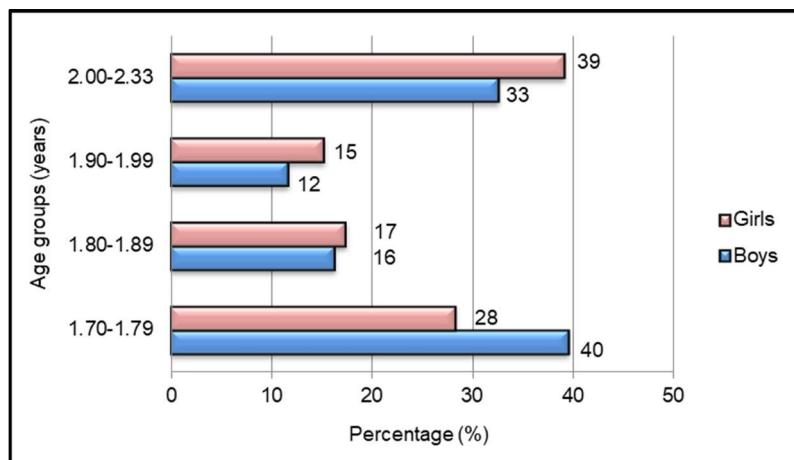
Table 4.3 Infants' age and sex distribution

	n	Mean	SD	p25	Median	p75	IQR
Boys	43	1.9	0.2	1.8	1.8	2.1	0.3
Girls	46	2	0.2	1.8	1.9	2.1	0.3
Total	89	1.9	0.2	1.8	1.9	2.1	0.3

SD: Standard deviation; p25: 25th percentile; p75: 75th percentile; IQR: Interquartile range

Using Chi-square analysis I found no significant differences in the proportion of boys and girls by age group ($\chi^2 = 1.33$, $p = 0.72$) (**Figure 4.2**), indicating an adequate balance in age by sex.

Figure 4.2 Infant age distribution



The mothers' mean age was 30.2 (± 5.3) years. Eighty-nine per cent of women were born and raised in Merida, Yucatan, and 11% were born in another Mexican state. Those who were born in a different state had moved to Merida, on average, 24 (± 8) years previously. The grandmothers' age range was between 40 and 67 years

(median= 58 years, IQR= 52.2-62.6 years) (**Table 4.4**), with 66% of the grandmothers born and raised in Merida, and 34% in small villages near Merida.

All mothers were married or lived with the biological father of the participant infant, had no diagnosis of diabetes and/or high blood pressure during pregnancy/delivery, or depression and/or anxiety, and were non-pregnant at the time of the study. All grandmothers were the biological mothers of the mothers and all children were the first-born child. None of the women had a previous unsuccessful pregnancy before they got pregnant with the participant child.

Table 4.4 Grandmothers' and mothers' age distribution

Age groups (years)	Mother		Grandmother	
	n	%	n	%
20.0 - 24.9	17	19		
25.0 - 29.9	29	32		
30.0 - 34.9	27	30		
35.0 - 39.9	14	16		
40.0 - 44.9	3	3	4	8
45.0 - 49.9			5	10
50.0 - 54.9			7	14
55.0 - 59.9			15	30
60.0 - 64.9			13	26
65.0 - 69.9			6	12
Total	90	100	50	100

In summary, few centres were authorized to be part of the study which made recruitment of participants difficult. However, through other means, I was able to increase the sample. Moreover, all the participants met the inclusion criteria and I obtained an adequate sample balanced by infant age and sex.

4.5 Sociodemographic data

4.5.1 General description

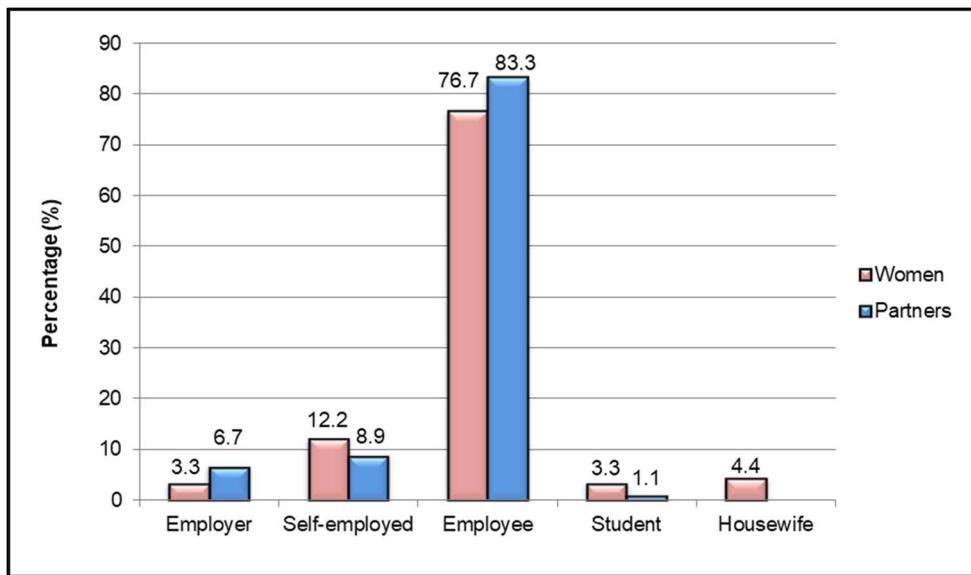
Eighty-four per cent of the women were married while 16% were unmarried and living with the partner at the time of the study. Most (78%) of the households were nuclear (mother, partner and the child), and 28% were extended families in which at least one other paternal and/or maternal family member shared the home. In 14 of these 25 extended families, the maternal grandmother was one of the members. Some mothers pointed out that the reason why the grandmother was living with them was that the partner spent six days per week working outside the city, and the grandmothers supported them by helping care for the child.

In relation to the number of rooms that the families used for sleep, 89% of the families used one to two rooms and 11%, three to four rooms. The median of persons per room in the sample was three (IQR= 1). According to the National Council for the Evaluation of Social Development Policy of Mexico (CONEVAL), it is considered overcrowding when the number of persons per room is ≥ 2.5 . In this sample, 63% of the studied families were considered “overcrowded” at the moment of the research.

However, during the home visits, I was able to observe that of the 57 mothers that mentioned use of only one room for sleep, 35 had at least one other room that could have been used. Once I finished the interviews, I asked them why they used only one room for sleep if they had another available for the child. All mothers answered that the child was too young to sleep alone and they were afraid that something might happen during the night. Therefore, the ‘overcrowding’ experienced by the families was more related to the fear of leaving the children alone in their rooms than not having the appropriate household conditions.

Sample distribution by employment status indicated that most of the infants belonged to families of employees (**Figure 4.3**). However, this was expected considering that one of the requirements to have access to the nursery service is to perform a paid job.

Figure 4.3 Distribution of parental employment status



Seventy-seven per cent of the women and 66% of the partners had completed an undergraduate degree and 18% and 23% respectively, high school degree. The median total years of education of the women and their partners was 16.5 (IQR=15-18 years) and 16 (IQR=12-17 years) respectively. According to official reports (INEGI 2016, data available at: <https://www.inegi.org.mx/>), the average duration of education among Yucatecan women and men in 2015 was 8.6 and 9 years respectively, which is equivalent to middle school. Based on this local data, my sample showed greater educational attainment.

All the families had access to piped potable water in their houses. However, due to the high levels of groundwater contamination in Yucatan (Perez-Ceballos and Pacheco-Avila, 2004), this water is not frequently consumed by the population. Although it has been advised that the chlorination or boiling of water before consumption minimizes the risk of diseases, most of the population has decided to obtain drinking water that they consider safe from other sources. In my sample, all families decided to purchase bottles of purified water that had been mechanically filtered or processed. In the local context, this water is commonly sold in convenience stores or by delivery trucks. The cost of a 20-litre bottle is between Mex\$28 (£1.1 GBP) and Mex\$30 (£1.2 GBP) depending on the brand. The Coca-Cola Company and PepsiCo produce the most common brands of purified water.

All the participant women reported having toilets with sinkhole and electricity at home. Information about the household conditions was also obtained. The type of building materials used for the construction of the house is relevant when analysing the living conditions and well-being of families. Regarding the floor, 94.4% of the houses were built with cement and slab covers, and 5.6% with cement. According to INEGI, by 2006 (Data available at: <https://www.inegi.org.mx/>), 1.6% of the Yucatecan population had a dirt floor; none of my sample showed this condition. In addition, a high percentage of the houses had ceilings (98%) and walls (97%) built with blocks and concrete.

Overall, a high percentage of the houses were constructed of durable materials (blocks, bricks, concrete, cement). The importance of these materials lies in their resistance to the high temperatures and rain that characterize the tropical climate of Yucatan. For example, in the case of ceilings and walls, the main difference between construction blocks with concrete and construction blocks without concrete is that the first is more resistant to water and humidity.

It must be noted that my sample is likely over-representative of women that use the service of specific childcare centres and who come from similar socioeconomic backgrounds. The idea to recruit participants from the 149 centres allocated around the city was to incorporate a wide range of socioeconomic variability. However, the Mexican authorities only approved carrying out the study in 28 centres. Therefore, this situation could have unintentionally excluded women from different socioeconomic conditions. In addition, although the other recruitment methods (Facebook, radio, universities) helped me to increase the sample, this could have increased any socioeconomic bias in the sample.

4.5.2 Living conditions and sociodemographic data stratified by grandmaternal groups

According to the information presented in **Table 4.5**, a high percentage of women and their partners were performing a paid job regardless of the study group they belonged to. Moreover, the percentage of overcrowding among the groups was similar, and women and their partners did not differ in their total years of education by grandmaternal group.

Regarding the sanitary conditions (access to potable water and toilet) and the characteristics of the house (household building materials), I did not find statistical differences between the groups in any of the variables presented in **Table 4.5**.

Table 4.5 Living conditions and sociodemographic data stratified by grandmaternal groups

Variable	With GM (n=52)	Without GM (n=38)	Differences
Age (years)	29.9	30.9	$\chi^2_{(2)} = -0.9, p=0.34$
<u>Employment status (%)</u>			
Employee	82.7	68.4	
Employer/Self-employed	13.5	18.4	Fisher's exact: $p = 0.20$
Other (student or housewife)	3.8	13.2	
<u>Partners' employment status (%)</u>			
Employee	86.5	78.9	
Employer/Self-employed	13.5	18.4	Fisher's exact: $p = 0.69$
Other (student)	---	2.6	
Mother's education (years)	16.2	16.8	$z = 0.20, p=0.83$, Wilcoxon rank sum
Partners' education (years)	14.9	15.5	$z = 0.80, p=0.40$, Wilcoxon rank sum
<u>Crowding index (%)</u>			
With overcrowding	65.4	60.5	$\chi^2_{(1)} = 0.22, p=0.64$
Without overcrowding	34.6	39.5	
<u>Access to potable water (%)</u>			
100	100	100	---
<u>Toilet access with sinkhole (%)</u>			
100	100	100	---
<u>House floor with cement (%)</u>			
With slab covers	94.2	94.7	
Without slab covers	5.8	5.3	Fisher's exact: $p = 0.65$
<u>Ceilings with blocks (%)</u>			
With concrete	98.1	97.4	
Without concrete	1.9	2.6	Fisher's exact: $p = 0.67$
<u>Walls with blocks (%)</u>			
With concrete	96.2	97.4	
Without concrete	3.8	2.6	Fisher's exact: $p = 0.62$

GM: Grandmother

4.6 Antenatal care and perinatal data

4.6.1 Women's antenatal care data

All the mothers had access to antenatal care services during their pregnancy and took prenatal micronutrient supplements such as iron, folic acid, and multivitamins which were provided by the public health system or prescribed by the private health system. Women did not remember the specific number of medical appointments, but reported that they attended their facility at least once per month throughout their pregnancy.

Infectious diseases were common health problems during pregnancy with a prevalence of 56%. Urinary tract infections represented 64% of the cases, followed by vaginal infections (36%). Concerning tobacco exposure, 13.3% of the mothers mentioned that they cohabited with smokers during pregnancy but none of the women reported having smoked during pregnancy.

On average, as a group, the mothers in the overall sample became pregnant at 27.4 (± 5.4) years of age. According to recalled data, the median weight before pregnancy was 56.5kg (IQR=52-63kg) and median pre-pregnancy BMI was 23.7 kg/m² (IQR=21.7-26.7). Ninety-three per cent of the women gained weight during pregnancy with a median of 10kg (IQR=9-14kg). The remaining 7% lost weight (between 4kg and 20kg) by medical recommendation.

4.6.2 Infants' data

Information on the duration of pregnancy and the infant's birth weight and length was obtained in two ways. First, I asked the mothers to report these data from recall and 77% of the mothers provided this data. Then, I asked the mothers to provide me with the infant's birth certificate that also contains the required information. Eighty per cent of the participants had a copy of the certificate at the moment of the interview. Due to the lack of statistical difference between the information provided by the mothers and the information provided by the certificates (Duration of pregnancy: $z = 0.17$, $p=0.86$, Wilcoxon rank sum; Birth weight: $z = 0.02$, $p=0.98$, Wilcoxon rank sum; Length at birth: $z = 0.61$, $p=0.54$, Wilcoxon rank sum), the information gathered in certificates was kept and recalled values were used in those cases where birth certificates were not available.

All children were born in hospitals (public or private) with 70% born by caesarean and 30%, vaginally. The median duration of gestation was 39 weeks (Min=34, IQR= 38-39, Max=42); 14% of the infants were born preterm (between 34 and 37 weeks of gestation). On average, the birth weight of children was 3.1 (± 0.5) kg; only 8% of the infants had a birth weight lower than 2.5kg. The median length at birth was 50 cm (Min=43, IQR=49-51, Max=54). No statistical differences were found in these variables between boys and girls (**Table 4.6**).

Table 4.6 Infants' birth data by sex

Variable	Boys (n=43)		Girls (n=47)		Differences Boys-Girls	
	Mean (\pm)	Median (IQR)	Mean (\pm)	Median (IQR)	mean	95% CI
Gestation weeks	38.8 (1.4)	39.0 (38-40)	38.6 (1.3)	39.0 (38-39)	0.23*	-0.31;0.77
Birth weight (kg)	3.1(0.5)	3.1(2.7-3.4)	3.0 (0.4)	3.0 (2.7-3.4)	0.09*	-0.11;0.29
Length at birth (cm)	49.5 (2.2)	50.0 (48-51)	49.5 (2.1)	50.0 (49-51)	-0.02*	-1.01;0.97

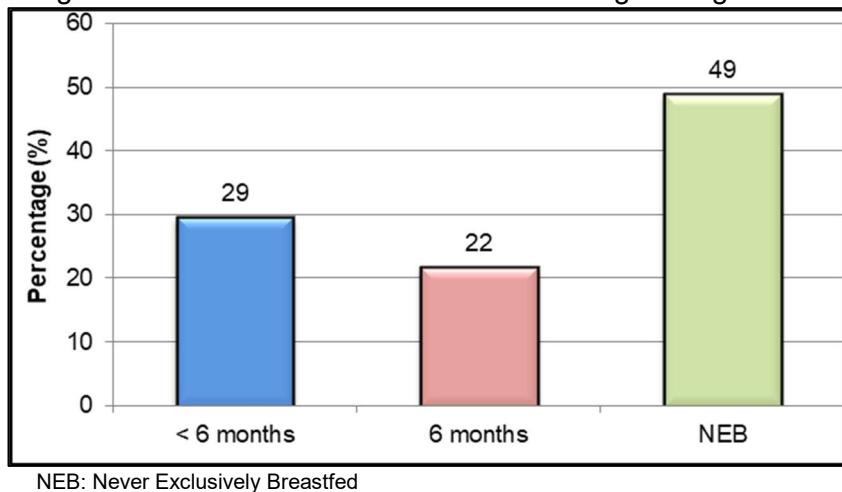
\pm : standard deviation; IQR: Interquartile range

*p>0.05

Most children (98%) consumed breastmilk, but only 22% of children were exclusively breastfed throughout the first six months of life (**Figure 4.4**), while 50% of the children were fed both breast-milk and formula from birth.

The median age of weaning (cessation of breastfeeding) among the children was six months (IQR = 3 – 10 months) and the median age at which the infants started complementary feeding was also six months (Min= 4 months, IQR= 5-6 months, Max=12 months); 62% of the children initiated their complementary feeding at six months of age.

Figure 4.4 Duration of exclusive breastfeeding among infants



4.6.3 Antenatal care and perinatal data stratified by grandmaternal groups

Regarding infectious diseases during pregnancy, no differences were found among the groups (Table 4.7). Moreover, exposure to cigarette smoke during pregnancy showed no difference between the groups. No differences were found in age at pregnancy, weight before pregnancy or weight gain during pregnancy among the groups (Table 4.8). In terms of BMI, women that received support from their mothers showed a higher rate of excess weight before pregnancy, although it did not reach statistical significance (Table 4.7). I did not find differences in the duration of pregnancy or the infant's weight and length at birth by grandmaternal groups (Table 4.8).

Table 4.7 Distribution of maternal pregnancy-related variables

Variable	With GM (n=52)	Without GM (n=38)	Differences
Mothers			
Infectious diseases (%) [#]			
Urinary tract	67.0	61.0	$\chi^2_{(1)}=0.18$, $p=0.67$
Vaginal infections	33.0	39.1	
Smoking status (%)			
Non smoker	87.0	87.0	$\chi^2_{(1)}=0.18$, $p=0.67$
Lived with smokers	13.0	13.0	
BMI before pregnancy (%)			
Healthy weight	57.7	71.1	$\chi^2_{(1)}=1.70$, $p=0.19$
Excess weight**	42.3	28.9	

GM: Grandmother

Only 50 mothers reported having an infection during pregnancy (23 from the grandmother group).

** Excess weight: overweight + obesity

Table 4.8 Distribution of maternal pregnancy-related variables and infants' birthing data stratified by grandmaternal groups

Variable	With GM (n=52)	Without GM (n=38)	Differences With GM – Without GM
Mothers			
Age at pregnancy (years)	27.0 (4.8)	27.9 (6.2)	-0.6 -3.4; 2.3
Weight before pregnancy (kg)	60.3 (12.8)	59.7 (13.8)	1.5 -5.1; 8.2
Weight gain during pregnancy (kg)*	11.9 (4.3)	12.0 (5.9)	-0.7 -3.6; 2.2
Infants			
Gestation weeks	38.7 (1.1)	38.7 (1.6)	0.0 -0.7; 0.7
Birth weight (kg)	3.0 (0.4)	3.2 (0.5)	-0.2** -0.4; -0.0
Length at birth (cm)	49.3 (1.9)	50.0 (2.3)	-0.9 -1.9; 0.1

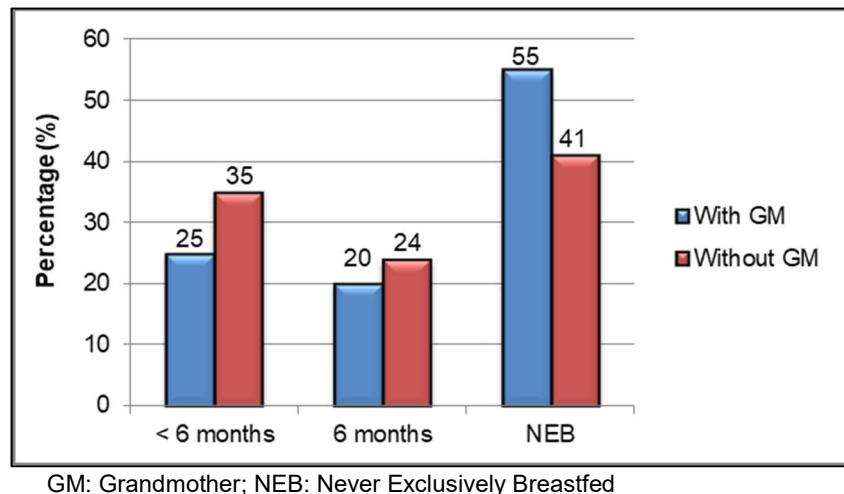
GM: Grandmother

* Six cases were excluded due to women lost weight during pregnancy

** $p=0.05$

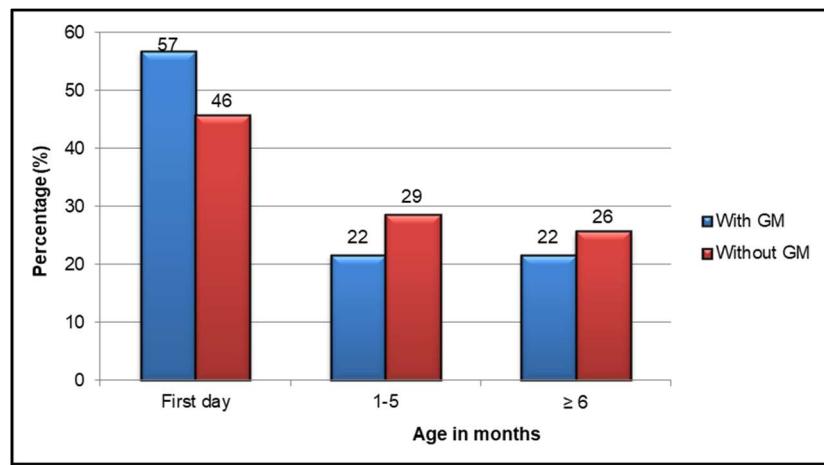
Although 60% of the women from the grandmother group received information from their mothers about the importance of exclusive breastfeeding, this group had a higher percentage of children who never exclusively breastfed (Figure 4.5). However, this difference was not significant among the groups ($\chi^2_{(2)} = 1.8$, $p=0.41$). Forty-two per cent of the grandmothers recommended their daughters to breastfeed exclusively during the first six months, while 52% emphasised the importance of breastfeeding without specifying any particular optimal duration.

Figure 4.5 Duration of infants' exclusive breastfeeding stratified by grandmaternal groups



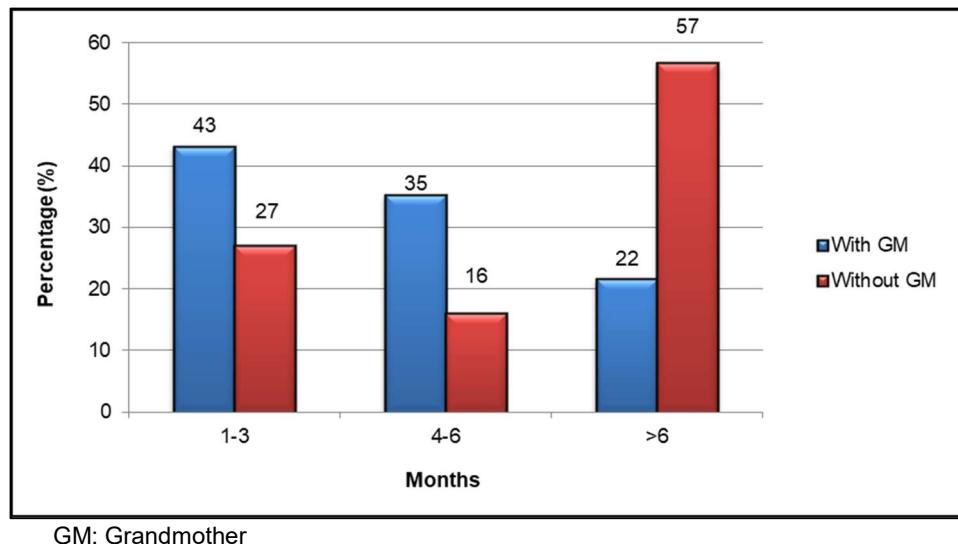
Although not significant ($\chi^2_{(2)} = 1.1$, $p=0.58$), formula milk was introduced at younger ages in children whose mothers received support from the maternal grandmother (Figure 4.6).

Figure 4.6 Infants' age at which formula milk was introduced



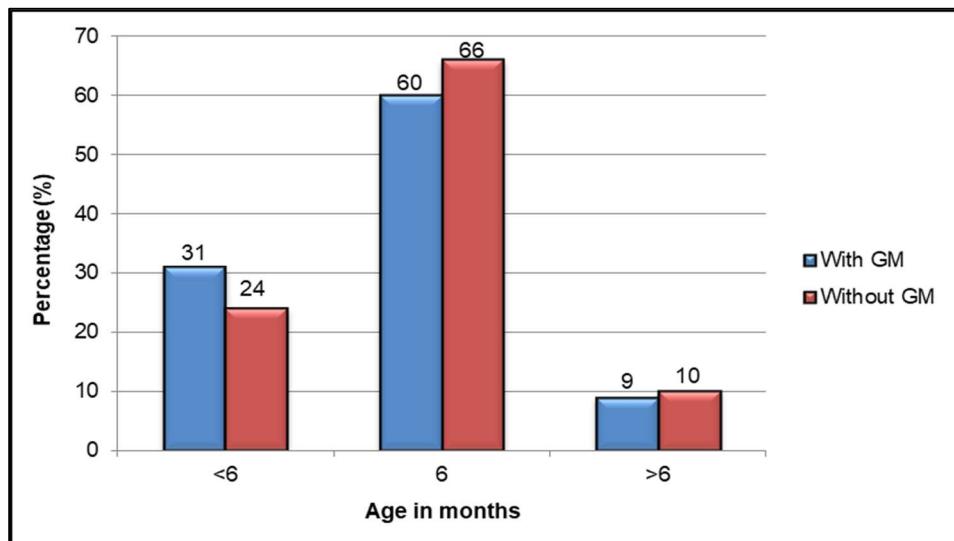
According to Figure 4.7, women who did not receive support from the grandmother breastfed their children for longer (>6 months) and this difference reached statistical significance ($\chi^2_{(2)} = 11.7$, $p=0.003$).

Figure 4.7 Duration of breastfeeding by grandmaternal groups



Finally, the age of initiation of complementary feeding was similar among the groups, with most of the mother initiating complementary feeding at 6 months of their children life (Figure 4.8).

Figure 4.8 Age of initiation of complementary feeding*



*Complementary feeding: the introduction of solid foods and liquids into the diet of the child, along with breastmilk or formula milk.
GM: Grandmother

4.7 Attitudes towards breastfeeding: The Iowa Infant Feeding Attitude Scale

4.7.1 General description

Mothers were asked to complete the IIF-AS to assess maternal attitudes toward various aspects of infant feeding. Given that 60% of the women from the grandmother group received advice from their mothers about the duration and/or importance of exclusive breastfeeding, I expected that that this group would show more favourable attitudes to breastfeeding.

Overall, the complete sample of women showed a high favourable attitude to breastfeeding (**Table 4.9**) with a mean score of 67.2 (± 7.6).

Table 4.9 Descriptive statistics of the scores of the IIF-AS

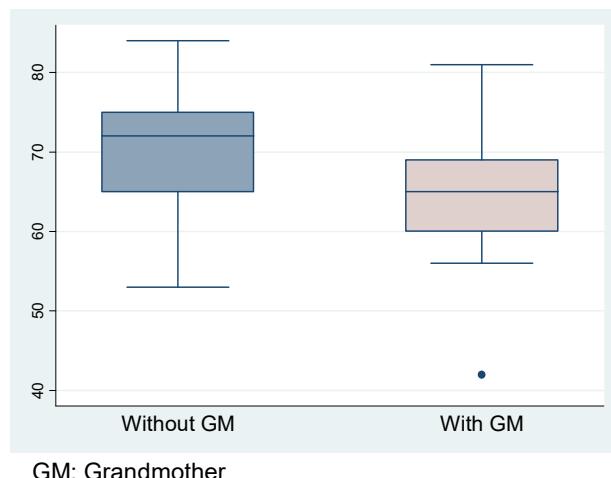
	N	mean	SD	median	IQR	Min	Max
IIF-AS	80	67.2	7.6	67	11	42	84

SD: standard deviation, IQR: interquartile range; Min: minimum; Max: maximum

4.7.2 Attitudes towards breastfeeding stratified by grandmaternal groups

Results by grandmaternal groups are shown in **Figure 4.9**. The median (IQR) scores from the group that received grandmaternal support were 65 (9) vs 72 (10) for the other group.

Figure 4.9 IIF-AS' scores by grandmaternal groups



Statistical differences were found among the groups ($z = 2.54$, $p=0.01$, Wilcoxon rank sum) with women from the no-grandmother group showing a more favourable attitude toward breastfeeding.

4.8 Discussion

183 mother-infant dyads were identified as potential participants through the information provided from the centres, but only 104 met the criteria and only 68 accepted to take part in the study. Through other recruitment methods I was able to increase the sample ($n=90$). In the current study, 58% of the mothers in the study sample indicated that they were receiving frequent support from their mothers as per my categorization criteria.

According to my review, there no studies in Mexico or Yucatan focusing on the role of the maternal grandmother in the care of their daughter and their grandchildren, even though they are the preferred substitute for childcare (Partidas, 2004) and tend to have more contact and a closer relationship with grandchildren than any other member of the mother's social network (León et al., 2016). As far as I know, my study is the first to provide data about grandmother's support in the Yucatecan population.

Accordingly to local standards, the studied families belonged to an overall advantaged socioeconomic group. Mothers and partners performed a paid job and showed relatively high levels of formal education. The advantaged socioeconomic conditions are also evident in their living conditions, with a high percentage of the participants having access to basic services (potable and drinking water, electricity and toilet access with sinkhole) and a high percentage of their homes constructed of durable materials (blocks, bricks, concrete, cement) resistant to water and humidity.

Regarding pregnancy-related variables, all women had access to health services during their pregnancy and none reported having smoked during pregnancy, while few of their partners smoked. Although 56% of women reported having infectious diseases during pregnancy, none of them reported having important implications.

Concerning the children, most showed adequate weight and length at birth with no statistical differences by sex. The overall prevalence of exclusive breastfeeding was low in the sample.

By grandmaternal group, differences were found only in attitudes to, and the actual duration of, breastfeeding. Counter-intuitively, women who did not have grandmaternal support showed higher scores of the IIF-AS and breastfed their children for longer.

I do not have information to ensure that women from the non-grandmother group did not receive advice from their mothers. It is possible that despite the distance (in those cases where the grandmother lived in another city) women still communicated with their mothers and received advice and knowledge from them. In addition, in those cases in which the grandmother was not available, due to having passed away or where the mother did not have a good relationship with her, it is possible that mothers benefited from the contact and support from other female relatives. Nonetheless, it is possible to consider that women without grandmaternal support have a higher disposition to intend to exclusively breastfeed their babies for the recommended time. There is evidence that shows that mothers that have the intention (highly motivated and confident) to exclusively breastfeed, had the highest IFFAS scores (Ishak et al., 2014).

Although mothers from the grandmother group showed a high positive attitude (median=65) to breastfeeding, considering that the maximum possible score is 85, is it interesting to analyse if the frequent presence of the grandmother could have an influence in their lower attitude about breastfeeding in comparison to the other group. I identified one study carried out in Brazil that found that non-daily contact with the maternal grandmother was a protective factor for maintaining breastfeeding until six months and decreased the risk of early supplementation with formula (Susin et al., 2005). Moreover, evidence has suggested that there are other barriers related to breastfeeding that need to be considered. For example, grandmothers could perceive breastfeeding as an obstacle to their daughters to getting adequate rest and as a barrier to bonding with their grandchildren (Grassley and Eschiti, 2008). Hence, early initiation of formula feeding could be seen by some grandmothers as a mean to increase the contact with their grandchildren.

Overall, advice and concerns of the grandmothers about breastfeeding may reflect cultural beliefs and practices that do not protect breastfeeding. Nevertheless, there is evidence about the positive influence of grandmothers towards breastfeeding in other populations (Mahoney and James, 2000). Hence, detailed information about each population is revealing variability in the grandmother's influence on breastfeeding.

In future studies in Mexico and Yucatan, it will be important to consider more data about maternal attitudes toward breastfeeding, such as their motivation and confidence levels in achieving a long duration of this activity, and about cultural beliefs and practices commonly used by grandmothers, as well as their attitudes and perceptions. Additionally, it would also be interesting to know if the effect of the grandmother depends on whether she breastfed, ie, if she had the experience.

4.9 Conclusions

Overall, this chapter suggests the existence of a broadly favourable environment for healthy growth and development of the children in my sample. Women who enrolled, as well as their partners, were better educated than the general Yucatecan population and showed a positive attitude towards breastfeeding. The socioeconomic background was similar among the two groups, which helped to verify if the differences found among the groups were related to the grandmother's support and/or other factors not associated with the socioeconomic conditions. In the next chapter, the results of the maternal social capital data are presented, comparing the grandmother groups.

Summary points

- I. The studied families belonged to an overall advantaged socioeconomic group, accordingly to local standards.
- II. The advantaged socioeconomic status is evident in the level of education and employment status of the families and in their living conditions, with a high percentage having access to basic services and their homes constructed of durable materials resistant to water and humidity.
- III. All women had access to health services during their pregnancies and none reported having smoked during pregnancy.
- IV. Most of the children showed adequate weight and length at birth.
- V. The study population had a positive attitude toward breastfeeding. However, women who did not have grandmaternal support showed higher scores of the IIF-AS and breastfed their children for longer.
- VI. Advice and concerns of the grandmothers about breastfeeding may reflect their experience, cultural beliefs, and practices that do not protect breastfeeding and early initiation of formula feeding could be seen by some grandmothers as a mean to increase the contact with their grandchildren.

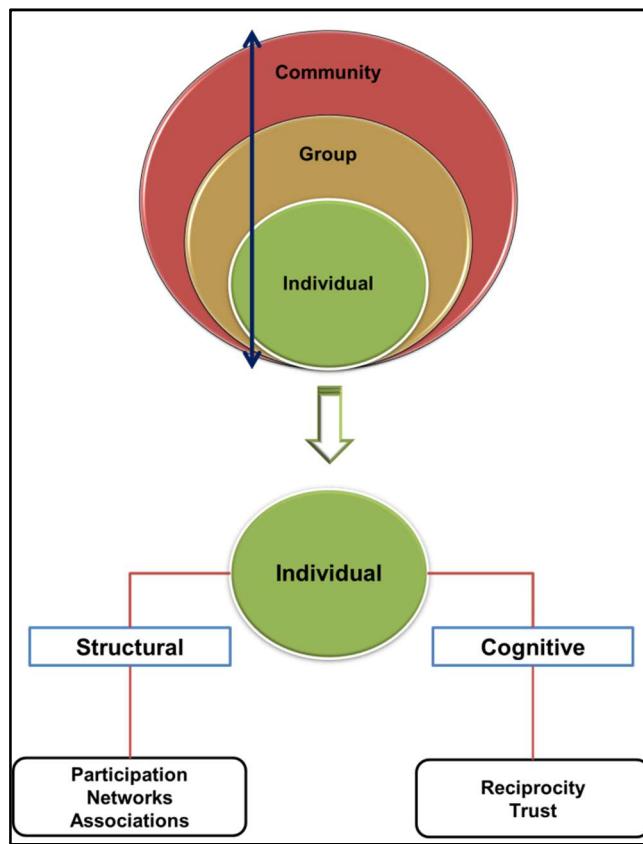
CHAPTER 5

MATERNAL SOCIAL CAPITAL

5.1 Introduction

This chapter provides a description of the social capital of the mothers. Social capital has been located at the community level, at the informal social group level and at the level of the individual (Coleman, 1988; Portes, 2000; Bankston III and Zhou, 2002). Therefore, social capital has an individual and an aggregate component, which means that it can be used by the group or individuals within the group and therefore, it can be considered that a dynamic component exists across the levels at which social capital exists (Adler and Kwon, 2002; Sander, 2002; Slangen et al., 2004) (Figure 5.1).

Figure 5.1 Levels and types of social capital



Compiled through literature review

In the current study, social capital was measured at the individual level considering that the decision to invest in social relationships is first made by individuals (Glaeser, 2001). At the individual level, social capital can be measured by asking an individual about the quantity and quality of own networks, for example by asking about their participation in networks (structural social capital) and about whether they are trusting (cognitive social capital) (Kemenade, 2003) (**Figure 5.1**).

The purpose was to identify the members of the mother's personal social network, which may become available to the mother as a result of the history of these relationships. I obtained data about the quantity and quality of the mother's social relationships (family, friends, neighbours and co-workers).

Because it was important to know if women invest in their social relationships, they were asked about the number of days and hours per month that they participate in activities related to these groups.

This chapter also presents data about the support provided by the maternal grandmother and the partner. All women were asked about the support provided by the partner (n=90) and only women from the grandmaternal group (n=52) were asked about the support provided by their mothers.

The reasons why I did not obtain data about grandmaternal support in the 'Without GM' group were several. First, 14% of the women reported that their mothers passed away before the child was born. Second, 22% of the grandmothers were not living in Merida at the time of the study. Third, although 63% of the women reported that their mothers were living in Merida, the grandmother helped, but below the threshold established in the criteria of the study and.

As seen in **Table 5.1**, some grandmothers were not providing frequent support because they were still performing a paid job or because they only were available to help on an emergency, such as when the child is sick. Thirty-two per cent of the mothers reported that another family member was the other main caregiver. This group included the aunts, sisters, sisters in law and mothers in law. Finally, 16% reported that only them and their partners were taking care of the child because they did not want to burden their mothers with childcare or because the grandmother was very sick and hence unable to take care of the child or because they did not have a good relationship with the grandmother.

Table 5.1 Grandmaternal support in the non-grandmother group

	Without GM support (n=38)
Passed away (n=5)	14%
Does not live in Merida (n=8)	22%
Live in Merida (n=25)	
Works	8%
Only in emergencies	8%
Other main caregiver	32%
Only parents take care of the child	16%

The main aims of this chapter were: a) to identify the members of the mother's social network and the support provided by them (in terms of quality and quantity) and b) to identify differences between the grandmaternal groups.

In **Figure 5.2** I present the way this chapter was organized. First, I provide a general description of the social capital data of the whole sample (n=90). This includes the number of relatives and friends with whom they maintain regular communication and a close relationship, and their participation in social networks and the quality of their relationships. I explained to them that when I was asking about 'regular communication', I was referring to people with whom they maintained some contact i.e. see them, talk to them, send or receive messages from them at least once a month. Regarding 'close relationships', I was asking about the number of people with whom they felt comfortable and could talk about their problems or personal issues.

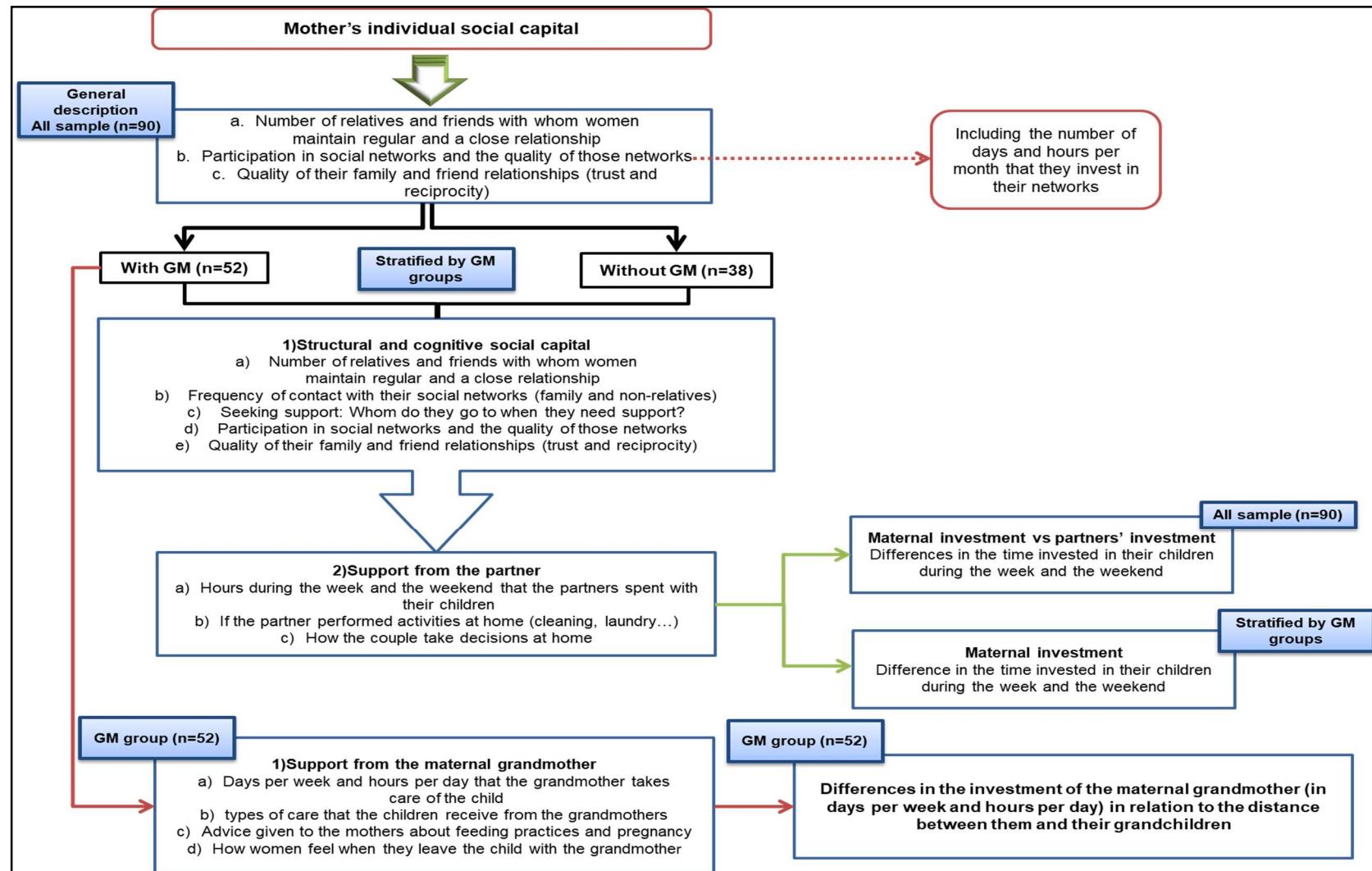
Then, the study sample was categorised in two groups: 'with GM support' and 'without GM support', and their results were compared. Differences were assessed between the groups in structural and cognitive capital. Moreover, data about the partner's support was compared across grandmaternal groups. Due to the partner's support section including information about paternal investment (number of hours that fathers invested in their children), I decided to include two subsections: 1) exploring the differences in investment among the parents (maternal vs paternal investment), and b) assessing differences in investment among the mothers stratified by grandmaternal groups.

Finally, the last section focused on grandmaternal support. Only women from the grandmaternal group (n=52) were asked about the support provided by their mothers. In addition, differences in grandmaternal investment (in days per week and hours per day) were assessed considering the distance (in km) between the mothers and grandmothers.

5.2 Statistical analysis

All participant women (n=90) completed the maternal social capital section of the interview. Some data was presented as frequency and or percentages, while univariate analyses (Chi-square/Fisher Exact test) were used to compare groups. Continuous data were tested first for normality distribution by using histograms, Q-Q plots and statistical tests, such as Kolmogorov-Smirnov test and Shapiro-Wilk test. For normally distributed data, mean \pm standard deviation (SD) was presented along with the statistical results (T-test) for group comparisons. In those cases in which a non-parametric test (Mann-Whitney) was used, the median \pm interquartile range (IQR) was presented.

Figure 5.2 Diagram of chapter structure



5.3 Social capital: general description of the data

Women were asked about the number of relatives and friends with whom they maintained regular communication and kept a close relationship (Table 5.2). Overall, women maintained regular contact with many relatives and friends. However, regarding close relationships, a large percentage of the women perceived having only one to four friends or relatives they could trust, mainly their mother who in 66% of the cases was the relative they felt closest to.

Table 5.2 Distribution of the number of relatives and friends with whom women maintain regular contact

Variable	Women
Relatives	
Regular contact (%)	n=89
1-4	35.0
5-8	37.0
≥ 9	28.0
Close relationship (%)	n=88
1-4	71.6
5-8	16.0
≥ 9	12.5
Relative with whom they maintain the closest relationship (%)	n=90
Mother	67.0
Other (stepmother, aunt, niece, husband, mother in law)	33.0
Friends	
Regular contact (%)	n=87
1-4	50.0
5-8	25.0
≥ 9	24.0
Close relationship (%)	n=87
1-4	81.6
5-8	16.0
≥ 9	2.3
If they consider having more female or male friends (%)	n=90
Female	67.0
Male	16.0
Without distinction	17.0

5.3.1 Structural social capital

I asked women about their own participation in social networks and the quality of those networks to explore their individual structural capital. Specifically, I asked the mothers if they were active members in groups in their community and if they received support from these networks. Only 13% of women stated being a member of a sports club. However, they clarified that they were members of gyms in which they did not participate in-group activities. A high percentage (92%) of women did not belong to any artistic association, such as literature, design, dance or performing arts, and only 10% were members of a religious association.

Sixteen per cent of women belonged to labour unions due to their jobs; however, they did not participate in activities organized by these unions. Only 11% of the women stated that their neighbourhoods had a neighbourhood association where, at least once per month, they met to talk and solve problems related to security and infrastructure of their neighbourhoods. Finally, a small percentage of women belonged to environmental (8%), political (15%) or other (10%) associations, such as volunteering in hospitals or social support programs and university associations. Participant mothers that were members of these associations reported that they did not receive support from these groups. However, this may be because they are not very active members. All the women invested less than two hours per month in activities related to some of these associations.

5.3.2 Cognitive social capital

Given the lack of belonging to social networks other than family and friends, cognitive social capital was measured by considering how the women perceived the quality of their family and friend relationships, for example if they are trustworthy. I read them some statements and they told me how much they agreed or disagreed with each of them (**Table 5.3**).

Levels of cognitive social capital were very high in this sample of women. The mother, partner, other relatives, such as siblings, aunts, fathers and mothers in law, or friends were the people the women trusted the most. Outside family and friend networks, most women felt they could not trust their neighbours and although they reported a high level of trust in their closest co-worker, this was due to most of them being friends outside the workplace.

In terms of reciprocity, 47% of the mothers agreed or strongly agreed that when someone's support or help is received during a special situation, people respond by returning the favour. Finally, 63% of the women from both groups agreed or strongly agreed that when they give their trust to someone, that person also returns that trust (**Table 5.3**).

Table 5.3 Maternal cognitive social capital

		Women (n=90)		
		Disagree	Indifferent	Agree
TRUST				
I trust my mother a lot (%)		2	4	94
I trust another relative a lot (%)		1	2	97
I trust my partner a lot (%)		1	1	97
I trust my closest friend a lot (%)		---	4	96
I trust my closest neighbour a lot (%)		61	12	27
I trust my closest co-worker a lot (%)		17	15	67
RECIPROCITY				
If I help someone, this person will help me when I need it (%)		31	21	47
If I trust someone, that person will also give me his or her trust (%)		17	20	63

5.4 Social capital by grandmaternal groups

5.4.1 Family

Figure 5.3 illustrates that the distribution of women, according to the number of relatives with whom they maintained regular contact, was similar among the groups ($\chi^2_{(2)} = 0.48$, $p=0.78$). Regarding the distribution according the number of relatives they maintained a close relationship, the distribution did not significantly vary between the groups ($\chi^2_{(2)} = 1.70$, $p=0.45$) (**Figure 5.4**).

Figure 5.3 Distribution of the number of relatives with whom women maintain regular contact stratified by grandmaternal groups

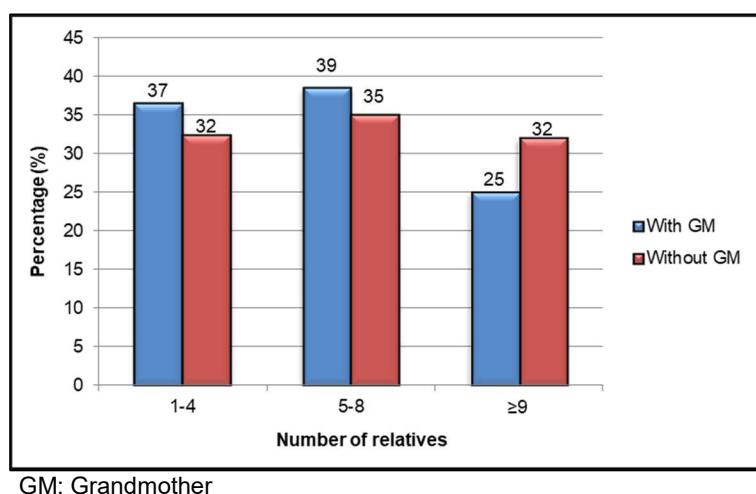
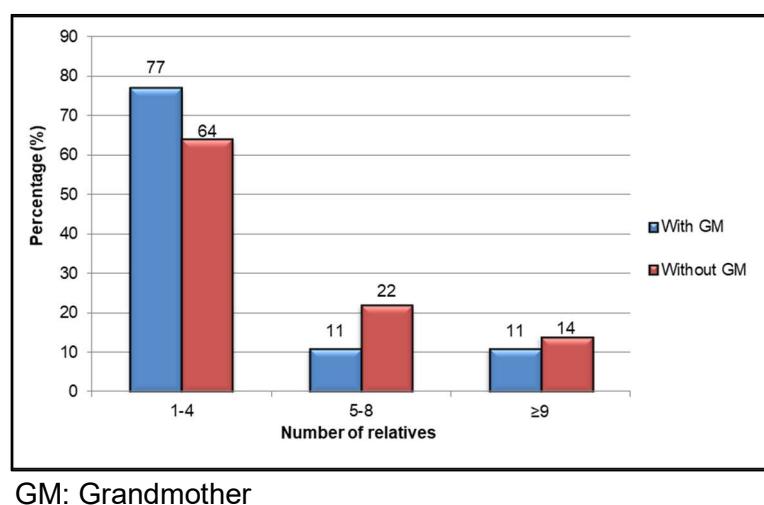


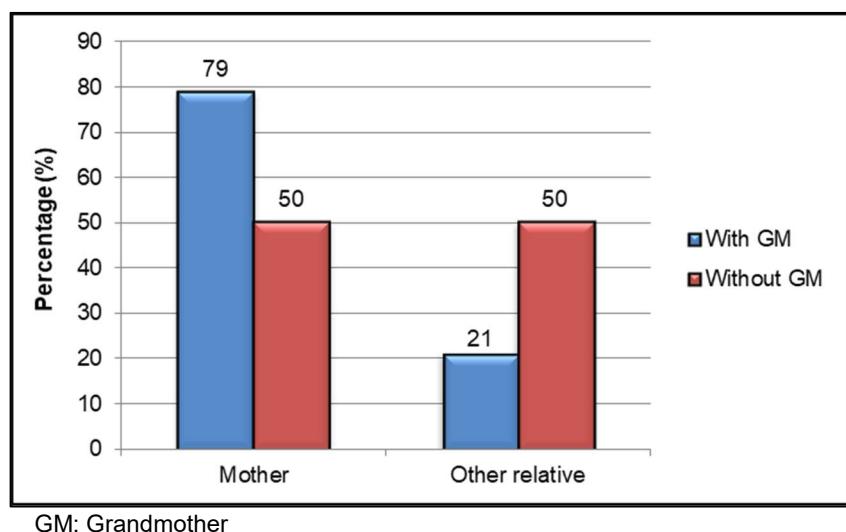
Figure 5.4 Distribution of the number of relatives with whom women maintain a close relationship stratified by grandmaternal groups



5.4.2 Family members with whom women feel the closest relationship

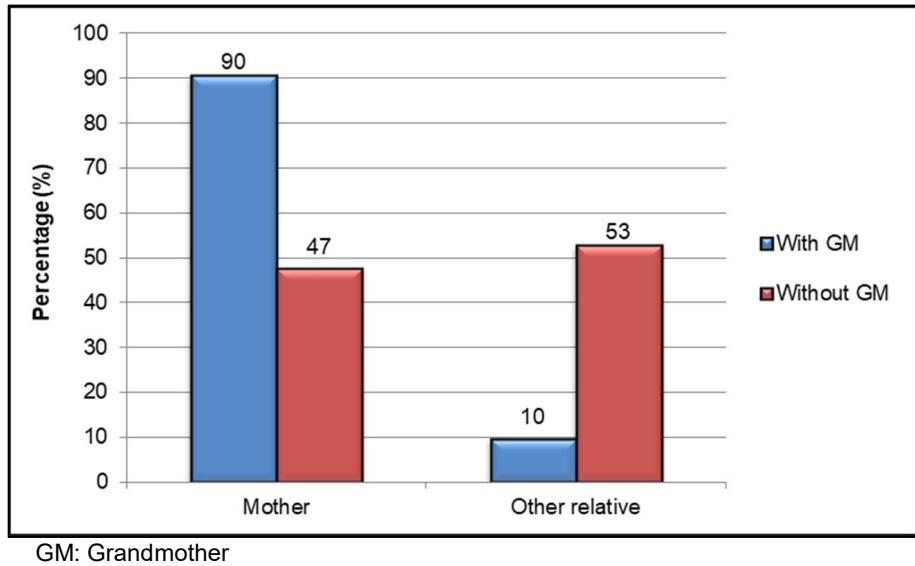
Women who had the support from the maternal grandmother felt closer to their mothers than women who did not have that support ($\chi^2_{(1)} = 8.22$, $p=0.004$) (Figure 5.5). However, those from the non-grandmother group who mentioned not feeling close to their mothers (50%), including those cases in which the grandmother had passed away, found this closeness with other relatives like aunts, cousins, mothers-in-law and stepmothers. This indicates that they were able to find other sources of support within the family.

Figure 5.5 Family members with whom women feel closest to stratified by
grandmaternal groups



As an example of this support found in other relatives, Figure 5.6 shows that 53% of women that did not have frequent contact and support from their mothers went to other relatives, such as female siblings, mother-in-law and sister-in-law, when they needed advice on childcare.

Figure 5.6 Family members that women ask for advice about childcare



5.4.3 Friends

When it comes to the number of friends with whom they kept in regular communication (Figure 5.7) and had a close relationship (Figure 5.8), the two study groups showed similar distributions, without significant differences ($\chi^2_{(2)} = 1.34$, $p=0.51$, and $\chi^2_{(1)} = 0.02$, $p=0.89$, respectively).

Figure 5.7 Distribution of the number of friends with whom women maintain regular contact stratified by grandmaternal groups

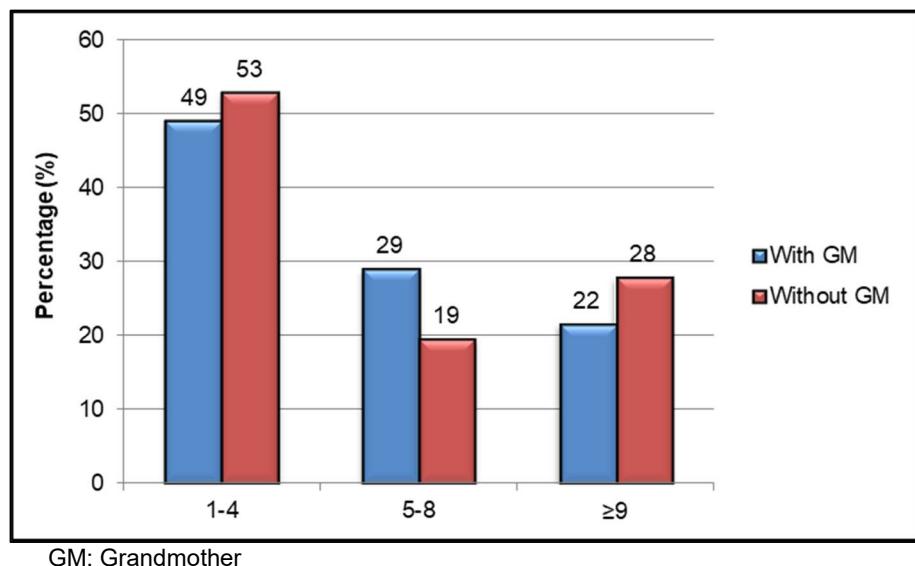
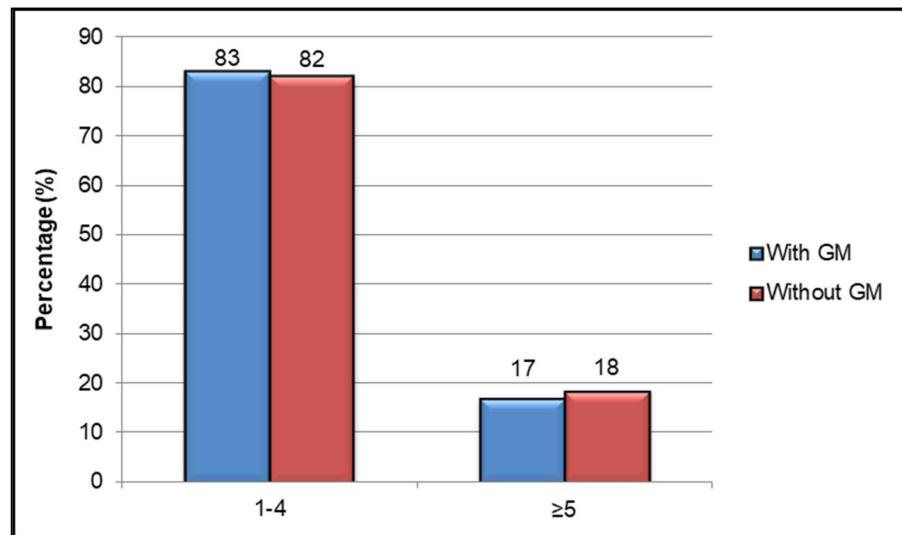


Figure 5.8 Distribution of the number of friends with whom women maintain a close relationship stratified by grandmaternal groups



GM: Grandmother

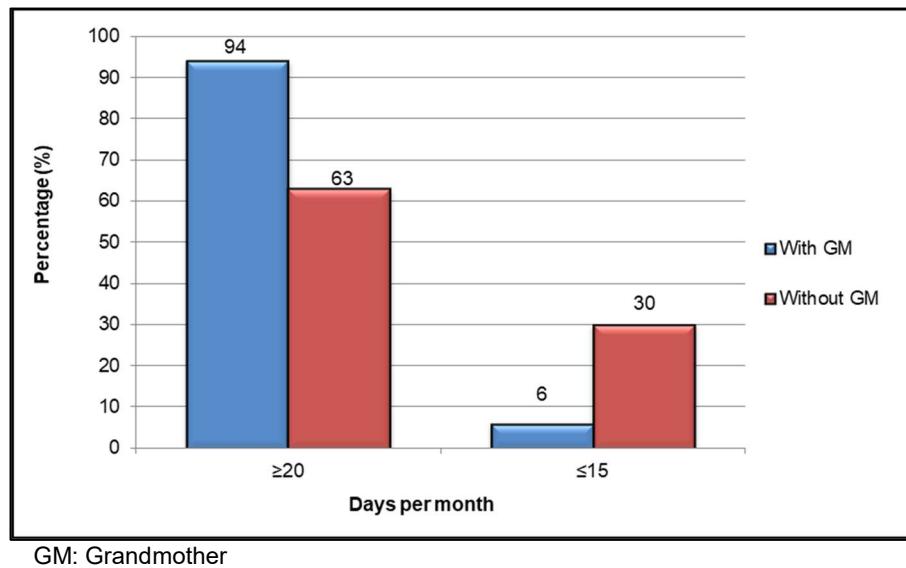
5.4.4 Frequency of contact with social networks

One important issue to consider is the frequency of contact that the participant women had with their close family members, including her mother, and with non-relatives.

5.4.4.1 Maternal grandmother

Women were asked how many days per month they talked with their mothers about personal issues not related to motherhood. As shown in **Figure 5.9**, 94% of women from the grandmother group talked with their mothers at least 20 days per month. From the other group, I excluded 21% of cases in which the grandmother had passed away. Only 7% of the participants stated that they never talked with their mothers, while 63% of the women communicated with their mothers least 20 days per month. Women from the grandmother group talked significantly more ($\chi^2_{(1)} = 12.83$, $p < 0.01$) with their mothers about personal issues.

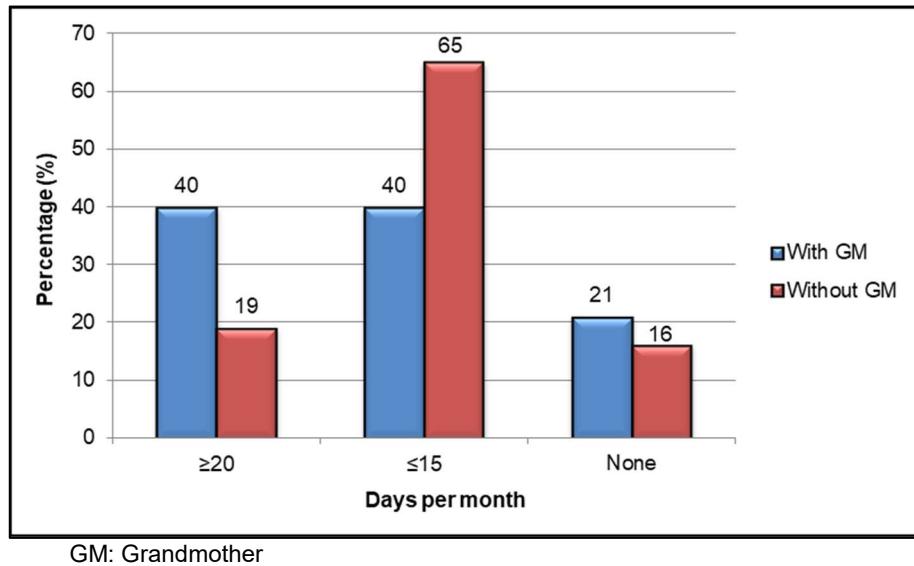
Figure 5.9 Days per month that women talk with their mothers about personal issues stratified by grandmaternal groups



5.4.4.2 Maternal grandfather and siblings

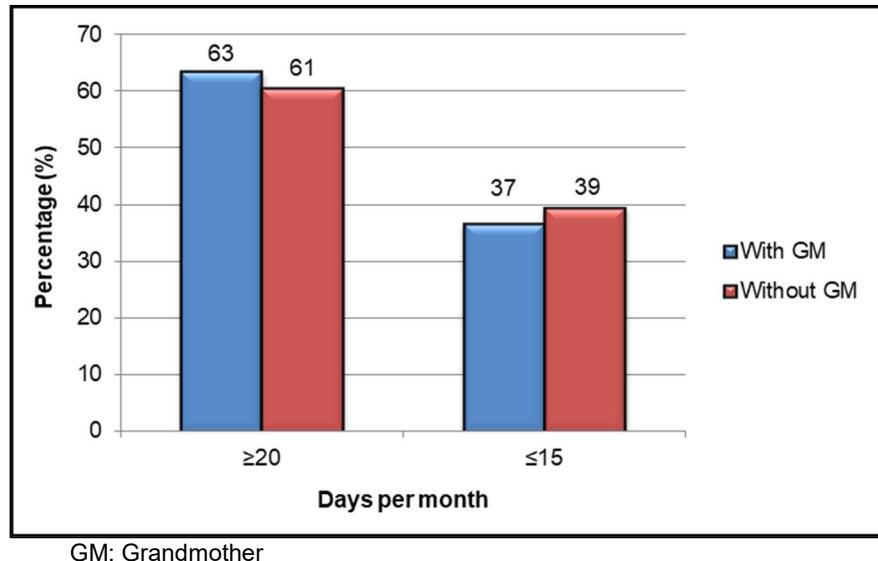
Figure 5.10 shows the women's frequency of communication with their fathers. I excluded 16% of the cases in which the father had passed away. In comparison to Figure 5.9, women had less contact with their fathers. Only 19% of the women from the no-grandmother group and 40% of women from the grandmother group stated they had frequent communication with their fathers (≥ 20 days). Although this difference was not significant ($\chi^2_{(2)} = 5.03$, $p=0.08$), probably the presence of the grandmother could be related to a higher frequent contact between daughters and fathers.

Figure 5.10 Days per month that women talk with their fathers about personal issues stratified by grandmaternal groups



Both participating groups had similar frequency of communication with their siblings (Figure 5.11) ($\chi^2_{(1)} = 0.08$, $p=0.77$).

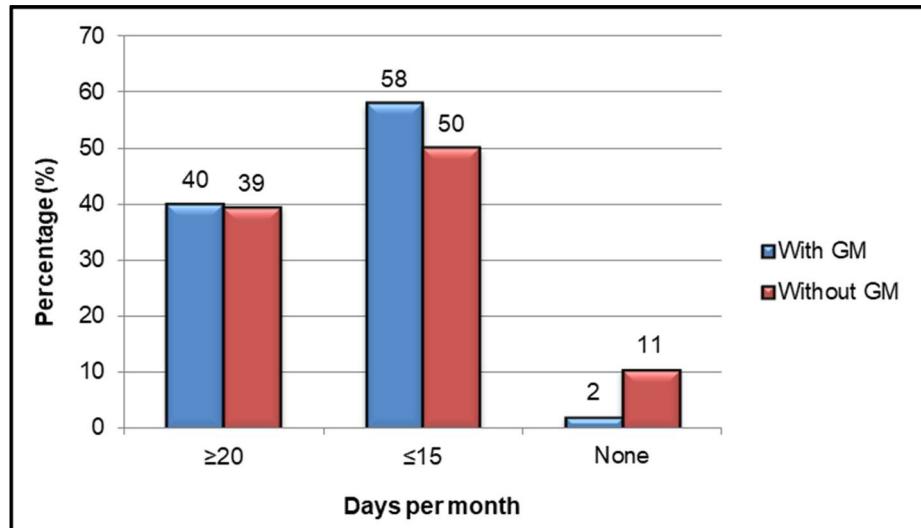
Figure 5.11 Days per month that women talk with their siblings about personal issues stratified by grandmaternal groups



5.4.4.3 Non-relatives

On the other hand, when I analysed the frequency of contact with non-relatives, such as friends (Figure 5.12) and neighbours (Figure 5.13), the distributions were very different to those related to their family members. Women had less contact with friends and neighbours, regardless of the group they belong to.

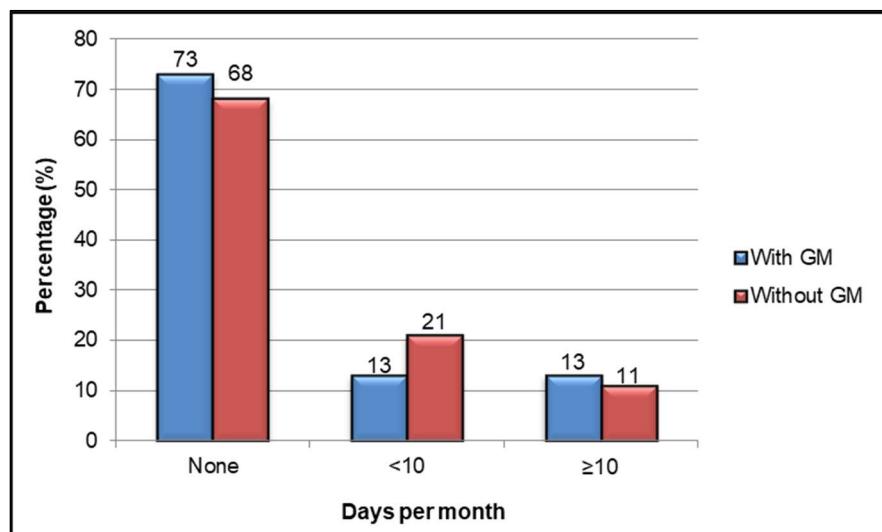
Figure 5.12 Days per month that women talk with their friends about personal issues stratified by grandmaternal groups*



GM: Grandmother

*No statistical differences among the groups: Fisher's exact: $p = 0.24$

Figure 5.13 Days per month that women talk with their neighbours about personal issues stratified by grandmaternal groups*

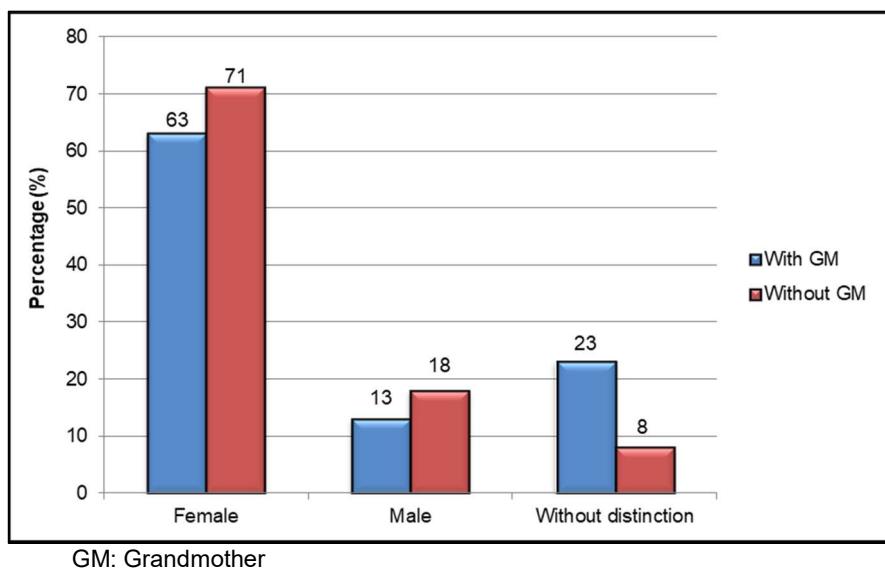


GM: Grandmother

*No statistical differences among the groups: Fisher's exact: $p = 0.67$

Most of the women reported having more female than male friends; however, a higher percentage of women from the grandmother group stated having equal numbers of female and male friends (**Figure 5.14**). Although this result cannot be generalized and the difference among the groups does not reach statistical significance ($\chi^2_{(2)} = 3.57$, $p=0.17$), it is possible that having regular contact with the grandmother can make a difference at the time of establishing social relationships and seeking support. Women who have access to their mothers and feel a close relationship with them could perceive them as their main support during stressful events.

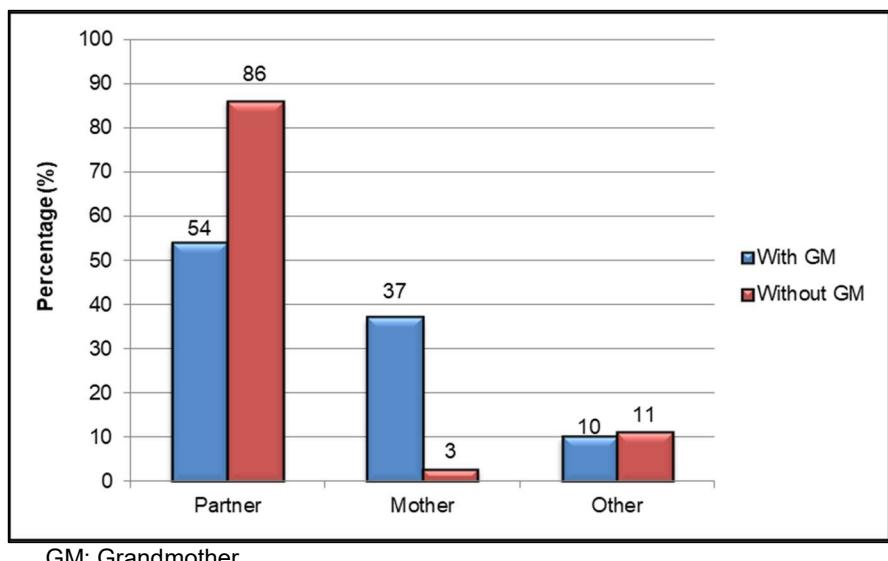
Figure 5.14 Distribution of female and male friends stratified by grandmaternal groups



5.4.5 Seeking support: Whom do they go to when they need support?

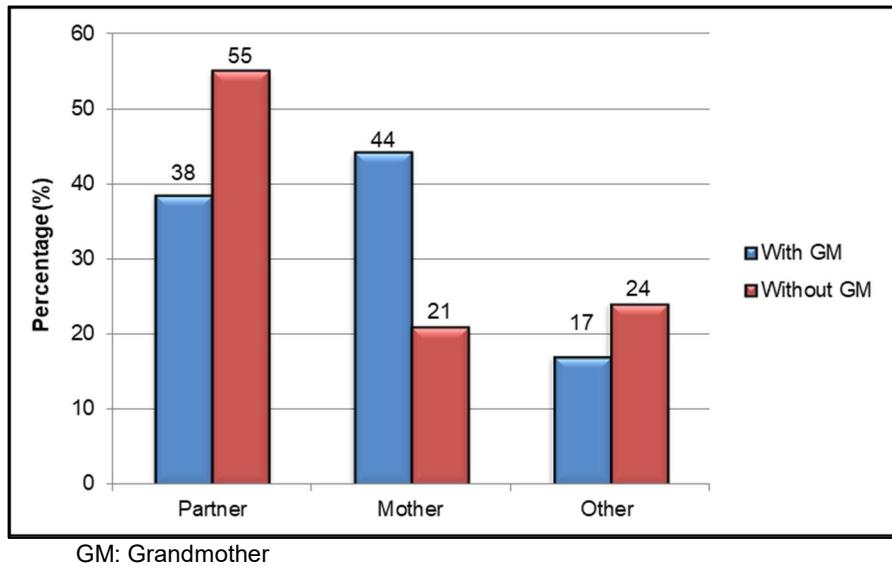
I read the mothers a series of hypothetical situations or problems that they might face and asked them who were the people they would ask for support to solve those problems. First, they told me the person they could rely on if they experienced a problem at their work (Figure 5.15). Most of the women without the support of the grandmother relied on their partners while mothers from the other group relied on the partners and the grandmothers (Fisher's exact: $p < 0.01$).

Figure 5.15 Person that women ask for support when they have a work issue stratified by grandmaternal groups



The same pattern was found when I analysed answers in relation to the person the women would ask for help if they have financial problems (Figure 5.16). Women that did not have the support of their mothers relied on their partners and other relatives, such as female siblings, mothers-in-law, and non-relatives such as friends, although this difference among the groups did not quite reach significance ($\chi^2_{(2)} = 5.23$, $p=0.07$).

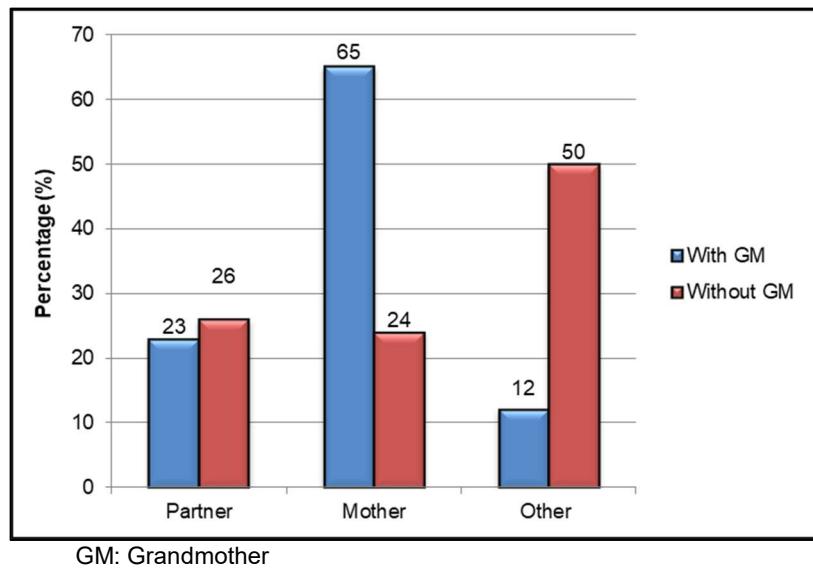
Figure 5.16 Person that women ask for support when they have financial problems stratified by grandmaternal groups



In comparison with **Figures 5.15 and 5.16**, women from the grandmother group felt that they could rely more on their mothers when they need help at home (for cleaning, cooking or taking care of the child) if they experienced a health problem (**Figure 5.17**). In contrast, women from the other group used different sources of support, including the partner, the mother, the mother in law or other relatives such as female siblings, sisters in law, aunts or female cousins ($\chi^2_{(2)} = 19.77$, $p=0.07$).

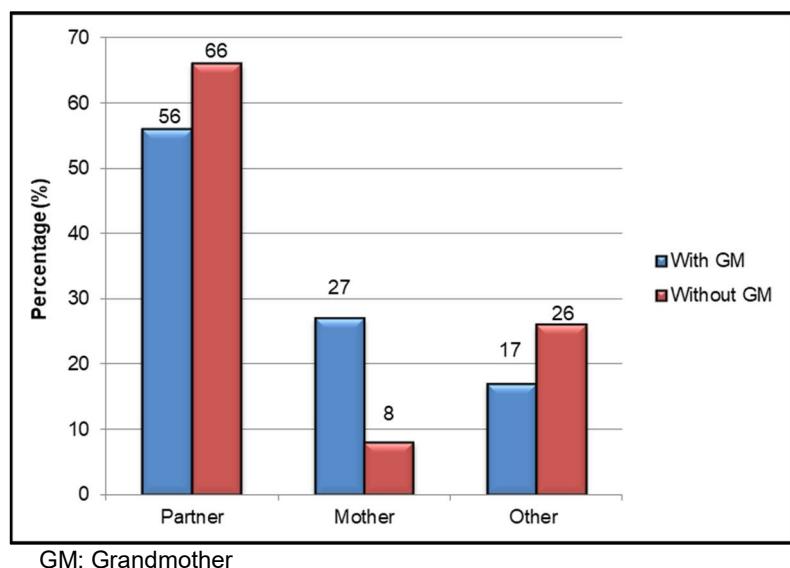
It is important to consider that this result was expected considering that the recruitment was conducted based on this kind of support. However, the difference between the groups is relevant since women from the non-grandmother group do not have this kind of support on a regular basis. Women without the support from their mothers pointed out that they received this kind of help when the situation is categorized as an emergency and the help provided comes from the female relative that is available at that moment.

Figure 5.17 Person that women ask for support when they needed help at home



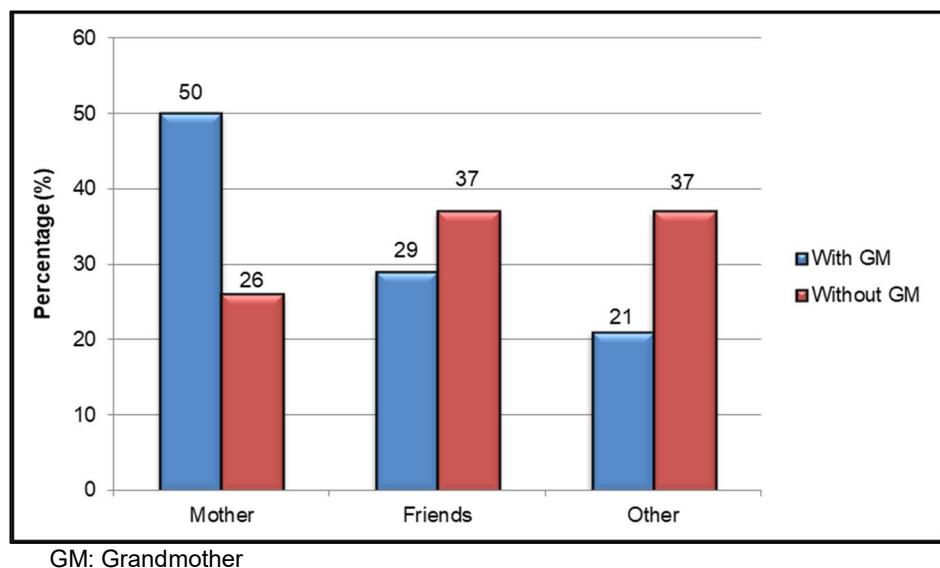
If women experienced a personal problem such as the loss of a close friend or relative, most of them relied on their partners for support. However, women from the grandmother group also asked for support from their mothers, whereas women from the other group sought support from friends, female siblings, aunts and female cousins ($\chi^2_{(2)} = 5.42$, $p=0.06$) (Figure 5.18).

Figure 5.18 Person that women ask for support when they have personal problems stratified by grandmaternal groups



Finally, the last hypothetical situation was related to the person they would ask for support if they had a problem with their partners. Fifty per cent of the women in the grandmother group relied on their mothers and 29% on friends. Meanwhile, women without support from their mothers relied more on friends and other people, such as siblings, the mother in law or the sister-in-law (Figure 5.19) ($\chi^2_{(2)} = 5.23$, $p=0.07$). It is clear that if women have access to their mothers, they represent an important source of support.

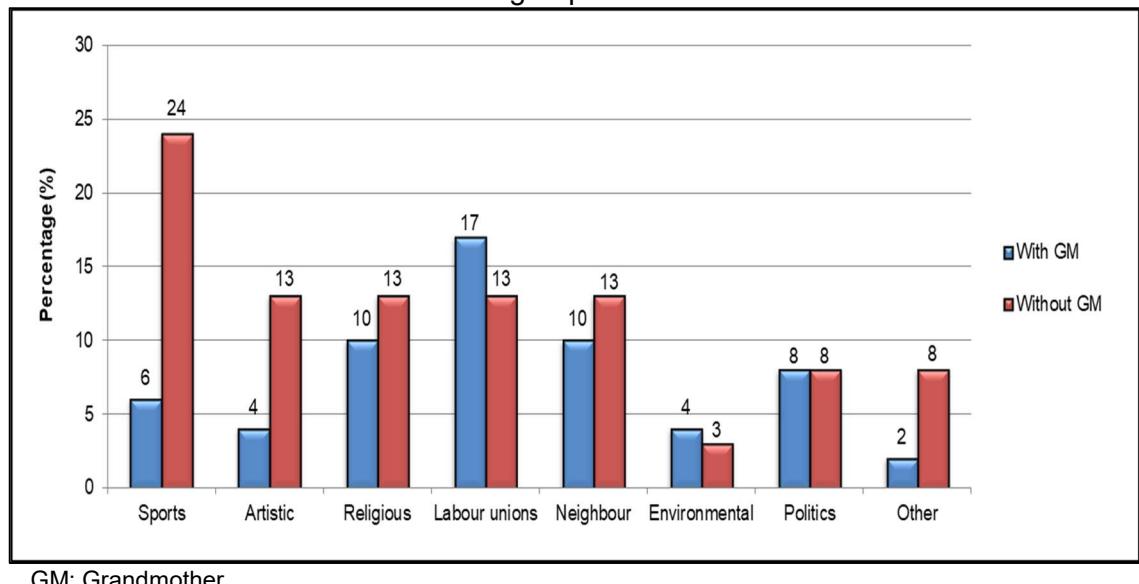
Figure 5.19 Person that women ask for support when they have a problem with the partner stratified by grandmaternal groups



5.4.6 Structural social capital

Levels of structural social capital were low between both groups (Figure 5.20). However, among those who belonged to a social group (i.e. affiliation to and/or participation in social groups, such as religious or neighbour associations), a higher percentage of women from the non-grandmother group were members of sport, artistic and other social groups. Nevertheless, as I stated before (see section 5.3.1), women reported not receiving support from these groups and this may be because they are not active members (women invested less than two hours per month in activities related to some of these associations).

Figure 5.20 Women's participation in social networks stratified by grandmaternal groups



Considering all the data presented above, it seems that women from both groups invest more in their relationships with family and, relatively, in friends than in other social groups.

5.4.7 Cognitive social capital

Levels of cognitive social capital related to family and close friends were very high between both groups (**Table 5.4**). The mother, partner, other relatives, such as siblings, aunts, fathers and mothers in law, and friends were the people women trusted the most. One important point to consider is that although the mothers reported that their mother was the person they could trust the most (**Table 5.4**), only 83% of women from the grandmother group and 29% of women from the other group reported that the grandmother was the most reliable person for taking care of the child. Probably when it comes to childcare, differences in parenting methods could be causing this discrepancy. For example, some of the mothers reported that one reason not to trust their mothers was the lack of boundaries with her child, i.e. they felt that the grandmother was spoiling the child.

In terms of reciprocity, 49% and 44% of the mothers from the grandmother and non-grandmother groups, respectively, agreed that when someone's support or help is received during a special situation, people should respond by returning the favour. Finally, around 60% of the women from both groups agreed that when they gave their trust to someone, that person should return their trust.

Table 5.4 Maternal cognitive social capital, stratified by grandmaternal groups

	With GM (n=52)			Without GM (n=38)		
	Disagree	Indifferent	Agree	Disagree	Indifferent	Agree
TRUST						
I trust my mother a lot (%) [*]	---	6	94	6	---	93
I trust another relative a lot (%) **	---	2	98	3	---	97
I trust my partner a lot (%)	5	---	95	---	3	97
I trust my closest friend a lot (%)	---	4	96	---	3	97
I trust my closest neighbour a lot (%)	64	11	25	58	13	29
I trust my closest co-worker a lot (%)	12	20	67	25	6	69
RECIPROCITY						
If I help someone, this person will help me when I need it (%)	29	21	49	34	21	44
If I trust someone, that person will also give me his or her trust (%)	16	21	63	20	18	62

GM: Grandmother

*I excluded five cases in which the grandmother passed away

**I asked them who their closest relative was after their mothers and partners.

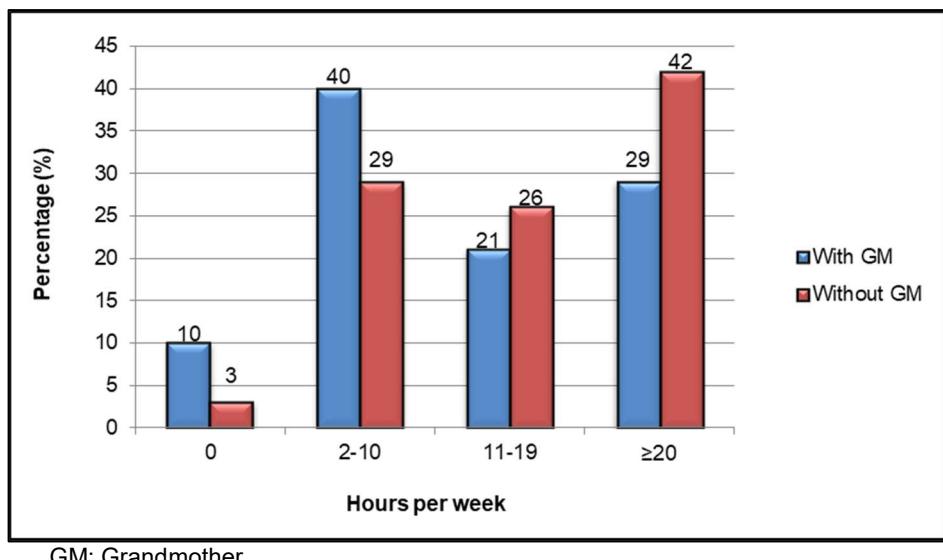
5.4.8 Support from the partner

A key member of the mother's social network is her partner. Cooperation between partners is particularly relevant for emotional and economic support and childcare assistance. In this sense, it was important to obtain information about partner support patterns. As stated in [Chapter 4](#), all the women were married to or living with the biological father of the studied child. This criterion was selected because biological fathers are known to invest more in their children than stepfathers in their partners' children (Salmon and Malcolm, 2011), and I wanted to hold this factor constant in my study.

To evaluate paternal investment, I obtained information about the total hours during the week (Monday-Friday, Saturday and Sunday) that the partners spent with their children. I specifically asked about the time they spent together when the child was not sleeping. Moreover, I obtained information about the activities at home in which the fathers played an active role, considering also activities related to childcare.

First, I asked the mothers the hours per week (Monday to Friday) that their partners invested in their children and found that the partners of the women that did not receive support from the grandmother were more likely to invest more time taking care of their children (media=15, IQR=25 vs median=15, IQR=20) during the week, although this difference was not statistically significant ($z = 1.78$, $p=0.08$, Wilcoxon rank sum) (**Figure 5.21**).

Figure 5.21 Hours per week that fathers invest in their children



On Saturdays, the pattern was similar to that observed during the week (Figure 5.22). Those partners that did not receive support from the mother-in-law (maternal grandmother) were more likely to invest more time taking care of their children (media=9.5, IQR=7 vs median=7.5, IQR=7; $z = 0.60$, $p=0.56$, Wilcoxon rank sum). However, on Sunday, partners that received support from the mother-in-law increased their own time investment (media=11, IQR=4.5), which was similar to the other group's investment (median=10, IQR=6) (Figure 5.23).

Figure 5.22 Hours on Saturday that fathers invest in their children

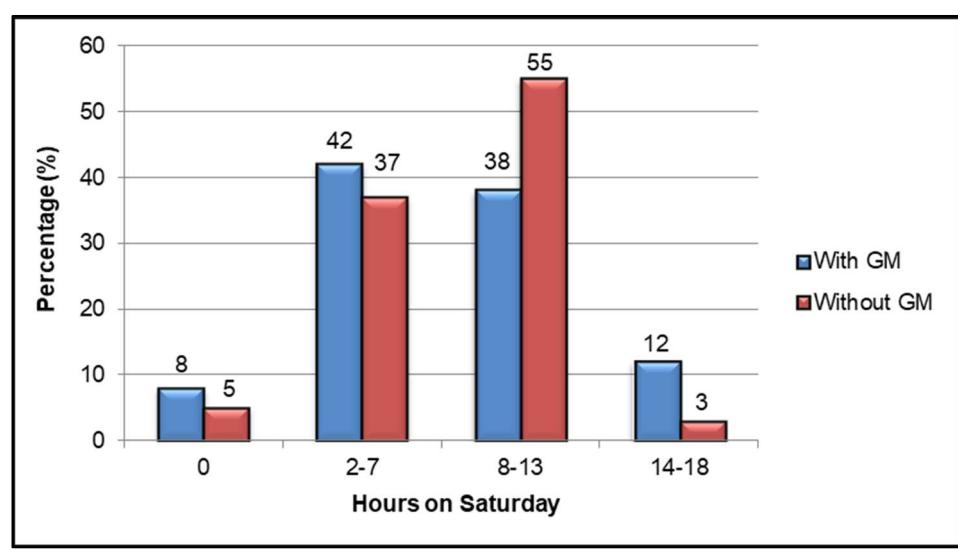
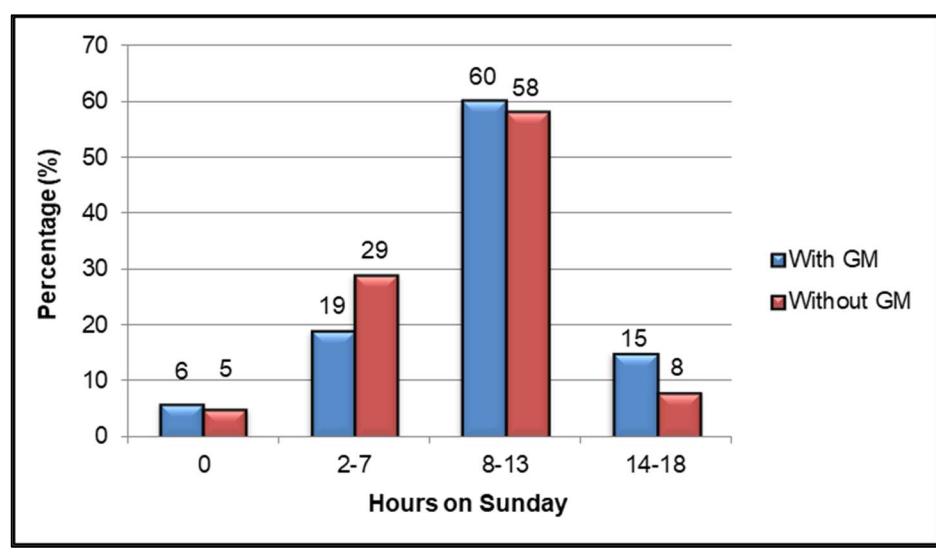
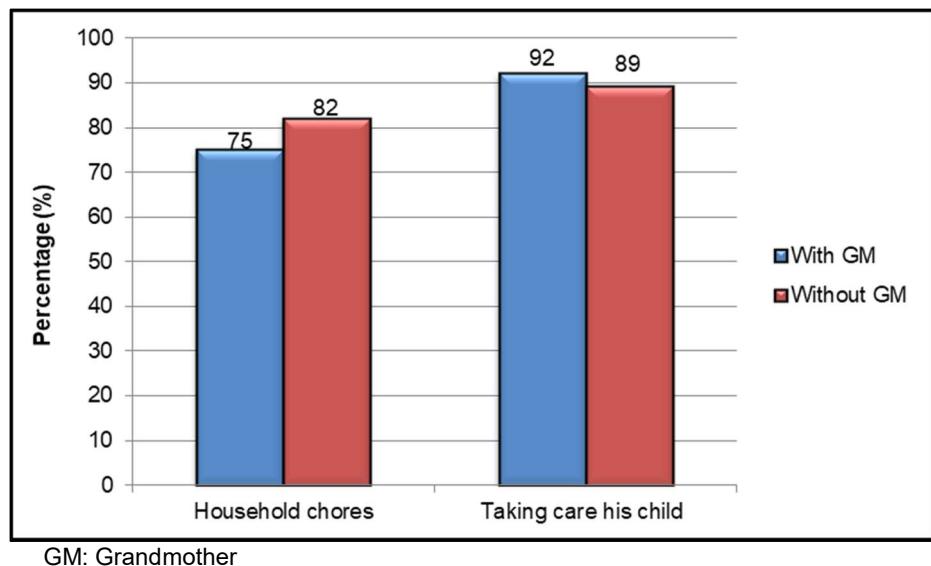


Figure 5.23 Hours on Sunday that fathers invest in their children



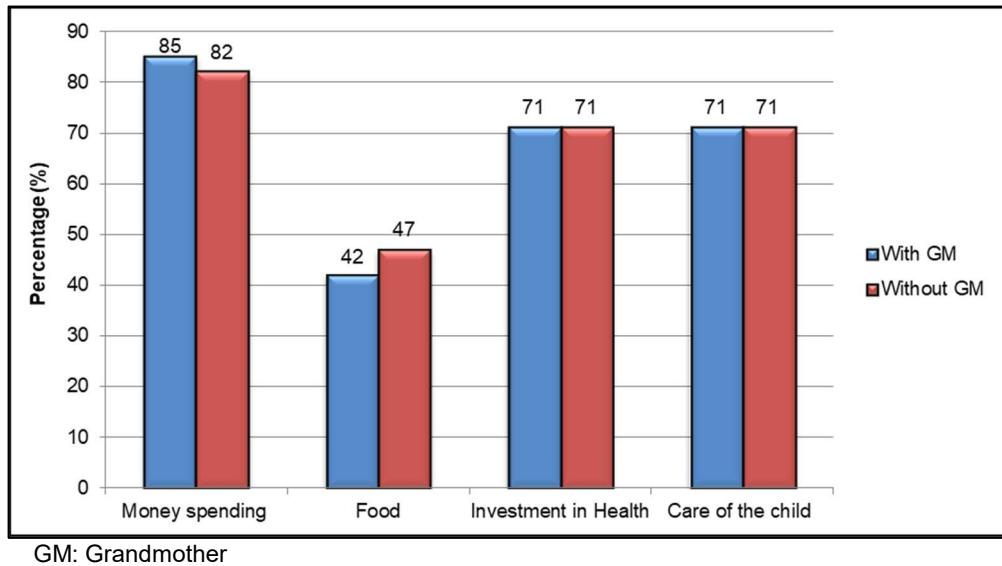
Most of the partners performed activities at home, such as cleaning, cooking, laundry, as well as activities related to the care of their children, such as giving them a bath and feeding them (Figure 5.24). However, although the partners were involved in activities related to their children, 63% of the women from both groups reported that they did not feel that the partner is the most reliable person for taking care of the child.

Figure 5.24 Activities in which the partner supports at home stratified by grandmaternal groups



Moreover, I asked mothers how they took decisions at home related to feeding practices and child healthcare, for example when the child got sick. The responses were similar between the two groups (Figure 5.25). When it came to decisions about how to spend household income, most of the women took decisions with the partner. The same applied when they had to make financial decisions about healthcare, such as paying for medical bills, and decisions about how to take care of the child. However, regarding decisions about the type of food consumed at home, partners were less involved.

Figure 5.25 Decisions that the mother and partner make at home stratified by grandmaternal groups



Women were involved in all decisions related to the home and when they did not have the support of the partner for decision-making, other relatives such as their mother; mother-in-law, aunts and/or both parents supported them.

5.4.8.1 Maternal investment vs partners' investment

In comparison with the partner, women invested more time during the week in taking care of their children. According to women, their partners invest between zero to 40 hrs per week (median=15 hrs per week, IQR=10) while they invested between four to 77 hrs per week (median=30 hrs per week, IQR=20). This difference was statistically significant ($z = 5.97$, $p<0.01$, Wilcoxon rank sum).

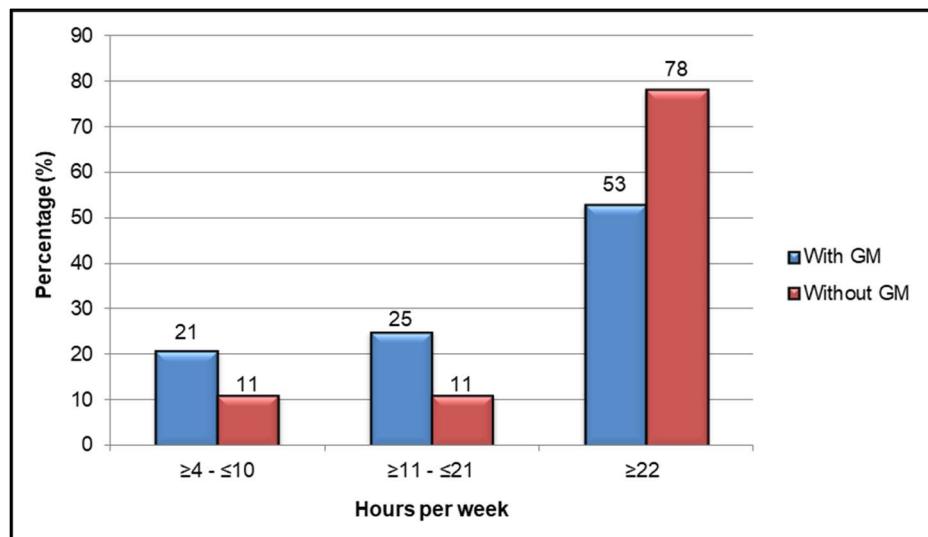
Differences were also found in parental investment during the weekends (Saturday: $z = 5.21$, $p<0.01$, Wilcoxon rank sum; Sunday: $z = 3.35$, $p<0.01$, Wilcoxon rank sum), mothers (median=12 hrs on Saturday, IQR: two; median=12 hrs on Sunday, IQR=3) invest more than fathers (median= 8 hrs Saturday, IQR=7; median=11 hrs on Sunday, IQR=5).

5.4.8.2 Maternal investment by grandmaternal groups

Although overall women invested more than their partners in taking care of their children, it is possible that women that had support from their mothers tended to invest less than women that did not have this support. During the week (Monday to Friday)

women from the no-grandmother group invested more time in taking care of their children ($\chi^2_{(2)} = 5.68$, $p=0.058$) (Figure 5.26). During the weekends, no differences were found by grandmaternal groups (Figures 5.27 and 5.28), all women invested similar amount of time.

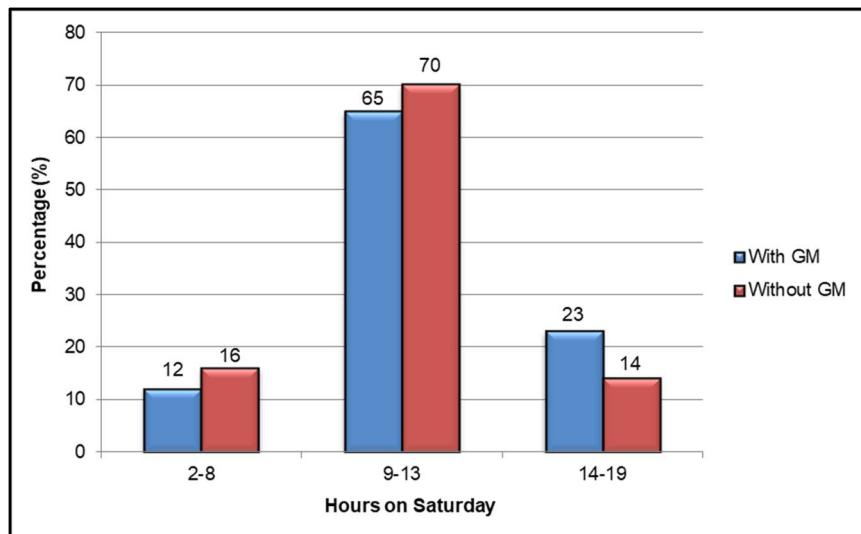
Figure 5.26 Hours per week that mothers invest in their children stratified by grandmaternal groups*



GM: Grandmother

$*(\chi^2_{(2)} = 5.68, p=0.058)$

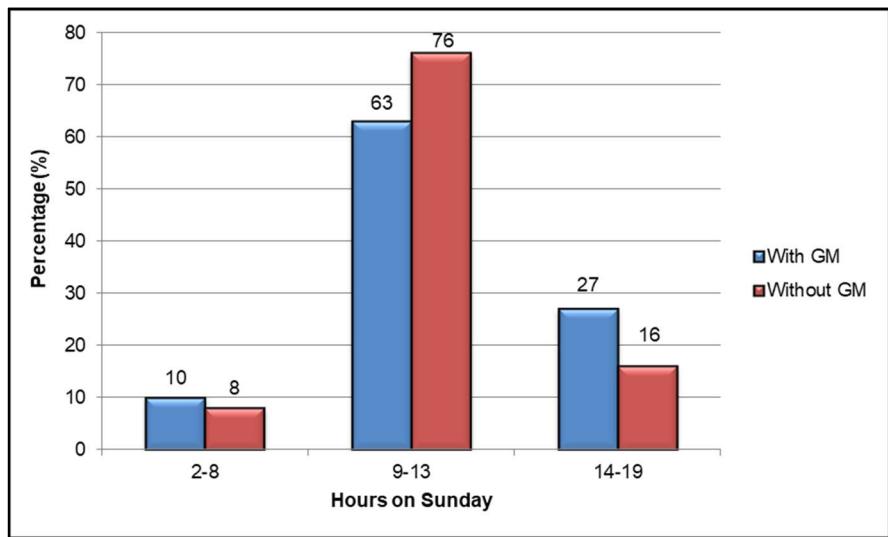
Figure 5.27 Hours on Saturday that mothers invested in their children stratified by grandmaternal groups*



GM: Grandmother;

$*\text{No statistical differences among the groups: } \chi^2_{(2)} = 1.50, p=0.48$

Figure 5.28 Hours on Sunday that mothers invested in their children stratified by grandmaternal groups*



GM: Grandmother

*No statistical differences among the groups: Fisher's exact: $p = 0.46$

5.4.9 Support from the maternal grandmother

As part of the maternal social network, grandmothers have a central role as an advisor to younger mothers and as a caregiver for children. In this sense, women from the grandmother group were asked about the types of support they received from their mother. First, I obtained information about the days per week and hours per day (**Table 5.5**) that the grandmothers took care of the child and the types of care that the children received from the grandmother.

Most of the participants reported that they asked their mothers to take care of the child during the week, between two to five days, due to their working commitments. The hours per day varied according to their work activities, but most of the mothers stated that their child spent between three to six hours with the grandmother. The types of care that the children received from the grandmothers were diverse and included spending time playing, feeding, bathing and taking them to the doctor if necessary.

Then, women were asked about the types of advice that they received from their mothers during their pregnancy. As shown in **Table 5.5**, not all women received advice from their mothers.

Table 5.5 Days per week and hours per day that the grandmother takes care of the child and grandmaternal advice given to the mothers

Variable	Women (n=52)
Days per week that her mother took care of their children (%)	
2-3	48
4-5	40
6-7	12
Hours per day (%)	(n=52)
3-6	79
7-10	15
11-12	6
Advice about weight gain during pregnancy (%)	(n=23)
9 kg	48
≥12 kg	17
Not specified	35
Advice about exclusive breastfeeding (months) (%)	(n=31)
Six	42
12	6
Not specified	52
Advice about weaning age (months) (%)	(n=25)
6-8	36
12	28
≥24	8
Not specified	28

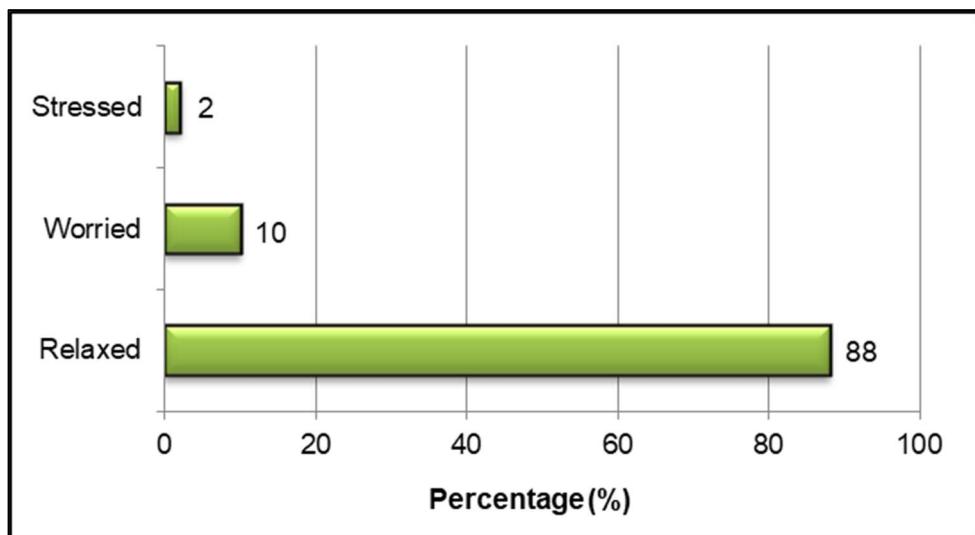
Regarding weight gain during pregnancy, most (48%) of the grandmothers recommended that one kilogram per month of the pregnancy was ideal, while 35% advised that gaining weight during pregnancy was healthy but did not specify how much weight. After the birth of the child, women received advice related to exclusive breastfeeding, weaning age and age of offering the first food, other than milk. Forty-two per cent of grandmothers advised their daughters to exclusively breastfeed their child for the first six months of life and 52% talked with their daughters about the importance of exclusive breastfeeding without specifying the time they should invest in breastfeeding.

Regarding weaning age, 36% of participants reported that their mothers told them that the weaning age should be between six and eight months and 28%, at 12 months. Twenty-eight per cent of the mothers reported that their mothers told them that breastfeeding should last until 'they stopped producing milk'. Due to the lack of specificity of this advice, I asked the grandmothers how they explained to their daughters how they could know that they were not producing milk. They considered the quality of the infant's cry and the colour of the breast milk. If after breastfeeding the baby was still crying, that meant that the child was still hungry and probably the mother did not have enough milk. On the other hand, if the breast milk had a 'pale colour' (like diluted milk), that meant that the mother was not producing good quality milk.

Regarding the first food they should offer to the child, most of the grandmothers told their daughters that it had to be vegetables. The remaining grandmothers advised fruits or other foods such as cereals or legumes.

Finally, I asked the women how they felt when they left the child with their mother (Figure 5.29). Most of the mothers reported feeling relaxed.

Figure 5.29 How women feel when they leave the child with the grandmother



5.4.9.1 Support of the grandmother during *la cuarentena*

La cuarentena (quarantine) is a term commonly used in Mexico to refer to the 40 days of recovery after the birth of a child. This period could be critical for women considering that they become involved in a learning process about childcare as well as specific beliefs and fears about recovery (Waugh, 2011).

During *la cuarentena*, women have expectations about social support and those who experience negative support are at a higher risk for experiencing postpartum depression (Martinez-Schallmoser et al., 2005). According to Clark (2001), the family could represent an important source of instrumental and emotional support during the postpartum period.

Therefore, I was interested to know if the women spent time in the grandmother's home or if the grandmother spent time in the mother's home during the postpartum period. According to the data, 29% of the women were living with their mothers at the time of birth and were still living together. From the remaining percentage, 18% spent less than a month with their mothers, 24% between one to two months, 18% between three to four and 11% equal to or greater than 7 months.

Women reported that during the postpartum period, their mothers had helped them in diverse activities related to the child such as bathing, cleaning and feeding them. In addition, grandmothers cooked for their daughters and the partners and took care of the child so that their daughters could sleep.

5.4.9.2 Differences in the investment of the maternal grandmother in relation to the distance between them and their grandchildren

In my study, I did not obtain information about how often, during a specific period of time, grandmothers and infants saw each other. However, the days per week that grandmothers took care of their grandchildren could be considered as a *proxy* of frequent contact between them. To measure geographical distance, I obtained the mothers' and grandmothers' addresses and calculated the GPS coordinates and the distance in km between the two homes using Google Maps. Then, I obtained the satellite image from OGC Web Map Service version 1.3 data provider (Map data © 2015 Google. <https://www.google.at/permissions/geoquidelines/attr-guide.html>; Retrieved: 11/19/2018). **Figure 5.30** shows the distribution of the mothers and grandmothers around Merida City. The sample (n=52) was divided into four groups according to the distance (**Table 5.6**).

According to the Urban Development Program, Merida is divided into eight districts (D-I to D-VIII) (Fuentes, 2005) (**Figure 5.30a**). Overall, mothers and grandmothers were distributed among the D-II to D-IV and D-VI and D-VII districts.

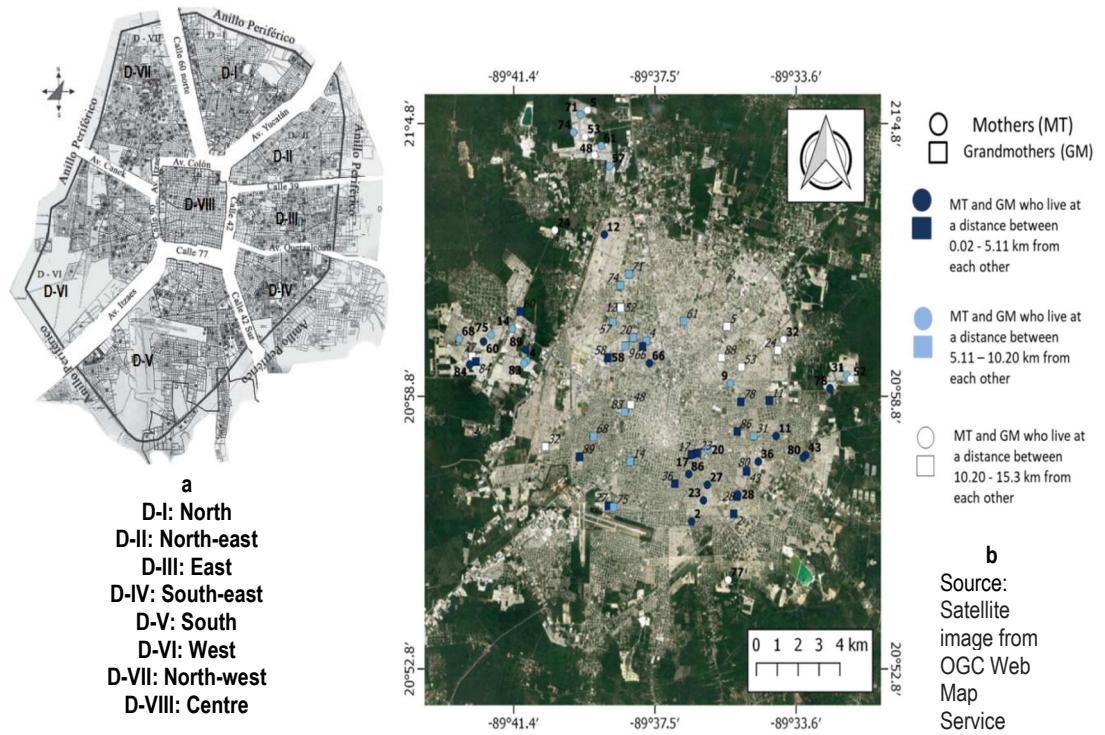
Due to the sample size, this analysis was only descriptive. I wanted to observe a possible pattern between the distance and the days per week and hours per day that the grandmothers invested in taking care of their grandchildren. **Table 5.6** shows the descriptive results.

Table 5.6 Grandmothers investment by distance from mother's home

Distance (km)	N	Days per week				Hours per day			
		mean	SD	median	IQR	mean	SD	median	IQR
Lived together	14	4.8	1.8	5	3	4.0	2.3	3.3	3
0.002-5.11	18	3.4	1.6	3	3	5.4	2.6	5	2
5.11-10.20	12	3.3	1.8	4	3.5	5.7	2.9	4.5	4.5
10.20-15.3	8	3.3	1.8	2.5	3	5.1	1.2	5	1.5

SD: standard deviation; IQR: Interquartile range

Figure 5.30 Distribution of grandmothers and mothers across Merida City



Source: Fuentes 2005

According to the median (IQR), the days per week of contact seemed to decrease in association with geographical distance (Table 5.6); however, it seems that as this distance increased, grandmothers increased the hours per day that they invested in taking care of their grandchildren.

5.5 Discussion

In this sample women invested more in family relationships than in friends, neighbours or other social groups and even though I did not ask the reasons why, it is possible that, in general, individuals invest more in their relatives, especially in maternal relatives (McBurney et al., 2002; Hrdy, 2009). Strong cultural values could explain this result. According to the Yucatan encyclopaedia, the Mayan family is characterized by a strong sense of unity and is considered a fundamental element of the society (Novelo Torres and Echanove Trujillo, 1945). Although I did not work with families with characteristics that can identify them as a contemporary Maya group, such as the presence of Mayan surnames (Relethford, 1995), this strong sense of family unity could be part of general Yucatecan ideology.

However, although relationships with family and friends were important, forming connections and a sense of community with work colleagues, neighbours and other groups (such as sports, hobbies and religious and community groups) could also contribute to women's well-being. In future studies, it will be important to explore more the reasons why women invest less in social relationships outside the family.

In terms of the differences between the maternal groups, the mother-daughter dyads were engaged in more frequent phone/personal contact that provided emotional support and advice. However, women without the support of their mothers were able to find other sources of support within their own family. Interestingly, they found closeness with other female relatives (aunts, cousins, sisters, stepmothers, mothers-in-law and sisters-in-law) that could compensate for the absence of the maternal grandmother. For example, in kinship studies it has been found that in the absence of grandmothers, aunts assume a maternal role and are considered as 'second mothers' (Thornton, 1991; Davidson, 1997; Bastida, 2001; Ellingson and Sotirin, 2006).

In terms of frequency of contact with their grandchildren, it has been described that maternal grandmothers are more inclined to maintain frequent contact as geographical distance increases (Pollet et al., 2006, 2007). In my analysis, it seems that as this distance increased, grandmothers increased the hours per day that they invested in taking care of their grandchildren. However, the pattern is not clear and a larger sample and more detailed data would be needed to analyse the association between the distance and the frequent support from the maternal grandmother.

In relation to partners' support, it is clear that they participated in decision-making at home, home tasks (cleaning, cooking) and tasks related to childcare (spending time with the child, feeding and bathing him/her). It was interesting to observe that women and their partners performed similar activities, although partners participated more in decisions related to economic investment and less in household chores.

Moreover, although fathers invested in their children, this investment was different from the maternal investment. Maternal investment was higher, mainly in terms of the time invested in childcare. This result could be because, due to traditional family and gender roles, women are expected to be the primary caretaker of the children and the house, regardless of the time they need to invest in other activities such as employment, which could represent a further energetic demand on mothers.

Probably, this energetic demand could be higher in women without grandmaternal support considering that they invested more time in taking care of their children during the week than women who have support from their mothers.

5.6 Conclusions

In the present study, maternal social structure is strongly based on family ties. Women's social networks in my sample were mostly composed of family female members, including the maternal grandmother, which could mean that women tended to seek the support and advice of other women within the family, especially if the grandmother is absent.

However, in terms of childcare, women that did not have the support of their mothers invested more in taking care of their children during the week. This could be because they could not feel secure about leaving their children with other family members or because their relatives were not available to provide this support. In future studies, it will be necessary to obtain more detail data about the childcare support sources of women who do not have grandmaternal support.

Partner's participation in home tasks, decisions and childcare activities is part of every couple's dynamic, but the investment is different. Fathers invested more in decisions related to economic investment while women invested more in household chores and childcare.

Summary points

- I. Women's social structure is strongly composed of female family members.
- II. If the mother had access to the grandmother, she represented an important and reliable source of support.
- III. If the grandmother was absent, women found closeness with other female relatives (aunts, cousins, sisters, stepmothers, mothers-in-law and sisters-in-law).
- IV. Women that did not have much time to invest in childcare, asked their mothers for help.
- V. Overall, the families showed a traditional pattern, with fathers investing more in economic decisions and women in household activities and childcare.

CHAPTER 6

NUTRITIONAL STATUS OF THE PARTICIPANTS

PRIMARY HYPOTHESIS AND OTHER ANTHROPOMETRIC OUTCOMES

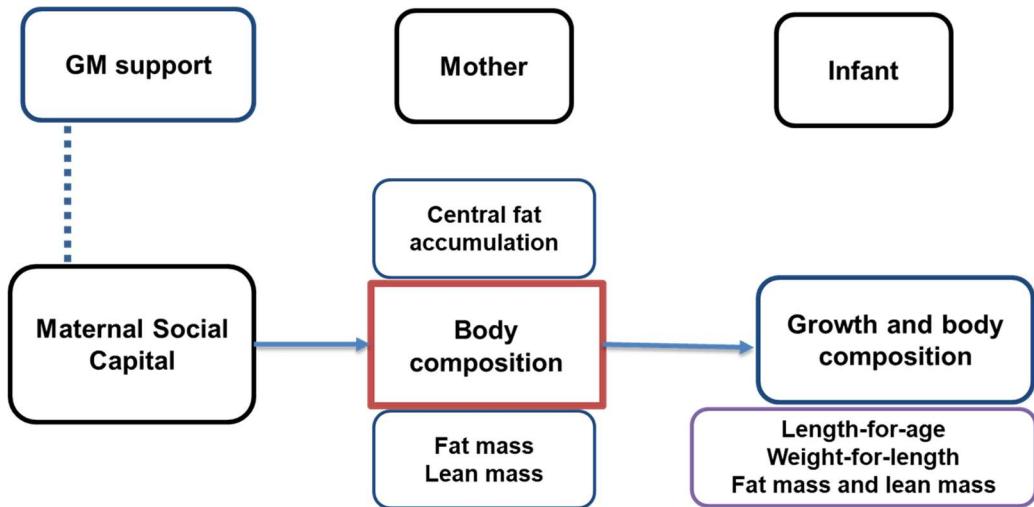
6.1 Introduction

This chapter provides outcomes related to the nutritional status of the mother-infant dyads and maternal grandmothers. Studies from traditional and modern societies show that grandmaternal support is correlated with children's nutritional status and survival (Beise and Voland, 2002; Sear and Mace, 2008; Hrdy, 2009) and that maternal grandmothers are the most influential 'allomother' with a positive effect on the growth of young children (Meehan et al., 2014).

The data obtained and analysed in this section let me test my primary hypothesis, which was that children whose mothers received support from the grandmother would be taller and have higher lean mass (**Table 3.1**). Following the study design (**Figure 3.1**), I also tested the hypothesis that women with grandmothers' support would have more favourable nutritional characteristics.

Hence, I was expecting that women with grandmothers' support would have a healthier nutritional profile which, in turn, would be associated with healthier growth patterns in their children (**Figure 6.1**). There is substantial evidence that the greatest period of plasticity is during prenatal and early postnatal life (Smith et al., 1976; Lucas, 1991; Zhu et al., 2004) when the impact of the external environment on infants is buffered by maternal phenotype (Wells, 2003a). Hence, the association of grandmaternal support and maternal phenotype is important to consider in order to identify potential effects on children's growth at early stages of life.

Figure 6.1 Grandmother's support associated with maternal and infant nutritional conditions



The study examined how the support provided by the grandmother is related to child nutritional conditions through the mediation of the maternal phenotype. Hence, I was expecting that women with grandmothers' support would have a healthier nutritional profile which, in turn, would be associated with healthier growth patterns in their children.

6.2 Data analysis

The descriptive results for the anthropometric data and body composition analysis are presented in this chapter. Moreover, I described body composition results in its relationship with other anthropometric variables for the dyad mother-infant.

The study sample was categorised in two groups: 'with GM support' and 'without GM support', and their results are compared. The main aims of this chapter were: a) to compare my study sample with the references and b) to identify differences between the groups. All participant mothers ($n=90$) completed the anthropometric assessment and measures of 89 children and 48 grandmothers were obtained. For body composition analysis, only 80 dyads mother-infant provided the samples (saliva and urine).

6.3 Statistical analysis

Some data was presented as frequency and or percentages, while univariate analyses (Chi-square/Fisher Exact test) were used to compare groups. Continuous data were tested first for normality distribution by using histograms, Q-Q plots and statistical tests, such as the Kolmogorov-Smirnov test and Shapiro-Wilk test. For normally distributed data, mean \pm standard deviation (SD) was presented along with

the statistical results (T-test) for group comparisons. In those cases in which non-parametric test (Mann-Whitney) was used, the median \pm interquartile range (IQR) was presented.

6.4 Infants

6.4.1 Description of the whole sample (n=89)

Descriptive statistics of anthropometric variables by sex are given in **Table 6.1**. No significant differences were found by sex.

Table 6.1 Descriptive statistics of anthropometric and derived anthropometric variables (Z-scores) of children by sex

	Children (n=89)					
	Boys (n=43)		Girls (n=46)		Sex difference	
	mean	95% CI	mean	95% CI	Mean	95% CI
Total length (cm)	85.9	84.7;87.1	84.9	83.8;86.1	0.9*	-0.7;2.5
Length-for-age (z-score) ^{&}	-0.3	-0.6;0.04	-0.2	-0.5;0.08	-0.1*	-0.5;0.3
Trunk length (cm)	52.2	51.5;52.8	51.3	50.7;52.0	0.8*	-0.2;1.7
Trunk length (z-score) ^{&}	-0.4	-0.7;-0.2	-0.4	-0.7;-0.2	-0.0*	-0.4;0.3
Leg length (cm)	33.7	33.0;34.4	33.8	33.1;34.4	0.0*	-0.9;0.9
Leg length (z-score) ^{&}	-0.4	-0.7;-0.1	-0.4	-0.8;-0.1	0.1*	-0.3;0.5
Weight (kg)	12.2	11.8;12.7	11.8	11.4;12.3	0.4*	-0.1;1.0
Weight-for-length (z-score) ^{&}	0.4	0.1;0.7	0.5	0.2;0.8	-0.0*	-0.4;0.3
Abdominal circumference (cm)	48.5	47.5;49.5	47.9	47.1;48.6	0.7*	-0.4;1.7
Subscapular skinfold (mm)	6.3	5.8;6.8	6.7	6.3;7.0	-0.4*	-1.0;0.2

[#]Boys minus girls

[&]For total length and weight-for-length I compared against the 2006 WHO Growth Standards and for trunk and leg length, I used the 1990 British Growth Reference.

*p>0.05

Only nine per cent of the children met the criteria for stunting (**Table 6.2**). The z-scores for trunk and leg length range from -3.2 to 1.9 (mean = -0.4 ± 0.83) and from -3.2 to 1.8 (mean = -0.4 ± 1.0) respectively. Only six per cent of the infants showed height components below <-2 z-scores.

In terms of weight-for-length, 22.5% were classified as at risk of overweight according to WHO cut-offs, and 4.5% with overweight. There were no significant differences among boys and girls in the variables presented in **Table 6.2**.

Table 6.2 Distribution of nutritional status categories of children by sex

	Children						Total (n=89)	
	Boys (n=43)		Girls (n=46)		Differences			
	n	%	N	%	n	%		
<u>Height (%)</u>								
Non-stunted	38	88.0	43	93.0	Fisher's exact:	81	91.0	
Stunted (<-2 z-score)	5	11.6	3	6.5	p = 0.48	8	8.9	
<u>Height components (%)</u>								
TL (Above <-2 z-score)	41	95.0	41	93.0	Fisher's exact:	82	94.2	
TL (<-2 z-score)	2	4.7	3	6.8	p = 0.51	5	5.7	
LL (Above <-2 z-score)	41	95.0	41	93.0	Fisher's exact:	82	94.2	
LL (<-2 z-score)	2	4.7	3	6.8	p = 0.51	5	5.7	
<u>Weight-for-length (%)</u>								
Healthy	32	74.4	33	71.7	Fisher's exact: p = 0.92	65	73.0	
Risk of overweight*	9	20.9	11	23.9		20	22.5	
Overweight**	2	4.7	2	4.3		4	4.5	

TL: Trunk length; LL: Leg length

* Plotted point above 1 z-score shows possible risk of overweight

** Plotted point above 2 z-score

6.4.2 Stratified by grandmaternal groups

Descriptive statistics of the children's anthropometric variables by grandmaternal group are given in **Table 6.3**. No statistical differences were found among the groups.

Table 6.3 Descriptive statistics of anthropometric and derived anthropometric variables (Z-scores) of children stratified by grandmaternal groups

	Children (n=89)					
	With GM (n=52)		Without GM (n=37)		Differences [#]	
	mean	95% CI	mean	95% CI	mean	95% CI
Length-for-age (z-score) ^{&}	-0.24	-0.50;0.03	-0.31	-0.72;0.10	0.1*	-0.4;0.6
Trunk length (z-score)**	-0.37	-0.56;-0.17	-0.53	-0.87;-0.19	0.1*	-0.3;0.6
Leg length (z-score)**	-0.46	-0.73;-0.19	-0.34	-0.71;0.02	-0.1*	-0.6;0.3
Weight-for-length (z-score) ^{&}	0.31	0.06;0.56	0.28	-0.04;0.60	0.0*	-0.4;0.4
Abdominal circumference (cm)	48.5	47.7;49.3	47.8	46.8; 48.8	0.4*	-0.9;1.7
Subscapular skinfold (mm)	6.2	5.9;6.6	6.5	5.9;7.1	-0.3*	-1.0;0.4

GM: Grandmother

With GM minus Without GM

[&]For total length and weight-for-length I compared against the 2006 WHO Growth Standards

^{**}For trunk and leg length I used the 1990 British Growth Reference.

*p>0.05

No significant differences were found among the children's outcomes by grandmaternal groups in the assessments of growth and nutrition status in **Table 6.4**.

Table 6.4 Distribution of nutritional status categories of children by grandmaternal groups

Children							
	With GM (n=52)		Without GM (n=37)		Differences	Total (n=89)	
	n	%	n	%		n	%
<u>Height (%)</u>							
Non-stunted	48	92.3	33	89.2	Fisher's exact: p = 0.44	81	91.0
Stunted (<-2 z-score)	4	7.6	4	10.8		8	8.9
<u>Height components (%)</u>							
TL (Above <-2 z-score)	49	96.0	33	92.0	Fisher's exact: p = 0.34	82	94.3
TL (<-2 z-score)	2	4.0	3	8.0		5	5.7
LL (Above <-2 z-score)	49	96.0	33	92.0	Fisher's exact: p = 0.34	82	94.3
LL (<-2 z-score)	2	4.0	3	8.0		5	5.7
<u>Weight-for-length (%)</u>							
Healthy	37	71.1	28	75.7	Fisher's exact: p = 0.86	65	73.0
Risk of overweight*	12	23.0	8	21.6		20	22.5
Overweight**	3	5.8	1	2.7		4	4.5

GM: Grandmother; TL: Trunk length; LL: Leg length

* Plotted point above 1 z-score shows possible risk of overweight

** Plotted point above 2 z-score

6.5 Mothers

6.5.1 Description of the whole sample (n=90)

Descriptive statistics of anthropometric variables by age group are given in **Table 6.5**. No statistical differences were found within the anthropometric variables among the age groups. Average height of the women was 155.8cm \pm 6, range 144.5 to 172.5 cm. On average, maternal leg length represented 47% of total height and sitting height 53%.

When compared against the 2006 WHO Growth Standards, 17% were classified as stunted (z-score below <-2). In terms of height components, using the 1990 British Growth Reference, 19% and 12% of the women were below -2 z-scores for sitting height and leg length, respectively.

Table 6.5 Descriptive statistics of anthropometric and derived anthropometric variables (Z-scores) of mothers by age groups

	Women					
	20-29 yrs. (n=46)		30-42 yrs. (n=44)		Differences	
	mean	95% CI	mean	95% CI	mean	95% CI
Height (cm)	156.0	154.3; 157.7	155.5	153.6;157.5	0.2*	-2.4;2.8
Height-for-age (z-score)	-1.1	-1.4;-0.8	-1.2	-1.5;-0.9	0.1*	-0.5;0.5
Sitting height (cm)	82.8	82.1;83.5	82.4	81.2;83.2	0.4*	-0.7;1.5
Sitting height (z-score)	-1.2	-1.4;-1.1	-1.4	-1.6;-1.2	0.1*	-0.2;0.4
Leg length (cm)	73.0	71.7;74.2	73.2	71.8;74.5	-0.2*	-2.1;1.8
Leg length (z-score)	-0.79	-1.1;-0.5	-0.75	-1.1;-0.4	-0.1*	-0.5;0.4
Weight (kg)	63.5	59.8;67.2	67.9	62.7;73.0	-4.4*	-10.7;1.9
BMI	26.3	24.7;27.8	28.2	26.0;30.3	-1.9*	-4.5;0.7
Waist circumference (cm)	85.9	82.5;89.4	91.6	86.7;96.5	-5.7**	-11.6;0.2
Hip circumference (cm)	101.6	98.8;104.5	105.2	101.5;109.0	-3.6*	-8.2;0.9
Tricipital skinfold (mm)	23.3	21.3;25.3	25.0	22.6;27.2	-1.6*	-4.2;1.0
Subscapular skinfold (mm)	18.8	16.5;21.2	21.7	19.1;24.4	-2.9*	-6.7;0.9
Supra-iliac skinfold (mm)	28.2	25.2;31.2	31.0	28.2;33.7	-2.7*	-6.7;1.2

#Differences: G1(20-29 yrs) - G2 (30-42 yrs)

^{*}For height I compared against the 2006 WHO Growth Standards and for trunk (sitting-height) and leg length, I used the 1990 British Growth Reference.

^{*}p>0.05

^{**}p=0.056

Descriptive statistics of body mass variables are also given in **Table 6.5**. No statistical differences were found in weight, BMI, waist and hip circumferences and skinfolds among the age groups. Maternal weight ranged from 44.8 to 121.7 kg (Median=62.1, IQR=14.9), waist circumference from 65.5 to 142.7 cm (Median=87.9, IQR=16.3) and hip circumference from 84.5 to 150.4 cm (Median=101.5, IQR=11.5).

I used waist circumference to determine the risk of abdominal obesity in women. Using the cut-off point of 88 cm proposed by The National Cholesterol Education Program (NCEP) Adult Treatment Panel III (ATP III) (the most commonly used by the Mexican Health authorities), 51% of the mothers were at risk for abdominal obesity. In terms of BMI, 53% of the mothers were classified as having excess weight (overweight + obesity).

Waist-to-hip ratio (WHR) is an important indicator of health risk. Applying a WHR cut-off point of ≥ 0.85 , 54% of mothers showed a high risk to develop metabolic diseases.

6.5.2 Stratified by grandmaternal groups

No statistical differences were found within the anthropometric variables among the grandmaternal groups (**Table 6.6**).

Table 6.6 Descriptive statistics of anthropometric and derived anthropometric variables (Z-scores) of mothers stratified by grandmaternal groups

	Women					
	With GM (n=52)		Without GM (n=38)		Differences [#]	
	mean	95% CI	mean	95% CI	mean	95% CI
Height (cm)	155.0	152.6;156.4	156.4	154.3;158.5	-1.8*	-4.4;0.7
Height (z-score)	-1.3	-1.6;-1.0	-1.0	-1.4;-0.7	-0.3*	-0.7;0.1
Sitting height (cm)	82.1	81.1;83.0	82.7	81.8;83.6	-0.7*	-1.8;0.5
Sitting height (z-score)	-1.4	-1.7;-1.2	-1.3	-1.5;-1.1	-0.2*	-0.5;0.1
Leg length (cm)	72.4	71.1;74.0	73.6	72.3;75.0	-1.2*	-3.0;0.6
Leg length (z-score)	-0.9	-1.2;-0.6	-0.6	-1.0;-0.3	-0.3*	-0.8;0.2
Weight (kg)	66.3	61.2;71.4	65.3	60.3;70.2	1.0*	-6.5;8.5
BMI	27.8	25.6;30.1	26.7	24.8;28.5	1.2*	-1.9;4.4
Waist circumference (cm)	88.9	84.8;93.9	88.1	83.8;92.4	0.8*	-6.2;7.8
Hip circumference (cm)	103.9	100.6;107.1	102.6	99.3;105.9	1.5*	-3.9;6.9
Tricipital skinfold (mm)	23.0	20.6;25.4	24.4	22.1;26.7	-1.4*	-4.7;1.9
Subscapular skinfold (mm)	20.6	18.0;23.2	20.4	17.7;23.1	0.2*	-4.0;4.5
Supra-iliac skinfold (mm)	29.1	26.0;32.4	29.1	26.5;31.7	0.0*	-4.5;4.5

GM: Grandmother

[#]With GM minus Without GM

*p>0.05

In terms of BMI, although not significant, women who received support from the grandmother showed a higher prevalence of excess weight (**Table 6.7**).

Table 6.7 Distribution of categorized maternal body mass variables stratified by
grandmaternal groups

	Women			Total (n=90)
	With GM (n=52)	Without GM (n=38)	Differences	
<u>Waist circumference (%)</u>				
<88 cm	48.0	50.0	$\chi^2_{(1)} = 0.03$, $p = 0.86$	49.0
>88 cm	52.0	50.0		51.0
<u>BMI (%)</u>				
<25	42.3	52.6	$\chi^2_{(1)} = 0.94$, $p = 0.33$	46.7
≥ 25	57.7	47.4		53.3
<u>WHR (%)</u>				
<0.85	44.2	47.4	$\chi^2_{(1)} = 0.09$, $p = 0.77$	45.6
≥ 0.85	55.7	52.6		54.4

GM: Grandmother

WHR: Waist to Hip Ratio

6.6 Grandmothers (n=48)

Given that biological changes may occur during ageing, grandmothers were grouped into three categories of age: 1) 40-49 years, 2) 50-59 years and 3) 60-69 years. Descriptive statistics of anthropometric variables are given in **Table 6.8**. No statistical differences were found in the linear growth variables (height, sitting height and leg length) or body mass variables (weight and BMI) among the age groups (Kruskal-Wallis non-parametric test $p>0.05$).

Table 6.8 Descriptive statistics of anthropometric and derived anthropometric variables (Z-scores) of maternal grandmothers by age groups

	Grandmothers (n=48)					
	40-49 yrs. (n=9)*		50-59 yrs. (n=22)*		60-69 yrs. (n=17)*	
	Mean (\pm)	Median (IQR)	Mean (\pm)	Median (IQR)	Mean (\pm)	Median (IQR)
Height (cm)	149.1(5.5)	148.7(5.4)	150.1(6.7)	151.0(11.2)	147.9(7.0)	148.94(9.2)
Height (z-score)	-2.2(0.8)	-2.2(0.8)	-2.0(1.0)	-1.85(1.7)	-2.2(0.9)	-2.2(1.4)
Sitting height (cm)	78.9(2.8)	79.1(2.7)	80.2(3.3)	80.1(5.1)	78.3(4.0)	79.1(3.0)
Sitting height (z-score)	-2.3(0.7)	-2.2(0.7)	-2.0(0.9)	-2.0(1.3)	-2.2(0.7)	-2.1(0.7)
Leg length (cm)	70.2(3.1)	69.7(2.9)	69.9(4.4)	69.9(7.0)	69.6(3.7)	69.9(5.8)
Leg length (z-score)	-1.5(0.8)	-1.6(0.7)	-1.6(1.1)	-1.5(1.7)	-1.5(0.8)	-1.5(1.5)
Weight (kg)	73.3(17.5)	65.5(8.5)	72.3(13.8)	72.5(22.1)	66.3(16.7)	68.5(24.0)
BMI	32.8(6.1)	31.2(2.4)	32.2(6.4)	30.5(10.0)	29.8(5.4)	30.6(7.8)

*No differences among the age groups in any of the variables (Kruskal-Wallis non-parametric test $p>0.05$).

Due to the lack of statistical differences in the anthropometric variables among the age groups, I now report the values of the same variables without considering age variability among the grandmothers (**Table 6.9**).

Table 6.9 Descriptive statistics of anthropometric and derived anthropometric variables (Z-scores) of maternal grandmothers

	Maternal grandmothers (n=48)			
	mean	SD	Median	IQR
Height (cm)	149.13	6.5	148.9	9.0
Height (z-score)	-2.1	0.9	-2.2	1.4
Sitting height (cm)	79.3	3.5	79.7	3.5
Sitting height (z-score)	-2.1	0.8	-2.0	0.9
Leg length (cm)	69.8	3.9	69.8	5.3
Leg length (z-score)	-1.5	0.9	-1.6	1.3
Weight (kg)	70.2	15.6	68.6	19.1
BMI	31.4	6.0	30.6	6.8

Grandmothers showed very low z-scores for height and sitting height (**Table 6.9**). Fifty-six per cent of them were <-2 z-scores for height-for-age, the indicator for short stature/adult stunting. In terms of height components, 51% and 36% of grandmothers showed z-scores <-2 in trunk and leg length, respectively. In terms of BMI, 86% of the grandmothers were classified as having excess weight (overweight + obesity) and the group as a whole showed a high prevalence of obesity (57.1%).

6.7 Additional analysis

According to **Table 6.3** and **6.5**, there were no differences between the groups in terms of nutritional status. All the comparisons pointed out to the same conclusion: Grandmaternal support did not have any significant association with infants and mothers' nutritional status - the mean differences between the groups were small and not significant. Since the groups also did not differ in background characteristics such as education, it does not seem appropriate to control for such variables.

However, one nutritional variable that was different between the groups was the attitudes of the mothers about breastfeeding. Overall, the complete sample of women showed a high favourable attitude to breastfeeding with a mean score of 67.2 (± 7.6). Nevertheless, statistical differences were found among the groups ($z = 2.54$, $p=0.01$, Wilcoxon rank sum) with women from the no-grandmother group showing a more favourable attitude toward breastfeeding.

Considering that one important factor related to breastfeeding patterns is the women level of education (Brand et al., 2011; Tsai, 2013; Roshan et al., 2018) I did, following the commentaries of the examiners, a multivariate analysis of the attitude of breastfeeding data and include the grandmaternal support and women years of education as a predictors.

The model was validated by confirming that it met residual assumptions, such as normality, variance homogeneity and independence. Non-collinearity between explicative variables was corroborated by calculating the inflation factors for variance between them.

Multiple regression results (**Table 6.10**) for attitudes toward breastfeeding showed that the variables used explained 12% of the variance in this measurement. Having grandmaternal support had a negative and significant effect on breastfeeding attitudes. In contrast, the years of education showed a positive and significant effect in this measurement.

Table 6.10 Multiple regression model for breastfeeding attitudes*

Variable	Coefficient	S. E.	t	P	95% CI
Years of education	0.69	0.28	2.45	0.02	0.12 1.25
Grandmaternal support (with GM)	-4.45	1.60	-2.78	0.007	-7.64 -1.27
Constant	58.52	4.68	12.49	0.000	49.19 67.85

*Breastfeeding attitude scores were normally distributed: Shapiro-Wilk: $w= 0.98$, $p = 0.33$

S.E.: standard error; CI: Confident Interval

$n = 80$, $F (2,77) = 6.48$, $p = 0.003$, adjusted $R^2 = 0.12$, root mean square error = 7.09; Testing residual normality:

Kolmogorov-Smirnov: $k-s= 0.08$, $p = 0.74$; Breusch-Pagan/Cook-Weisberg homocedasticity test: $\chi^2_{(1)} = 1.1$, $p = 0.30$; There was no collinearity of the variables of the model according to the Inflation Factor Variance (IFV).

The previous analysis confirm and complement the results presented in section 4.7.2 'Attitudes towards breastfeeding stratified by grandmaternal groups' of my thesis.

6.8 Discussion

Overall, stunting rates in children were low (9%) and no differences were found among the physical health indicators by sex and grandmaternal groups. Although only 5% of the children were classified with overweight, 23% showed risk for this condition which could compromise their future health.

Compared to the local data (Instituto Nacional de Salud Pública, 2012), the prevalence of stunting and excess weight was lower in my urban sample (Stunting: 9% vs 15%; Excess weight: 5% vs 15.1%).

Among the mothers, overall rates of excess weight (overweight + obesity) were high (53%) and even though no differences were found by grandmaternal groups, women from the grandmother group showed a higher prevalence of obesity according to BMI (58% vs 47%). Furthermore, 51% of the mothers were at risk for abdominal obesity and 54% showed a high risk to develop metabolic diseases according to WHR. Differences in these conditions were not found by grandmaternal groups.

According to the last nutritional survey carried out in 2012 (INSP 2013), in Yucatan the prevalence of overweight and obesity in women was 82.0%, which is higher in comparison to the prevalence showed by my sample (53%). However, it is still concerning that a high percentage of the mothers have poor nutritional status that is associated with an increased risk of chronic non-communicable disease. Considering that in Yucatan among the 5 main causes of mortality are cardiovascular and cerebrovascular disease and diabetes mellitus, this group could be at risk of developing those conditions.

Moreover, although no data was obtained about eating patterns, the excess weight could be related to a lack of physical activity and an inadequate diet, which are patterns that can be inherited and hence, compromise the future health of their children.

On the other hand, maternal grandmothers showed a high prevalence of short stature (56%) and excess weight (86%). In comparison to the children and women, maternal grandmothers showed the worst nutritional condition. However, this pattern could be due partly to the decline in stature that grandmothers are starting to experience. In general, after 40 years of age, total height is reduced by about 1 cm per decade, and this decline may accelerate after 70 years (Minaker, 2012). The reduction in total height affects relatively more the upper body segment than the legs (Chumlea et al., 1985), which could be one factor contributing to the greater reduction in sitting height.

In my sample, it seems that in children and mothers the risk and prevalence of excess weight could be considered until now the main nutritional issue, but in grandmothers, the presence of the 'dual burden' of malnutrition is the main health issue.

The 'dual burden' of malnutrition can be defined as the coexistence of undernutrition (mostly stunting, which indicates nutritional deprivation during the growth period) and excess weight (OW/OB). The double burden of malnutrition can be observed at the individual, household and mother-child levels (Varela-Silva et al., 2012). In my sample, 50% of the grandmothers showed individual-level nutritional dual burden.

In the mothers, 17% (n=15) were classified with adult stunting and only 13% (n=12) with the double burden of malnutrition. None of the infants presented this condition.

The anthropometric data reveal that grandmaternal support did not have any significant association with infant and maternal nutritional condition. However, at least some findings suggested that women from the grandmother group showed a higher prevalence of obesity according to BMI.

Finally, the question that arises from this analysis is whether the children of this sample are at risk of showing poor growth, or whether the population's short stature is (partly) due to genetic factors. Therefore, I incorporated an intergenerational analysis (**Chapter 8**) to address this question and test for secular trends in this sample. This analysis included data from the grandmother-mother-infant triads.

6.9 Conclusions

Overall, the analysis showed that the presence of the maternal grandmother did not show statistical associations with infant growth outcomes or with maternal nutritional status. Hence, my hypothesis was not supported by these results.

At the time of the study, the infants presented healthy growth patterns in terms of height-for-age and weight-for length, although 23% showed risk of overweight. Mothers were informed about this risk and nutritional advice was given.

Mother presented a high prevalence of excess weight, which can compromise their future health and possibly the future health of their children.

Summary points

- I. Stunting rates in the children were low (9%) and only 5% were classified with overweight.
- II. In mothers, rates of excess weight were high (53%), with women from the grandmother group showing a higher prevalence of obesity (58% vs 47%).
- III. A high percentage of the grandmothers (50%) showed an individual-level nutritional dual burden whereas only 13% of the mothers presented this condition.
- IV. Grandmaternal support did not have any significant association with infants and mothers nutritional conditions.

CHAPTER 7

PHYSICAL AND BEHAVIOURAL MARKERS OF STRESS

SECONDARY HYPOTHESIS

7.1 Introduction

Social capital could be an important resource that potentially could have a positive impact on mothers and children's health by decreasing stress responsiveness. Intimate and regular contact with relatives, neighbours, and friends has a beneficial influence on the stress responses (Turner et al., 1999; Light et al., 2000, 2005; Ditzen et al., 2009) probably because individuals with positive social interactions could be able to cope better with the challenges they face in their day-to-day lives.

Moreover, it has been reported that regular contact with social networks enhances competence as a parent (Pridham and Chang, 1992; Mercer and Ferketich, 1994), which could be particularly relevant for women when they are having their first experience as mothers (Leahy Warren, 2005; Leahy-Warren et al., 2012).

In **Chapter 2**, it was stated that maternal grandmothers are more consistent in their inclination to support their daughters during their reproductive careers through direct caregiving and as source of knowledge and resources. Moreover, due to higher levels of empathy with their daughters, support provided by the grandmother could facilitate the parenting process.

From the studies reviewed, it is clear that grandmothers transmit practical knowledge however, is also important to consider that they could increase confidence levels in their daughters, which could have an impact on maternal stress and on their experience as mothers.

Although it is not clear exactly how positive social interactions suppress the stress-response, it seems that one biological mechanism is through the suppression of cortisol concentrations (Heinrichs et al., 2003; Clodi et al., 2008; Ditzen et al., 2009), with positive impacts on mothers' and children behaviours.

Therefore, I hypothesized that women and infants who received support and care, respectively, from the grandmother would be less stressed and have lower levels of cortisol.

This chapter includes the results for the physical and behavioural markers of stress in infants and women. Physical markers of stress were assessed by analysing cortisol concentrations in saliva. The behavioural markers were assessed by the analysis of self-administered scales provided to the women. I explored the relationships between mothers' and infants physical and behavioural markers of stress.

Moreover, levels of resilience and self-efficacy about motherhood were described and their associations with the behavioural marker of stress were explored. Finally, information about maternal responses to child behaviour was assessed. The aim of this section was to explore the strategies used by the mothers when the child showed specific distress behaviours (fear, discomfort, frustration).

The study sample was categorised in two groups: 'with GM support' and 'without GM support', and their results are compared. Only 80 participant mothers and 71 infants provided saliva samples for cortisol analysis and 80 mothers provided the self-administered scales to evaluate their perception of stress, resilience and self-efficacy about motherhood and infant's temperament.

7.2 Methods

Salivary cortisol levels were determined by enzyme-linked immunosorbent assay (ELISA). Perception of stress was assessed through Cohen's Perceived Stress Scale and infant's temperament, by the Early Childhood Behaviour Questionnaire. The mothers filled both instruments. Perception of resilience was assessed through the Mexican Resilience Scale and self-efficacy, through the Parental Evaluation Scale.

7.3 Statistical analysis

Some data was presented as frequency and or percentages, while univariate analyses (Chi-square/Fisher Exact test) were used to compare groups. Continuous data were tested first for normality distribution by using histograms, Q-Q plots and statistical tests, such as Kolmogorov-Smirnov test and Shapiro-Wilk test. For normally distributed data, mean \pm standard deviation (SD) was presented along with the statistical results (T-test) for group comparisons. In those cases in which a non-parametric test (Mann-Whitney) was used, the median \pm interquartile range (IQR) was presented.

7.4 Physical markers of stress: Salivary cortisol levels

7.4.1 Cortisol levels in infants and mothers

One hundred and fifty-one saliva samples were analysed (71 infants, 80 mothers) to measure cortisol levels. I was unable to obtain saliva samples from nine infants, as they were reluctant to use the cotton wool or spit. Mothers tried to help me to obtain the samples, but they started to feel frustrated and stressed due to the infants' crying. Therefore, I decided to not obtain the samples.

As I stated in [section 3.11.1](#) of the Chapter 3, morning samples were obtained after waking and before breakfast and physical activity. Participant women were not breastfeeding when the samples were obtained and they were not in the menses phase of their menstrual cycle.

In order to determine a baseline range of salivary cortisol, I looked for studies that reported salivary cortisol concentrations in women and infants. This search was done considering the protocol used for samples collection and analysis. In my study, samples were obtained between 6:00 and 8:00 hrs (morning samples) and were analysed using a commercially available ELISA assay (DRG Diagnostics, Germany).

The studies found can be seen in [Table 7.1](#). All studies obtained morning saliva samples from premenopausal women and, in some studies, from infants, and analysed the samples using commercial ELISA kits from different manufacturers (Salimetrics, DRG and Cambio).

Table 7.1 Baseline cortisol concentrations presented in previous studies

Source	Sample	Protocol for hormone measures	Baseline cortisol measures
Harris et al 2000 Place of research: Islington, London	83 female subjects possibly vulnerable to depression and 33 non-vulnerable. None pregnant women with a mean age of 38.5 (± 7.06) years. They had at least one child at home, were cohabiting with the partner or spouse and were not taking corticoid medication.	Morning saliva samples (8:00hrs) and night samples (20:00hrs) on four consecutive days. Wash mouth before spitting, not clean teeth and not use any aids to salivation. None of the women was suffering for major depression at the time the baseline cortisol measures were carried out.	Average (SD) of the four morning samples Non-vulnerable women 3.30 (± 1.98) ng/ml Possibly vulnerable 3.63 (± 1.96) ng/ml
Pico-Alfonso et al 2004 Place of research: Alicante, Castellon and Valencia, Spain	46 non-abused, 70 physically abused and 46 psychologically abused women. None pregnant women with a mean age of 45.9 (± 11.2) years. From the non-abused group, 98% were cohabiting with the partner during saliva collection.	Morning saliva samples (8:00 and 9:00hrs) and night samples (20:00 and 21:00hrs) on four consecutive days. Starting the fourth day after the beginning of menstruation. Wash mouth before spitting, not clean teeth and not use any aids to salivation.	Average (SD) of the four morning samples Non-abused women 2.90 ng/ml
Feldman et al 2010 Place of research: Israel	53 mothers (28.9 ± 4.0 yrs.) and their 6-month-old infants (25 boys, 25.65 ± 1.3 weeks). Singleton healthy infant, mother married or cohabitating with the infant's father, mother with at least high-school education. Dyads were tested in two paradigms: still-face (SF) and still-face with maternal touch (SF+T)	Morning saliva samples obtained with Salivettes from both mother and infant to assess baseline salivary cortisol.	Average morning samples from both paradigms Infant SF+T: 2.2 ± 0.6 ng/ml and SF: 2.2 ± 0.5 ng/ml Mothers SF+T: 2.2 ± 0.5 ng/ml and SF: 2.3 ± 0.7 ng/ml
Govender 2010 Place of research: South Africa	5 women with psychiatric diagnosis and 4 non-psychiatric premenopausal women. Median age of control women: 26.6yrs.	Saliva samples were obtained at 7:00, 13:00, 18:00 and bedtime hours. Using this data, 24hr cortisol levels were obtained.	Median of the cortisol levels at 7:00am in control women 8.4 ng/ml

Table 7.1 Baseline cortisol concentrations presented in previous studies

Source	Sample	Protocol for hormone measures	Baseline cortisol measures
Blasco-Ros et al 2014 Place of research: Alicante, Castellon and Valencia, Spain	73 women victims of intimate male partner violence (IPV) and 31 not exposed to IPV. None-abused women: 46.03 (± 12.92) years. 97% married and/or living with the partner. They had at least one child at home, were cohabiting with the partner or spouse and without taking corticoid medication.	The participants provided saliva samples a minimum of twice a day (between 08:00 and 09:00 and between 20:00 and 21:00) for four consecutive days, starting the fourth day after the beginning of menstruation.	Average (SD) of the four morning samples Non-abused women 2.93 (± 1.1) ng/ml
Neelon et al 2015 Place of research: Durham, NC, USA	Participants of the Newborn Epigenetic study (NEST). Sub-sample of 54 mother/infant dyads. Women > 18 yrs. Infants' age: 8.1 (± 2.2) months.	Morning saliva samples: upon waking, 30 min after waking and at bedtime over the course of a single day. Women: passive drool method avoiding cleaning teeth, consuming food or beverage, and smoking. Infants: using a sorbetto after feeding and rinse and wipe their mouths prior to the second morning sample collection.	Mean (\pm SD) Awakening-Formula feeding dyads (n=34) Infants: 5.9 ± 8.7 ng/ml Mothers: 3.9 ± 1.6 ng/ml Awakening-Breastfeeding dyads (n=20) Infants: 7.5 ± 1.2 ng/ml Mothers: 3.5 ± 2.7 ng/ml 30 min after waking Formula feeding dyads Infants: 4.6 ± 5.4 ng/ml Mothers: 4.5 ± 1.7 ng/ml 30 min after waking Breastfeeding dyads Infants: 6.1 ± 1.0 ng/ml Mothers: 3.8 ± 1.9 ng/ml
Ewert et al 2016 Place of research: Indiana, USA	8 premenopausal women between 20 and 35 yrs.	Morning saliva samples obtained between 10:00 and 13:00 hrs. Participants were cautioned not to eat or drink anything one hour before the data collection time	Average of morning samples: 3.3ng/ml Range: 2.7-5.9 ng/ml

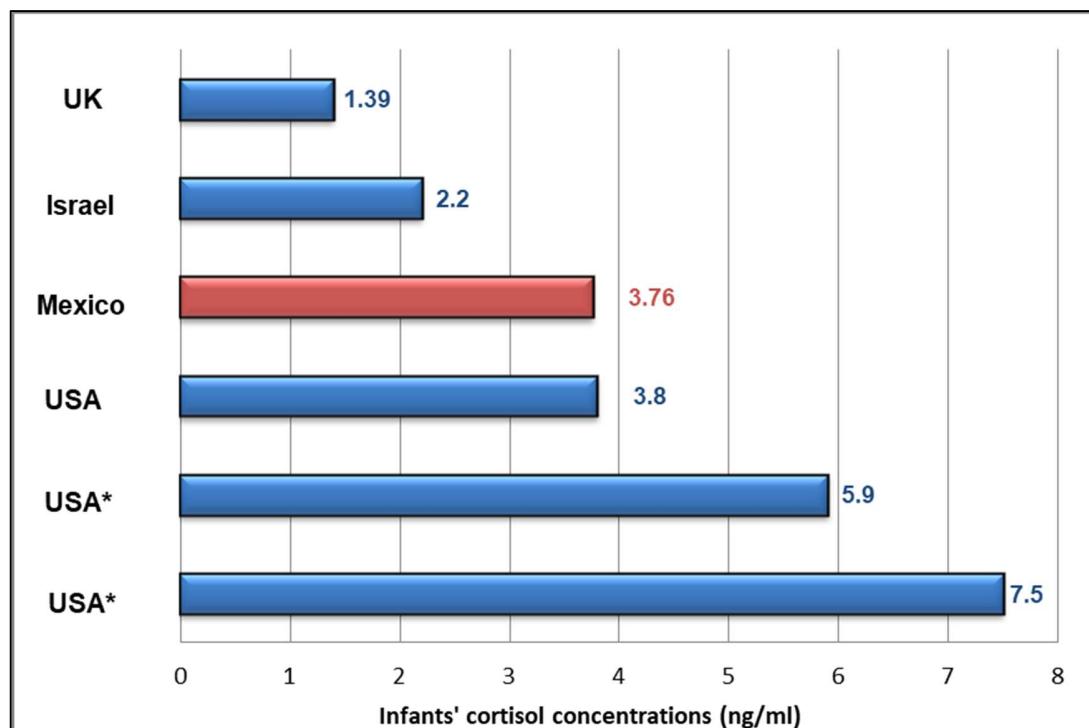
Table 7.1 Baseline cortisol concentrations presented in previous studies

Source	Sample	Protocol for hormone measures	Baseline cortisol measures
Heidemarie et al 2016 Place of research: Cardiff, UK	100 mother–infant dyads. Infants: 2yrs old Mothers: 22 to 43 yrs. (Mean: 33.4 ± 4.5 yrs.). 91% women were married and 78.2% had a university degree Note: This is a longitudinal study in which cortisol levels were measured when the child was 1, 2 and 3 yrs. I am only considering baseline cortisol levels at 2 yrs.	Two baseline samples obtained at 9:15am and 9:30am. Sorbettes were used for collecting saliva from the infant's mouth and women were asked not to feed their babies during assessment.	Mean ($\pm SD$) Baseline levels at 2 yrs. of age 9:15am (n=63 infants) 1.39 ± 1.49 ng/ml (Range: 0.10-11.66) 9:30am (n=60 infants) 2.63 ± 7.08 ng/ml (Range= 0.29-45.01) Without caffeine use: 8.0 ng/ml
Rieth et al 2016 Place of research: France	19 healthy young recreationally trained females (physical activity for 4–6 h/ week for at least 2 years) aged 19.5 ± 0.3 yrs. All taking low-dose oral contraceptive pills for at least 2yrs. No smokers, not using any medical drug and without chronic diseases.	Morning saliva samples were obtained between 6:00am and 7:00am using Salitubes. Two groups were formed: ten without any daily caffeine use, and 9 caffeine users.	With Caffeine use: 6.0 ng/ml Mean ($\pm SD$)
Tarullo et al 2017 Place of research: Boston, USA	121 mother–infant dyads. Infants: 6.67 ± 0.43 months of age Mothers: 33.4 ± 4 years. 93% women living with the partner and 87.4% having a college degree or higher. Infants were not from multiple births.	Morning saliva samples obtained at the time of awakening using synthetic swabs. Samples were excluded if the participants used any medication or collected the samples more than 60 min after waking.	Infants: 3.8 ± 2.7 ng/ml Mothers: 2.6 ± 1.5 ng/ml
Van Gaasbeck et al 2017 Place of research: North Carolina, USA	36 premenopausal women without health problems and no alcohol consumption.	Two samples obtained: one immediately after waking and one immediately before going to sleep on the same day.	Morning cortisol mean 5.7 ± 2.2 ng/ml

According to my review, in previous studies, morning cortisol levels in women have been reported to be between 2.2ng/ml and 8.4ng/ml. Moreover, the DRG assay using in my study establishes that the normal morning range expected in adults, after analysing samples from 109 healthy subjects, is between 1.2ng/ml and 14.7ng/ml. In infants, according to my review, morning cortisol levels have been reported to be between 2.2ng/ml and 7.5ng/ml. The DRG assay does not include information about salivary cortisol levels in infants.

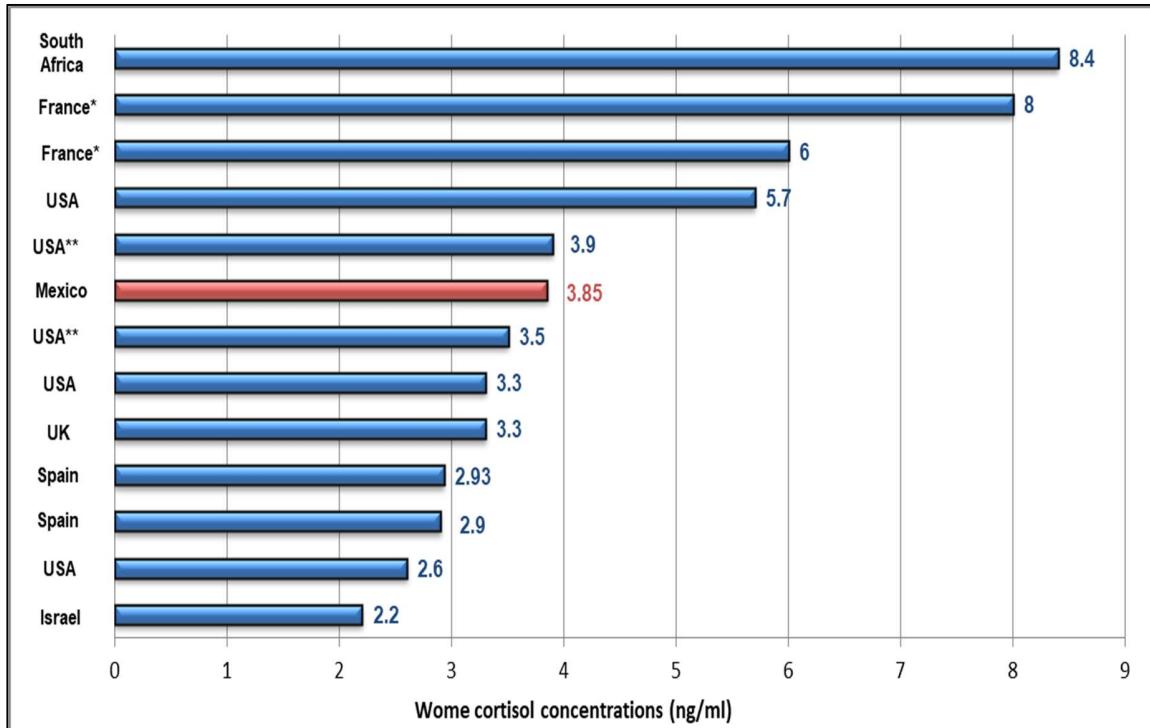
In my sample, the average morning cortisol level in women was $3.85(\pm 3.06)$ ng/ml and in infants, $3.76(\pm 3.32)$ ng/ml. Overall, according to the descriptive data, the cortisol concentrations found in my sample are within the range indicated above according to what was found in previous studies (Figure 7.1 and 7.2).

Figure 7.1 Awakening/Morning cortisol concentrations in infants reported by previous studies compared to the concentrations found in this study (in red)



*Same study, two groups of infants

Figure 7.2 Awakening/Morning cortisol concentrations in women reported by previous studies compared to the concentrations found in this study (in red)



*/**: Same study, two groups of women

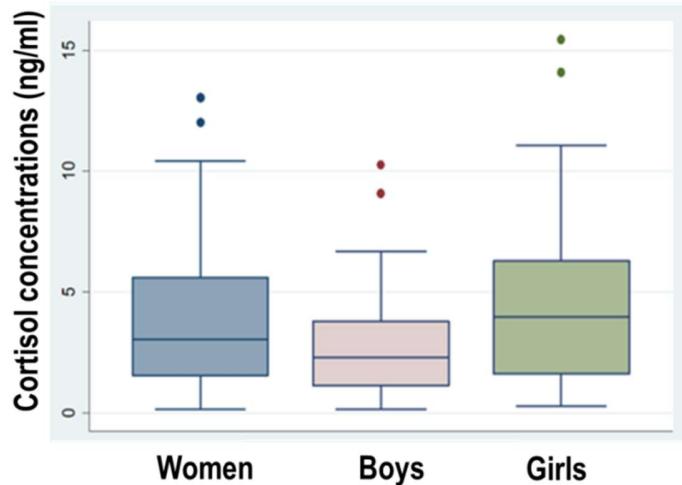
The data are strongly right-skewed as can be seen in Table 7.2 and Figure 7.3, so cortisol scores were log-transformed prior to analysis to correct positive skew.

Table 7.2 Women and infants' cortisol concentrations (ng/ml)

	n	mean	SD	p25	median	p75
Women	80	3.8	3.1	1.5	3.0	5.6
Infants	71	3.8	3.3	1.3	2.7	5.2
Boys	34	2.8	2.4	1.1	2.3	3.8
Girls	37	4.7	3.8	1.6	4.0	6.3

SD: Standard deviation; p25: 25th percentile; p75: 75th percentile

Figure 7.3 Women's and infants' cortisol concentrations (ng/ml)



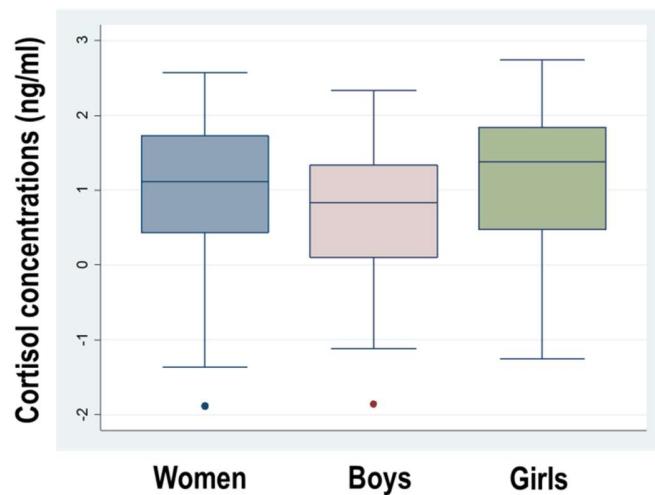
The LN-transformation of the data reduced the skew (Table 7.3 and Figure 7.4). I applied tests of normality to determine the goodness of fit of the distributions. According to the Kolmogorov-Smirnov normality test, the natural logarithm of the salivary concentration of cortisol in women is normally distributed (Combined K-S= 0.10, p=0.62). Shapiro-Wilk normality test was applied to determine normality distribution in infants' natural logarithm concentrations by sex and Kolmogorov-Smirnov, to determine data distribution fit of the overall sample. According to the results, cortisol concentrations by sex are normally distributed (Boys: w= 0.97, p = 0.57; Girls: w= 0.96, p = 0.15), similar to the overall sample data (Combined K-S= 0.10, p=0.83).

Table 7.3 Natural logarithm of the salivary concentration of cortisol in women and infants

	n	mean	SD	p25	median	p75
Women	80	0.98	0.95	0.43	1.11	1.72
Infants	71	0.91	1.00	0.24	0.99	1.64
Boys	34	0.67	0.97	0.10	0.83	1.33
Girls	37	1.13	1.00	0.46	1.40	1.84

SD: Standard deviation; p25: 25th percentile; p75: 75th percentile

Figure 7.4 Natural logarithm of the salivary concentration of cortisol in women and infants



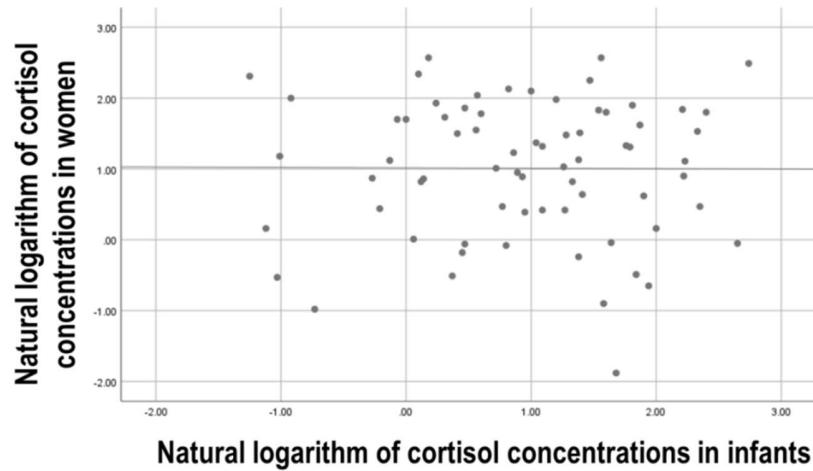
7.4.2 Correlation between maternal cortisol concentrations and infants' concentrations

Several studies have found that children's cortisol levels are positively correlated to maternal stress (Champagne et al., 2001, 2003; Ashman et al., 2002; Essex et al., 2002; Champagne and Meaney, 2007; Heim et al., 2008). I therefore explored the relationship between maternal and infant cortisol concentrations.

First, I tested for differences between women and children. No differences were found in the natural logarithm concentrations between women and infants ($t_{(149)} = 0.47$, $p=0.64$) and between boys and girls ($t_{(69)} = -1.97$, $p=0.05$).

Then, I analysed the correlation between these variables. In this analysis, I only included the cases in which samples from both the mother and her child were obtained ($n=71$). No correlation was found between the cortisol concentrations of the mothers and their children ($P_r = -0.01$, $p=0.96$) (Figure 7.5).

Figure 7.5 Relationship between women and infants' cortisol concentrations



7.4.3 Correlation between maternal cortisol concentrations and infants' concentrations stratified by grandmaternal groups

No differences were found in infant cortisol by grandmaternal groups. In women, the pattern observed in the salivary cortisol concentrations by grandmaternal groups was that mothers that belong to the non-grandmother group presented lower levels of cortisol in saliva (Table 7.4).

Table 7.4 Natural logarithm of cortisol concentrations in mothers and infants stratified by grandmaternal groups

	n	Mean	SD	p25	Median	p75	Differences
Infant							
With GM	43	0.98	0.96	0.18	1.19	1.76	$z = -0.60$, $p=0.56$
Without GM	28	0.80	1.10	0.42	0.84	1.48	Wilcoxon rank sum
Women							
With GM	45	1.15	0.94	0.64	1.32	1.80	$z = -1.88$, $p=0.059$
Without GM	35	0.76	0.93	-0.04	0.81	1.53	Wilcoxon rank sum

Considering that in **section 5.4.9** (see also **figure 5.29**) I showed that 88% (n=39) of the women reported feeling relaxed when they left their child with their mothers, I analysed if the cortisol levels were different between this group and the group without grandmaternal support (n=35). There was weak evidence of a difference was found between these groups ($z=-1.8$, $p=0.08$; Rank sum: women with GM support= 1626 and women without GM support= 1149), however contrary to expectations, the women who reported feeling relaxed showed higher cortisol concentrations (Median=1.4, IQR=2 VS Median= 0.8, IQR=1.5).

My secondary hypothesis was not supported by these results. Several reasons might explain why I found the opposite results in the cortisol saliva concentrations. First, some of the mothers that were living with the grandmothers pointed out that they were living together due to the partner spending five to six days outside the city due to work commitments. This low physical contact with the partner during the week might be causing stress to the mother. Second, it is necessary to know the quality of the relationship between the partner and the maternal grandmother. It is clear that they benefit from the support provided by the grandmother; however, childcare decisions made by the grandmother could be accepted by the mother, but not by the partner. In this sense, women could be experiencing stress because of a possible conflict between their partner and mothers.

On the other hand, a conflict between mothers and grandmothers over different styles of parenting might also be creating stress for the mothers. As I stated in **section 5.4.7** of the maternal social capital chapter (Cognitive social capital by grandmaternal groups), only 83% of women from the grandmother group reported that the grandmother was the most reliable person for taking care of the child. Some of the women reported that they felt that their mothers were spoiling their children. Therefore, it is possible that some children who are cared for by the grandmother behaved worse and showed lengthy periods of whining or tantrums due to the permissiveness of the grandmother. Commenting on a study carried out by Fouts et al (2005) of infant crying in relation to two types of parenting style, Wells (2005) argued that infants who receive frequent response from their parents or other relatives, such as the grandmother, to their needs, can feel more 'comfortable' in signalling their demands as a means to obtain more resources, since these signals tend to receive a response. Hence, the ways and the frequency in

which caregivers respond to infants needs can influence their personality. Permissiveness from the grandmother could be associated in the way in which infants signal their need to their mothers and the way in which women perceived their children behaviour. The analysis of the infants' behaviour by grandmaternal groups is presented in **section 7.5.6**.

Moreover, it is also important to consider that according to the results of **section 5.4.5**, women who received support from their mothers seek their support when they have a problem or a concern. It is possible to consider that these women could feel more confident in expressing their frustrations with their mothers, which could lead to 'revive the bad experience' or 'revive the concern', causing stress to the mothers.

7.5 Behavioural markers of stress in mothers and infants

7.5.1 Stress assessment in mothers: Cohen's Perceived Stress Scale (PSS-14)

To assess perceived stress in women, they were asked to complete the PSS-14. **Table 7.5** shows the descriptive statistics (median, IQR) of the PSS and its individual factors. Seven out of the fourteen items of PSS-14 are considered negative (1, 2, 3, 8, 11, 12, 14) and the remaining seven as positive (4, 5, 6, 7, 9, 10, 13), representing perceived helplessness and self-efficacy respectively. Total scores for PSS-14 range from zero to 56 with a higher score indicating greater stress (0-14: almost never stressed; 15-28: every now and then she is stressed; 29-42: she is often stressed; 43-56: very often she is stressed).

Overall, according to the scores classifications, women perceived being stressed 'every now and then' (median score=22.5) (**Table 7.5**). Hence, women from this sample perceived relatively low levels of stress.

Table 7.5 Descriptive statistics of the PSS-14 results

	mean	SD	p25	median	p75	IQR
PSS (total score)	22.4	7.0	18	22.5	27	9
Factors						
Helplessness	14.4	4.9	12	14	18	6
Self-efficacy	1.13	1.00	0.46	1.40	1.84	5

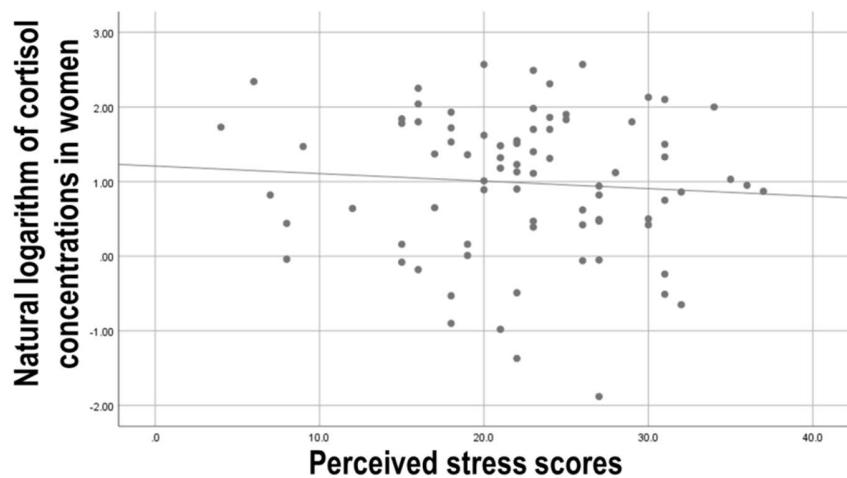
SD: Standard deviation; p25: 25th percentile; p75: 75th percentile; IQR: Interquartile range

7.5.2 Correlation between maternal perceived stress and cortisol concentrations

Correlations between physical and behavioural markers of stress were assessed in the women, to explore whether women reporting a higher level of perceived stress also showed higher cortisol scores.

However, no correlation was found between the cortisol concentrations in mothers and their levels of perceived stress ($S_r = -0.07$, $p=0.52$) (Figure 7.6).

Figure 7.6 Correlation between maternal cortisol levels and perceived stress



7.5.3 Stress assessment in mothers stratified by grandmaternal groups

I hypothesized that women that have support from their mother would have lower scores in their stress perception. However, no statistical differences were found in the mother's stress perception by grandmaternal groups (Table 7.6). Therefore, my secondary hypothesis was not supported by these results.

Table 7.6 Descriptive statistics (median, IQR) of the PSS-14 scores stratified by grandmaternal groups

	With GM	Without GM	Differences Statistical tests
PSS (total score)	22.0 (7)	23.0 (12)	$z = -0.78$, $p=0.43$, Wilcoxon rank sum
Factors			
Helplessness	14.0 (6)	15.0 (8)	$z = -0.88$, $p=0.38$, Wilcoxon rank sum
Self-efficacy	8.0 (4)	9.0 (5)	$z = -0.41$, $p=0.68$, Wilcoxon rank sum

IQR: Interquartile range; GM: Grandmother

Note: IQR are presented in parentheses under the median

Considering that in **section 5.4.9** (see also **Figure 5.29**) I showed that 88% ($n=39$) of women reported feeling relaxed when they leave their child with their mothers, I analysed if the perceived stress scores were different between this group and the group without grandmaternal support ($n=35$). No difference was found between these groups ($z=0.9$, $p=0.34$; Rank sum: women with GM support= 1375 and women without GM support= 1399).

7.5.4 Infant temperament assessment: The Early Childhood Behaviour Questionnaire

To assess infant temperament, mothers completed the ECBQ, which comprises 201 items that assess 18 domains of temperament, which form three broad factors: Surgency, Negative Affectivity and Regulatory Capacity/Effortful Control. Mothers were asked to rate the frequency of specific temperament-related behaviours observed over the past two weeks on an ordinal scale ranging from one (never) to five (always); a not applicable option was also provided. Scale scores for the eighteen dimensions represent the mean score of all scale items applicable to the child, as judged by the caregiver.

Table 7.7 shows the descriptive statistics of the 18 domains of temperament of the overall sample. The lowest scores (Median: ≤ 2.5) were found in four domains: discomfort, motor activation, fear, and sadness. These domains belong to the Negative Affectivity factor, which is characterized by sadness, discomfort, frustration, fear, and difficulty to soothe (Putnam et al., 2006). The higher scores (Median: ≥ 3.5) were found in three domains: impulsivity, sociability and low-intensity pleasure. The first two domains belong to the Surgency factor, which is characterized by high activity level, high-intensity pleasure

seeking, low shyness, and impulsivity. The third domain belongs to the Effortful control factor, which encompasses inhibitory control, attentional focusing, low-intensity pleasure, and perceptual sensitivity (Putnam et al., 2006).

Table 7.7 Descriptive statistics of the 18 domains of temperament in infants

	mean	SD	p25	median	p75	Min	Max
Activity level/Energy	2.9	0.4	2.7	2.9	3.2	1.8	3.8
Attentional focusing	3.3	0.3	3.1	3.2	3.5	2.5	4.0
Attentional shifting	3.0	0.4	2.7	2.9	3.2	2.1	4.2
Cuddliness	3.4	0.6	3.0	3.4	3.7	1.7	4.6
Discomfort	2.2	0.5	1.9	2.1	2.5	1.4	4.1
Fear	2.5	0.5	2.2	2.5	2.9	1.4	4.7
Frustration	3.0	0.5	2.7	2.9	3.3	1.8	4.3
High-intensity Pleasure	3.1	0.4	2.7	3.0	3.4	1.6	4.3
Impulsivity	3.5	0.4	3.2	3.5	3.7	2.8	4.5
Inhibitory control	2.8	0.6	2.4	2.8	3.3	1.6	4.5
Low-intensity pleasure	3.9	0.6	3.5	3.9	4.2	2.4	5.0
Motor activation	2.1	0.6	1.7	1.9	2.4	1.0	5.0
Perceptual sensitivity	3.4	0.6	3.0	3.4	3.8	1.6	5.0
Positive anticipation	3.3	0.6	2.9	3.3	3.7	1.7	5.0
Sadness	2.3	0.6	2.0	2.3	2.6	1.2	5.0
Shyness	2.7	0.5	2.3	2.7	3.2	1.7	4.0
Sociability	3.6	0.6	3.1	3.6	4.0	1.9	4.7
Soothability	3.3	0.4	3.0	3.3	3.6	2.0	4.3

Table 7.8 shows the descriptive statistics (median, IQR) of the three factors of the ECBQ by sex. No differences were found by sex in any of the evaluated factors (Wilcoxon rank sum $p>0.05$). Overall, according to the mothers, infants showed a higher frequency of

temperament behaviours related to extraversion and regulatory capacity over the past two weeks before answering the questionnaire.

Table 7.8 Descriptive statistics of the ECBQ factors by sex

ECBQ factors	Boys (n=40)		Girls (n=40)		Total (n=80)	
	Media n	IQR	Media n	IQR	Media n	IQR
Negative affectivity	2.8	0.4	2.9	0.4	2.8	0.4
Surgency/Extraversion	3.4	0.4	3.3	0.3	3.3	0.4
Regulatory capacity or effortful control	3.3	0.3	3.1	0.5	3.2	0.4

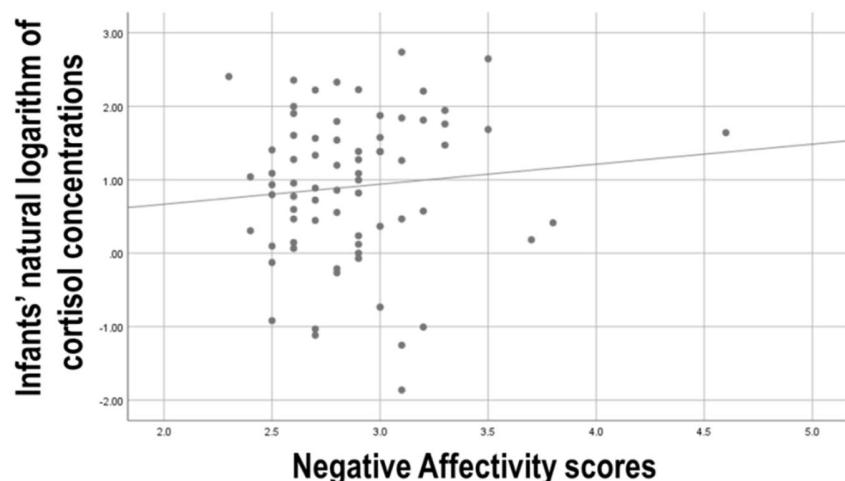
IQR: Interquartile range

7.5.5 Correlation between infants' temperament assessment and cortisol concentrations

Because the Negative affectivity factor might be related to distress in infants, I tested whether higher infants' cortisol concentrations correlated with higher levels of negative affectivity in infants.

The correlation was not significant ($S_r = 0.20$, $p=0.17$) and no clear pattern was found (Figure 7.7).

Figure 7.7 Relationship between infants' cortisol concentrations and negative affectivity



7.5.6 Infant temperament assessment stratified by grandmaternal groups

Caretaker behaviour can influence infant personality (Crockenberg and Acredolo, 1983). Consequently, the impact of grandmaternal support may potentially be associated with the infant's temperament. I hypothesized that infants that received care from their maternal grandmothers would have more frequent positive behaviours and hence, lower scores on the Negative affectivity factor. However, no statistical differences were found by grandmaternal group (**Table 7.9**) in the three temperament factors. Therefore, my secondary hypothesis was not supported by these results.

Table 7.9 Descriptive statistics (median, IQR) of the ECBQ factors stratified by grandmaternal groups

ECBQ factors	With GM	Without GM	Differences Statistical tests
Negative affectivity	2.8 (0.4)	2.9 (0.4)	$z = 0.96, p=0.34$, Wilcoxon rank sum
Surgency/Extraversion	3.2 (0.5)	3.4 (0.3)	$z = 1.72, p=0.08$, Wilcoxon rank sum
Regulatory capacity or effortful control	3.3 (0.3)	3.2 (0.2)	$z = -0.07, p=0.95$, Wilcoxon rank sum

IQR: Interquartile range; GM: Grandmother

Note: IQR are presented in parentheses under the median

In **section 7.5.3**, I stated that one reason why women from the grandmother group showed a higher mean saliva cortisol concentration could be that children who are cared for by the grandmother behaved worse and showed lengthy periods of whining or tantrums due to the permissiveness of the grandmother. However, according to the infant temperament evaluation made by the mothers, the children showed similar behaviour, regardless of the group they belonged to. Hence, differences in women's cortisol concentrations could be related to other factors, such as an event and/or concern that some mothers from the grandmaternal groups could experience before the saliva collection (Flinn and England, 2003; Nepomnaschy and Flinn, 2009). Moreover, there is still the possibility that differences in parenting styles between grandmothers and

daughters could be one of the factors; however, more data are needed to support this statement.

On the other hand, another possible explanation for the lack of differences is that infants from this sample are not only cared for by their mothers and maternal grandmothers. At least 83% of the infants spent time in childcare centres in which the caregiving engagement could be related to their emotional expressions. Hestenes et al (1993) found that children in higher quality school environments displayed more smiling, laughing, and showed greater intensity of this positive affect than children in lower quality environments. The best predictor of children's affect focused on the teachers' behaviour (e.g., appropriate involvement and interaction, encouragement of receptive and expressive language, appropriate scheduling and supervising of activities).

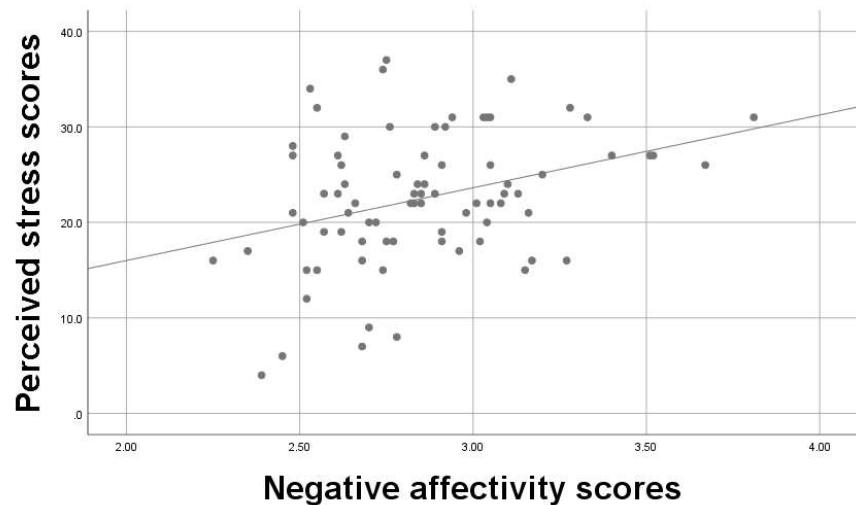
Hence, in my sample, infants from the grandmother group could experience positive interactions with their mothers, maternal grandmothers and teachers and infants from the none-grandmother group could have these interactions with their mothers, other women from their family, such as aunts, and teachers. Therefore, the different social relationships experienced by the infants could be related to the results obtained.

However, it is also important to consider that a potential limitation of the infant temperament evaluation is that only mothers provided this information and no other observer was in charge of this activity. Hence, a subjective positive bias of maternal perception on infants' temperament needs to be considered.

7.5.7 Correlation between maternal perceived stress and infants' negative affectivity

Due to children's behaviour being reported to correlate with maternal behaviour (Ashman et al., 2002), I explored whether infants' negative affectivity was related to maternal perceived stress. I hypothesized, that women that perceived higher levels of stress were have infants with higher levels of negative affectivity. My result showed that mothers who perceived high levels of stress perceived their children as having more behaviours related to the negative affectivity factor ($S_r = 0.31$, $p=0.005$) ([Figure 7.8](#)).

Figure 7.8 Relationship between mothers' perceived stress and infants' negative affectivity



7.6 Maternal resilience

7.6.1 Resilience assessment: Scale of Resilience in Mexicans (RESI-M)

Broadly, resilience is a quality or characteristic of a person that implies the possession of several skills that help individuals to overcome adversities and emerge strengthened from them (Reivich and Shatté, 2002).

Although there is not yet a consensus about the definition of resilience (Kalawski and Haz, 2003), this could refer to the capacity of individuals to face and overcome the problems and stressful events of life, and results in successful adjustment to their environments despite challenging circumstances (Masten et al., 1990; Suárez, 1996).

Table 7.10 shows the descriptive statistics of the RESI-M scale and its factors. Overall, according to the median, women showed high levels of resilience, with higher scores in strength and self-confidence, family support and social support factors.

Table 7.10 Descriptive statistics of the RESI-M and its factors

	mean	SD	p25	median	p75	Min	Max
Resilience (total score)	3.4	0.3	3.2	3.4	3.7	2.7	4.0
Factors							
Strength and self-confidence	3.5	0.4	3.2	3.6	3.7	2.6	4.0
Social competence	3.1	0.5	2.6	3.0	3.4	1.8	4.0
Family support	3.6	0.5	3.2	3.8	4.0	1.2	4.0
Social support	3.7	0.4	3.5	4.0	4.0	2.8	4.0
Structure (life organization)	3.2	0.5	3.0	3.2	3.6	2.2	4.0

SD: Standard deviation; p25: 25th percentile; p75: 75th percentile; Min: minimum; Max: Maximum

It has been suggested in the literature that social support can help to relieve the negative effects of stress and that the family can be the most important source of support promoting the skills and self-esteem needed to overcome adversity (Seccombe, 2000). According to my results, women perceived themselves as capable of achieving the goals they set and probably this confidence is reinforced by the high levels of family support they perceived. Although women showed a high score in social support, considering the results of the maternal social capital chapter, this social support comes more from the family than from friends and other social groups.

7.6.2 Resilience assessment stratified by grandmaternal groups

I hypothesized that women that received support from their mother would have higher scores in their resilience perception. **Table 7.11** shows the descriptive statistics (median, IQR) of the RESI-M scale and its factors by grandmaternal group. No statistical differences were found among the groups.

Table 7.11 Differences in the RESI-M results and its factors (median, IQR) stratified by grandmaternal groups

	With GM	Without GM	Differences	
			Statistical tests	
Resilience (total score)	3.4 (0.5)	3.4 (0.4)	$z = 0.96, p=0.34$, Wilcoxon rank sum	
Factors				
Strength and self-confidence	3.5 (0.6)	3.6 (0.6)	$z = 0.96, p=0.34$, Wilcoxon rank sum	
Social competence	3.0 (0.6)	3.1 (0.5)	$z = 0.96, p=0.34$, Wilcoxon rank sum	
Family support	3.8 (0.8)	3.7 (0.8)	$z = 0.96, p=0.34$, Wilcoxon rank sum	
Social support	4.0 (0.4)	4.0 (1.0)	$z = 0.96, p=0.34$, Wilcoxon rank sum	
Life organization (norms, rules, activities)	3.2 (0.6)	3.0 (1.0)	$z = 0.96, p=0.34$, Wilcoxon rank sum	

IQR: Interquartile range; GM: Grandmother

Note: IQR are presented in parentheses under the median

Having support from the maternal grandmother was not associated with the levels of resilience of women in this sample. As I stated before, mothers from the none-grandmother group found support and closeness with other female relatives (aunts, cousins, stepmothers, mothers-in-law and sisters-in-law) that could be compensating for the absence of the maternal grandmother and having a positive impact on their resilience perception.

7.6.3 Maternal resilience and perceived stress

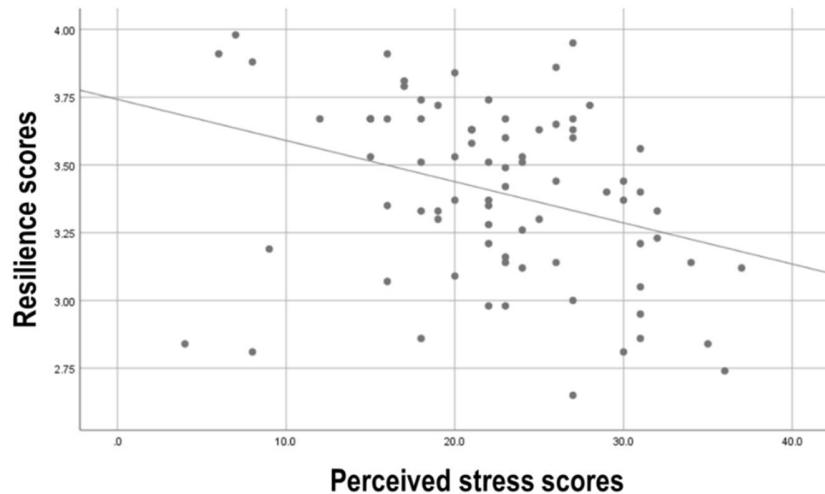
Although the presence of the maternal grandmother was not a determinant of women's levels of resilience, the results presented in this section are relevant considering that high levels of resilience could be helping mothers to cope with stressors.

Therefore, I explored the correlation between maternal levels of resilience and maternal perceived stress. A negative and significant correlation was found ($S_r = -0.40, p<0.001$).

Figure 7.9 shows graphically that the lower levels of resilience, the higher the levels of perceived stress.

Women from this sample perceived having high levels of resilience that could be helping to reduce their stress levels and hence, their stress perception.

Figure 7.9 Correlation between maternal resilience and perceived stress

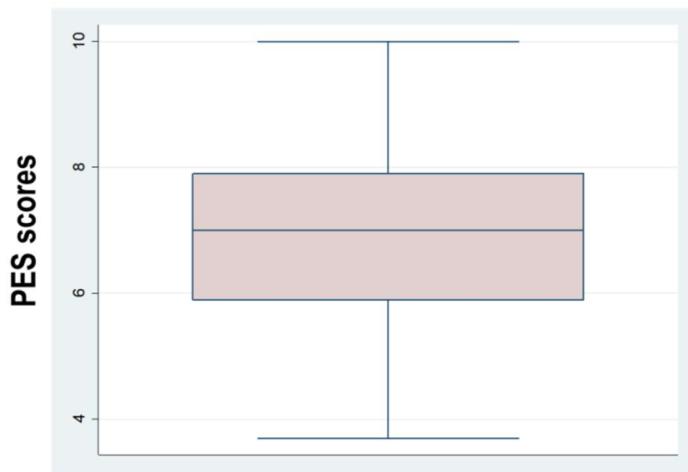


7.7 Satisfaction and self-efficacy about motherhood: Parental Evaluation Scale (PES)

Another important aim of this study was to evaluate how the mother perceived her ability to respond to the needs of her child. This perception could be mediated by her sense of social support. Therefore, co-parenting with other relatives, such as the grandmother, could have an impact on the way women perceived their own efficacy as mothers.

Figure 7.10 shows the PES scores of the overall sample. The median (IQR) score was 7.0 (2.0) with a range between four and 10. Considering that the maximum score was 10, overall, women showed high levels of self-efficacy and maternal satisfaction.

Figure 7.10 Women PES scores

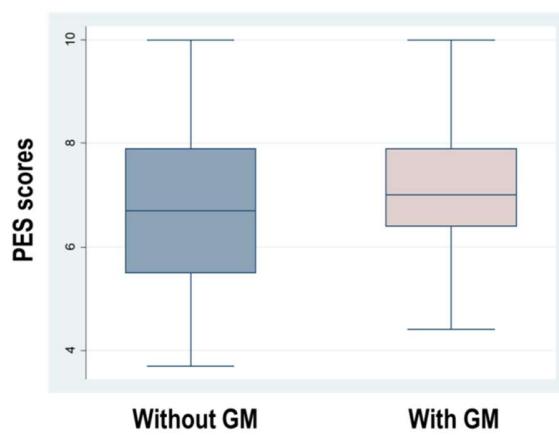


7.7.1 Satisfaction and self-efficacy stratified by grandmaternal groups

Results from the PES by grandmaternal groups are shown in **Figure 7.11**. The median (IQR) scores from the group that received grandmaternal support were 7.0 (1.5) vs 6.7 (2.4) for the other group.

No statistical differences were found among the groups ($z = -0.81$, $p=0.41$, Wilcoxon rank sum) and although women who did not receive support from their mothers showed more variability in their results and the lowest scores, neither of these differences was very large.

Figure 7.11 Scores from the Perceived Stress Scale stratified by grandmaternal groups



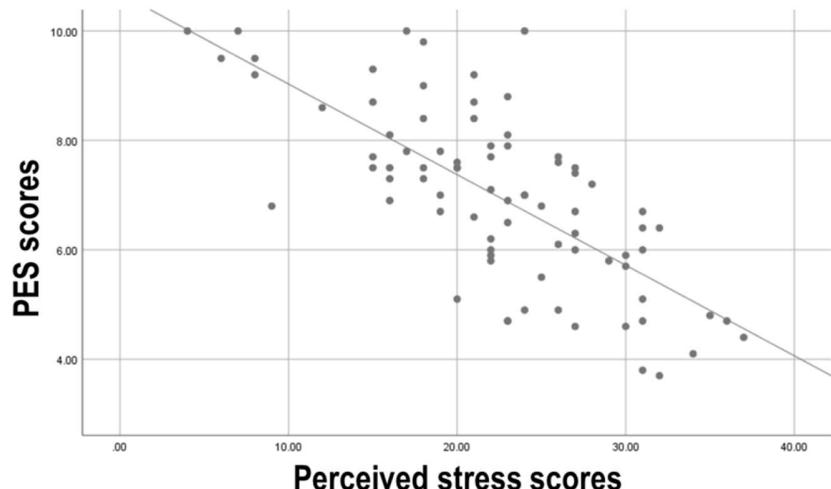
GM: Grandmother

7.7.2 Satisfaction and self-efficacy about motherhood and perceived stress

Although the presence of the maternal grandmother was not associated with women's self-efficacy about motherhood, the results presented in this section are relevant considering that in terms of maternal outcomes, women with greater parenting self-efficacy experienced fewer symptoms of postpartum depression and psychological distress (Cutrona and Troutman, 1986; Halpern and Mclean, 1997; Haslam et al., 2006).

In my sample, self-efficacy and perceived stress of women were negatively correlated ($S_r = -0.71$, $p < 0.001$) (Figure 7.12). The greater the perception of maternal efficacy, the lower the levels of perceived stress.

Figure 7.12 Correlation between maternal self-efficacy (PES scores) and perceived stress



Parents who believe in their own abilities will generally feel more satisfied and perceive themselves as competent parents (Kendall and Bloomfield, 2005). In the face of stressful situations, mothers with low self-efficacy could feel less satisfied and unable to do what is necessary to persevere and achieve certain tasks, which can result in an increase in negative emotions (Cutrona and Troutman, 1986). In contrast, mothers with high self-efficacy are more confident and experience less negative emotions (Bandura, 1982; Cutrona and Troutman, 1986).

In this sense, a high perception of self-efficacy could result in better care for the infants and in better mental health of the mother. In my sample, the women perceived themselves as efficient mothers with low levels of perceived stress.

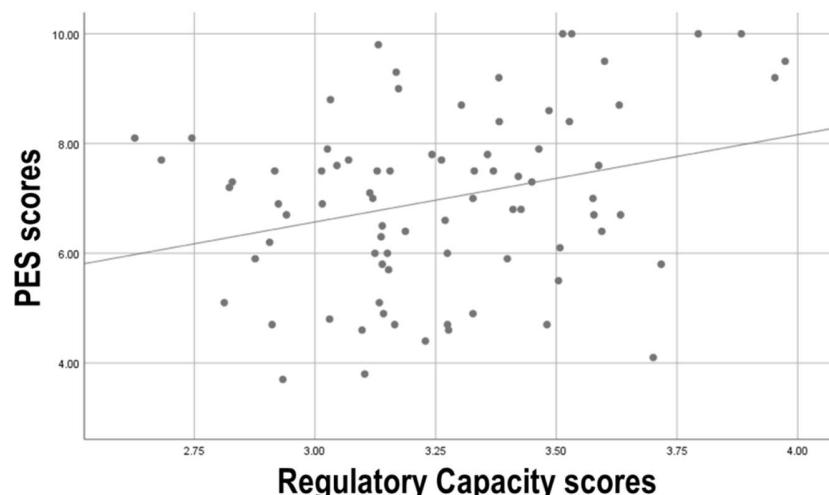
Being able to identify maternal self-efficacy could allow the implementation of interventions to strengthen efficacy beliefs and improve maternal skills and women's mental health.

7.7.3 Maternal satisfaction and self-efficacy and infant temperament assessment

Infants with negative emotionality could negatively affect maternal levels of self-efficacy (Leerkes and Crockenberg, 2002; Porter and Hsu, 2003; Hsu and Lavelli, 2005; Leerkes and Burney, 2007). In my sample, no significant correlation was found between mothers' self-efficacy and infants' negative affectivity ($S_r = -0.21$, $p=0.065$). However, it is still important to consider that positive and/or negative interaction between mothers and their children could be associated with maternal self-efficacy (Leerkes and Crockenberg, 2002; Hsu and Lavelli, 2005).

Considering positive interactions, maternal self-efficacy and infants' regulatory capacity were positively correlated in my sample ($S_r=0.23$, $p=0.038$). Mothers that perceived in their infants more behaviours related to the regulatory capacity factor, showed a greater perception of maternal efficacy (Figure 7.13).

Figure 7.13 Correlation between maternal self-efficacy and infants' regulatory capacity

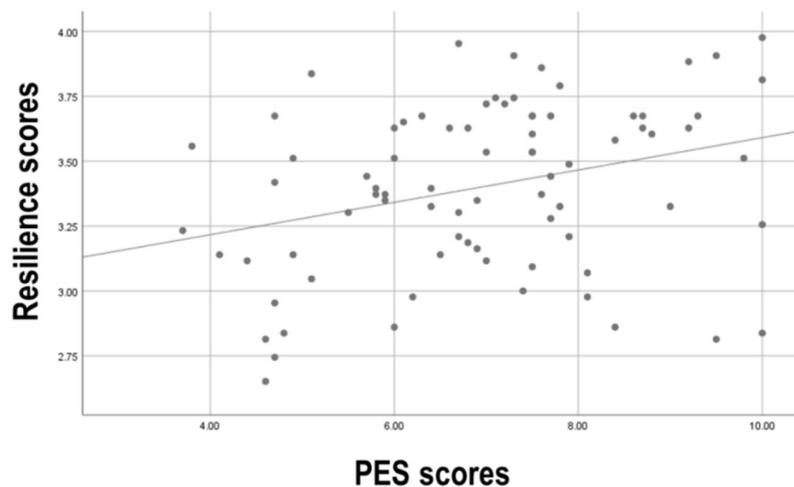


Hence, is clear that the way in which women perceived infants' temperament is associated with maternal efficacy. In my sample, women who stated that their children showed greater self-control and emotional-regulation (i.e. regulatory capacity) reported feeling more satisfied with their parental skills and perceived themselves as more efficient mothers.

7.7.4 Maternal satisfaction and self-efficacy and women's resilience

According to my results, women from my sample felt capable of achieving their goals and felt satisfied with their maternal skills, which translated into low levels of stress. It seems that resilience and self-efficacy are intimately related. Hence, I explored the correlation between them (Figure 7.14).

Figure 7.14 Correlation between maternal self-efficacy (PES scores) and resilience



Women's resilience and self-efficacy were significantly correlated ($S_r = 0.30, p < 0.01$). The greater the levels of resilience, the higher perception of maternal efficacy.

Resilience and self-efficacy influence the way people think and the way they respond to the challenges they face (Bandura, 1977; Seccombe, 2000) and hence, the coping behaviours they choose to overcome stressful events. Moreover, these abilities are related to social competence or sociability, which is the ability of individuals to develop

and establish healthy relationships with others (Murray, 2003). Having good quality social networks can significantly increase an individual's ability to adjust to environmental conditions and adapt to stressors (Garmezy, 1991; Pellegrini et al., 1999; Murray, 2003).

In my sample, family factors could be protecting mothers from the negative effects of stress. Grandmothers are important, however, it seems that women develop the ability to find support from other family members when the grandmother is absent.

Therefore, detailed information about the family environment could be relevant. Data about family cohesion and warmth, and the absence of family discord, as well as having in childhood at least one adult who served as social support (Grossman and Tierney, 1998) may be important to consider in future studies.

7.8 Responses to child behaviour: Parental strategies questionnaire

7.8.1 Mothers responses to child behaviour

Overall in my sample, women showed low levels of perceived stress, high levels of resilience and high levels of maternal self-efficacy, while according to the ECBQ, mothers rated their children with high scores in three domains of temperament (impulsivity, sociability and low-intensity pleasure) that belong to the Surgency and effortful control factors. This suggests that the women perceived their children as showing self-control and positive social behaviours.

However, is important to consider that women also perceived behaviours related to the negative affectivity factor (discomfort, motor activation, fear and sadness), although this factor was rated with lower scores. In this sense, I was interested to know how women responded to relieve infants' distress.

First, I asked the mothers to complete The Child Distress and Parental Response Questionnaire (Wells, 1998) to obtain information about how often the child was distress by different perceived causes (frustration, tiredness, discomfort or hungry). Then, I asked them which strategy they used to relieve distress (cuddle the baby, offer food/drink/pacifier, distract with toys/TV or ignore the child).

It has been suggested that infants perceived as difficult may be fed more to quieten them, which could be related to subsequent fatness (Carey, 1985; Wells et al., 1997). Therefore, caregivers' response to child behaviour may have important consequences for young children health.

Table 7.12 shows the frequency per day that according to the mothers, infants' showed distress behaviours. Only in those cases in which mothers perceived some of the behaviours more than once a day or once a day, women were asked to provide information about the strategy they used to relieve distress in the child.

Only 47.5% (n=38) of the mothers perceived tiredness in their children more than once a day/once a day. From this percentage, 8% (n=3) and 16% (n=6) of women admitted to using foods or drinks, respectively, to relieve child tiredness. The food provided were bananas, biscuits, jelly, cereal, chicken soup and rice and the drinks provided were juice, water and milk.

Table 7.12 Frequency of child distress behaviours perceived by the mothers

	Mothers (n=80)			
	More than once a day (%)	Once a day (%)	Less than once a day (%)	Never (%)
Tiredness	10.0	37.5	36.2	16.2
Discomfort	7.5	36.2	40.0	16.2
Hunger	68.7	23.7	7.5	---
Thirst	76.2	17.5	6.2	---
Fear	2.5	10.0	47.5	40.0
Frustration	2.5	8.7	37.5	51.2

Forty-four per cent (n=35) of the mothers perceived discomfort in their children more than once a day/once a day. From this percentage, 11% (n=4) and 23% (n=8) of women admitted to using foods or drinks, respectively, to relieve child discomfort. The food provided were fruits, vegetables (cucumber, broccoli), biscuits, jelly, cereal, bread and Danonino® and the drinks provided were juice, water, milk and Yakult®.

The highest percentages of daily distress were attributed to hunger and thirst (92% and 94% respectively). All the women responded by giving food or drink to their children. The food provided were fruits, vegetables, cereal, biscuits, bread, sandwich, rice, tortilla,

pancakes, pasta and chicken soup, eggs, meat, Danonino®, yoghurt, Gerber®, turkey sausage, beans, oats and sweets. The drinks provided were water, juice, milk, water with fruit (orange, lemon), rice water and PaediaSure®. Women did not specify more about the types of foods and drinks they gave to the infants.

Finally, only 12.5% and 11.2% of the women perceived fear and frustration, respectively, in their children more than once a day/once a day. When women perceived fear and frustration they responded by cuddling the child or distract the child with toys or the TV. None of the mothers provided foods or drinks when the child showed these behaviours.

Overall, I found mothers that admitted using foods to relieve some causes of their children distress, such as tiredness and discomfort. The use of food to relieve distress may have important consequences on infants eating patterns.

7.8.2 Mothers responses to child behaviour stratified by grandmaternal groups

Table 7.13 shows the percentage of women that perceived some of the different causes of child distress stratified by grandmaternal groups. Only 24%(n=11) of the mothers from the grandmother group and 29% (n=10) women from the other group admitted to using foods to relieve child distress that was not attribute to hunger or thirst. Specifically, they used food or drinks when they perceived tiredness and discomfort from the child once a day or more often. No differences were found between the groups.

Table 7.13 Frequency of child distress behaviours perceived by the mothers by grandmaternal groups

	More than once a day (%)		Once a day (%)		Less than once a day (%)		Never (%)		Differences
	GM	no GM	GM	no GM	GM	no GM	GM	no GM	
Tiredness	8.9	11.4	37.7	37.1	35.6	37.1	17.8	14.3	Fisher's exact: p = 0.97
Discomfort	4.4	11.4	35.6	37.1	40	40	20	11.4	Fisher's exact: p = 0.57
Hunger	68.8	68.6	24.4	22.9	6.7	8.6	---	---	Fisher's exact: p = 1.00
Thirst	75.6	77.1	15.6	20	8.8	2.9	---	---	Fisher's exact: p = 0.56
Fear	---	5.7	11.1	8.6	55.6	37.1	33.3	48.6	Fisher's exact: p = 0.15
Frustration	2.2	2.8	8.9	8.6	35.6	40	53.3	48.6	Fisher's exact: p = 0.97

GM: n=45

No GM: n= 35

7.8.3 Grandmothers responses to child behaviour

Due to 56% of the children being taken care for by the grandmother between two to seven days per week, and three to 12 hours per day, grandmothers were also asked to respond to this questionnaire to explore which strategy they used to relieve distress in their grandchildren. The aim was to explore if the strategies used by them were similar to the strategies used by their daughters.

Table 7.14 shows the frequency per day that according to the grandmothers the infants showed distress behaviours. Forty-nine per cent (n=22) of the grandmothers perceived tiredness in their grandchildren more than once a day/once a day. From this percentage, 27% (n=6) of grandmothers admitted to using drinks to relieve infants' tiredness. The drinks provided were juice or milk.

Table 7.14 Frequency of child distress behaviours perceived by the grandmothers

	Child distress grandparental responses			
	Grandmother (n=45)			
	More than once a day (%)	Once a day (%)	Less than once a day (%)	Never (%)
Tiredness	6.7	42.2	17.8	33.3
Discomfort	11	35.6	26.7	26.7
Hunger	57.7	35.6	6.7	---
Thirst	77.8	17.8	2.2	2.2
Fear	4.4	2.2	33.3	60
Frustration	2.2	8.9	35.6	53.3

Forty-seven per cent of (n=21) the grandmothers perceived discomfort in their children more than once a day/once a day. From this percentage, 33% (n=7) and 24% (n=5) of grandmothers admitted to using foods or drinks, respectively, to relieve child discomfort. The food provided were fruits, biscuits, jelly, cereal, bread and milk sweets and the drinks provided are juice or milk.

As with the mothers, the highest proportion of distress was attributed to hunger and thirst (93% and 96% respectively). All the grandmothers responded by giving food or drink to their grandchildren. The food provided were fruits (apple, banana, papaw), vegetables (broccoli), cereal, pancakes, biscuits, bread, sandwich, rice, potato, lentils, *tortilla*, pancakes, pasta and chicken soup, eggs, meat (pork), Danonino®, yoghurt, Gerber®, sausage, beans, oats, jelly, chocolate, and milk sweets. The drinks provided were juice and milk, water with fruit (orange, lemon) or rice water.

Only 7% and 11% of the grandmothers perceived fear and frustration, respectively, in their grandchildren more than once a day/once a day. When they perceived these behaviours in their grandchildren, they responded by cuddling the child or distracted the child with toys or the TV. None of the grandmothers provided foods or drinks when the child showed these behaviours.

As with the women group, I also found grandmothers that admitted using foods to relieve some causes of their grandchildren's distress, such as tiredness and discomfort. The use

of foods and drinks to soothe the infants may potentially have an impact on their eating and health patterns.

7.8.4 Comparison between grandmothers and mothers

Overall, mothers and grandmothers seem to perceive similarly the frequency of infants' distress behaviours. The only difference was found in the frequency of infants' fear behaviour perceived by the women (Table 7.15). Sixty-per cent of the grandmothers never perceived fear in their grandchildren.

Regarding the strategies they used to soothe infants, a higher percentage of grandmothers (40% vs 29%) used food or drinks to relieve distress behaviours, which may have an impact in on the infant's fatness.

Table 7.15 Frequency of child distress behaviours perceived by the mothers and grandmothers

	More than once a day (%)		Once a day (%)		Less than once a day (%)		Never (%)		Differences
	M	G	M	G	M	G	M	G	
Tiredness	8.9	6.7	37.7	42.2	35.6	17.8	17.8	33.3	Fisher's exact: p = 0.16
Discomfort	4.4	11	35.6	35.6	40	26.7	20	26.7	Fisher's exact: p = 0.41
Hunger	68.8	57.7	24.4	35.6	6.7	6.7	---	---	Fisher's exact: p = 0.55
Thirst	75.6	77.8	15.6	17.8	8.8	2.2	---	2.2	Fisher's exact: p = 0.52
Fear	---	4.4	11.1	2.2	55.6	33.3	33.3	60.0	Fisher's exact: p = 0.01
Frustration	2.2	2.2	8.9	8.9	35.6	35.6	53.3	53.3	Fisher's exact: p = 0.31

M: Mothers; G: Grandmothers

7.9 Additional analysis

The difficulty about re-analyse the behavioural data is that both outcomes and potential confounders between the groups did not show any indication of differing. The mean differences between the groups were small and not significant in terms of socioeconomic conditions and behavioural characteristics (**Table 7.6, 7.9 and 7.11** and **Figure 7.11**).

However, when I analysed this data without considering which grandmother group the mothers belonged to, some associations were found between maternal resilience, perceived stress, self-efficacy and infant's negative affectivity. Hence, following the hypothesis of the study, I did a multivariate analysis of the perceived stress data and include the following control variables: grandmaternal support, levels of resilience and self-efficacy, education and infant's negative affectivity.

Including the grandmaternal support variable helped me to verify if there is a chance to find differences between the groups once controls are included. The resilience, self-efficacy and infant's negative affectivity data were included due to their high correlation with perceived stress ($Sr=-0.40$ $p<0.001$, $Sr=-0.71$ $p<0.0001$ and $Sr=0.31$, $p=0.005$, respectively). Considering the possibility that different educational backgrounds could be related to different levels of stress (Makowsky et al., 1988; Hauksdóttir et al., 2013), I included the maternal years of education. For instance, it has been reported that women with lower levels education had higher levels of perceived stress, feelings of less control, and lower levels of satisfaction with family life than women with more education (Makowsky et al 1988). Moreover, a study carried out in Iceland found that psychological stress may have increased following the recent economic collapse, particularly among females in economically vulnerable groups (unemployed, students and women with middle level of education) (Hauksdóttir et al 2013).

The model was validated by confirming that it met residual assumptions, such as normality, variance homogeneity and independence. Non-collinearity between explicative variables was corroborated by calculating the inflation factors for variance between them.

Multiple regression results (**Table 7.16**) for perceived stress showed that the variables used explained 52% of the variance in this measurement. Having grandmaternal support

had a negative but not significant effect on stress. Moreover, the only variable that significantly explained the variance of this variable was the levels of resilience.

Table 7.16 Multiple regression model for Perceived Stress scores*

Variable	Coefficient	S. E.	t	P	95% CI	
Self-efficacy and satisfaction about motherhood	-2.96	0.36	-8.26	<0.001	-3.67	-2.24
Resilience	-1.72	1.85	-0.93	0.36	-5.42	1.98
Infant's negative affectivity	0.94	1.60	0.59	0.56	-2.23	4.1
Years of education	-0.31	0.20	-1.54	0.13	-0.71	0.09
Grandmaternal support (with GM)	-2.25	1.11	-0.23	0.82	-2.46	1.96
Constant	51.36	8.78	5.84	0.000	33.84	68.87

*Perceived stress scores were normally distributed: Shapiro-Wilk: $w = 0.98$, $p = 0.33$

S.E.: standard error; CI: Confident Interval

$n = 80$, $F(5,74) = 18.41$, $p < 0.001$, adjusted $R^2 = 0.52$, root mean square error = 4.86; Testing residual normality: Shapiro-Wilk: $w = 0.97$, $p = 0.20$; Breusch-Pagan/Cook-Weisberg homocedasticity test: $X^2(1) = 2.18$, $p = 0.14$; There was no collinearity of the variables of the model according to the Inflation Factor Variance (IFV).

The same procedure was applied for the resilience and self-efficacy variables and I obtained the same results: having grandmaternal support did not significantly explain the variance of these measurements.

7.10 Discussion

After completing the analyses described in this chapter, it is important to consider potential limitations or bias in the data that might explain the lack of associations between physiological and behavioural markers of stress. First, women were asked to obtain saliva samples at the moment of awakening. However, in 70% of the cases, women decided to wait for the research team to collect the samples. This implied a gap of 1 to 2 hrs (or probably more) between the moment of awakening from both mother and infant and the saliva collection, which might affect the analysis. Due to the cortisol circadian rhythm (cortisol levels reach their lowest levels at around midnight and levels start to rise at around 02:00 to 03:00 and reach a peak at around 08:30) higher levels are expected at the moment of awakening (Chan and Debono, 2010), and a decrease in levels can be seen 30 min after waking. Hence, the time (hour) in which the mother and child woke up and the samples were obtained, needed to be considered. Unfortunately, I did not measure the gap between waking and sampling. Second, during the time gap mentioned before, women probably had to attend to their infants' needs, which implied a moderate

physical activity, a factor that can also affect cortisol levels. Moreover, the infants might have commenced play activities while waiting for the research team to arrive, which also can be considered as physical activity. Third, mothers were asked to not feed the baby before obtaining the samples. However, I am not sure if women followed this instruction rigorously. During fieldwork, women reported that children asked, specifically for milk, at the time of awakening, so I am not sure if all women refused this demand of the child.

Other factors that need to be considered are health status and daily activities of both mother and child. Cortisol levels rise in acute medical illness (Cornil et al., 1968; Drucker and Shandling, 1985) and the events and concerns that people experience in their day-to-day lives can vary depending on the day of the week and the activities related to that day (Flinn and England, 1997, 2003; Nepomnaschy and Flinn, 2009), i.e., activities, demands and concerns during the week are not the same that during weekends. During fieldwork, several infants presented viral exanthema of the hands, feet and mouth (Coxsackie A16), and, in addition, several mothers and their children presented infectious conjunctivitis. However, a detailed register of the health conditions of the participants and the duration of the diseases at the time of saliva collection was not considered.

Moreover, cortisol levels can vary depending on the phase of the menstrual cycle (Nepomnaschy et al., 2004). However, during fieldwork, the research team decided to obtain women samples when they were not in their menstrual phase. This decision was made for two reasons: a) because bioimpedance data were being obtained and the protocol suggested doing this test during the absence of menstruation and b) to avoid more home visits to the mothers. Hence, the variability in my data could be related to the changes in the profiles of the reproductive hormones.

Although the range of cortisol concentrations in my sample lay within the range reported in other studies (Harris, 2000; Pico-Alfonso et al., 2004; Feldman et al., 2010; Govender et al., 2011; Bright et al., 2012; Blasco-Ros et al., 2015; Neelon et al., 2015; Rieth et al., 2016; Ewert et al., 2016; Laurent et al., 2016; Tarullo et al., 2017; Gaasbeck et al., 2017), my results need to be carefully considered due to the limitations mentioned above, and also because accurate baseline cortisol levels should ideally be calculated after obtaining several samples during the day, to consider circadian rhythm, and during the week and then, calculating baseline levels considering the variability obtained (Nepomnaschy et al.,

2004). Unfortunately, due to time constraints, it was not possible to obtain more than one sample per participant to calculate baseline cortisol levels.

Therefore, results presented by grandmaternal groups need to be carefully interpreted. I cannot state for sure that the grandmother is a factor causing higher cortisol levels in women from the grandmother group. More studies with a more rigorous methodology on hormone measurements will be needed to assess if the maternal grandmother support is related to their daughters' and grandchildren levels of cortisol.

On the other hand, no differences were found by grandmaternal groups in the behavioural markers of stress of mothers and infants. Women perceived themselves as resilient and self-efficacious mothers with low levels of perceived stress. Moreover, women perceived that their children show more behaviours related to regulatory capacity, which was related to women's perception about their efficacy as mothers. Hence, perceiving having high levels of resilience, self-efficacy and children with positive behaviours could be helping women to overcome stressful events and having good mental health. However, it is important to consider that their positive perception about themselves could be determining their perception about their children having high positive behaviours.

In the present study, maternal social structure is strongly based on family ties, therefore successful adjustment to the social environment could be related to the support and advice provided by family members. Women's social networks in my sample were mostly composed of family female members, including the maternal grandmother, which could mean that during stressful events women tended to seek the support and advice of other women within the family. In the case of infants and young children, due to their being in the process of learning about their environment, and acquiring the necessary skills to face stressful events, they could be sensitive to interactions with family caretakers (Belsky et al., 1996; Geary and Flinn, 2001; Geary et al., 2001). In my sample, according to the mothers, infants showed positive behaviours which are positively related to maternal resilience and self-efficacy perceptions and probably to positive interactions with the grandmother and other female family members as well as with caretakers outside the family, such as the teachers from the childcare centres.

Finally, through this analysis, I was able to identify a group of mothers and grandmothers that used food and drinks to relieve distress behaviours in the child. This could be a key

group to focus on, considering that the strategies used could be associated with later inadequate eating patterns and body composition outcomes. However, information about the strategies used by the additional caretakers from the non-grandmother group will be needed to make sure that the grandmother response to child distress is contributing to any differences in fatness between these groups.

7.11 Conclusions

Cortisol concentration analysis revealed that grandmothers' support did not have a statistically significant effect on mothers' and infants' cortisol levels, but women who received support from the grandmother showed a higher concentration. More studies with a more rigorous methodology on hormone measurements will be needed to assess if the maternal grandmother support is related to their daughters' and grandchildren levels of cortisol.

Overall, women perceived themselves as resilient and self-efficacious mothers with low levels of perceived stress, which could be determining their perception about their children temperament, ie, perceiving themselves as a person capable of solving problems and overcoming adversities could be causing mothers to perceive their children as having good temperament, even if this is not the case.

Summary points

- I. No statistical difference was found between the grandmaternal groups in salivary cortisol concentrations.
- II. Women from the grandmother group showed higher levels of cortisol; however more studies would be needed to establish grandmaternal support as a factor causing higher cortisol levels.
- III. No statistical differences were found in the women's behavioural markers of stress between the groups. Overall, women perceived themselves as resilient and self-efficacious mothers with low levels of perceived stress.
- IV. No statistical differences were found in the infants' behavioural markers of stress between the groups. Overall, mothers perceived that their children showed high levels of positive behaviours.

V. Results of infants' temperament need to be carefully interpreted because mothers provided this information and it could be biased considering that their high perception of resilience and self-efficacy may be causing mothers to perceive their children with good temperament, even if this is not the case.

CHAPTER 8

INTERGENERATIONAL ASSOCIATIONS

8.1 Introduction

In this chapter, I aimed to explore associations in the linear growth variables among three generations (maternal grandmother-mother-infant) to address the question of whether the low height z-scores reported in [Chapter 6](#) responded to genetic or environmental factors.

Physical growth is the result of a complex interaction between genes and environment (Tanner, 1978) and therefore, the health profile of a population or a group may reflect both genotype and the quality of the conditions experience during early and current periods.

Evidence has suggested that the environmental conditions experienced by one generation during its growth period can be related to the health, growth and development of the next generation (Emanuel, 1986). Some of these studies have found associations between parental and offspring height (Emanuel et al., 2004) and between maternal height and children risk of stunting and survival (Hernandez-Diaz et al., 1999; Monden and Smits, 2009; Özaltin et al., 2010).

In their theoretical framework for intergenerational associations, Martorel and Zongrone (2012) argue that the pre and postnatal experiences of a mother shape maternal traits (such as height and body composition), which have direct implications for birth characteristics (such as birth weight) and health conditions of children during their first years of post-natal growth.

Hence, mothers' own developmental experience shapes the maternal phenotype to which the offspring is firstly exposed and to which it adapts (Wells, 2003a, 2010a). Therefore, the growth profile attained by a mother and her recent ancestors, such as the

grandmother, may reflect the quality of the conditions experience both *in utero* and during postnatal growth.

8.2 Data

For this chapter, I analysed data from triads comprising the grandmother, mother and infant. The analysis was carried out only for those triads where all the necessary information was obtained (n=48).

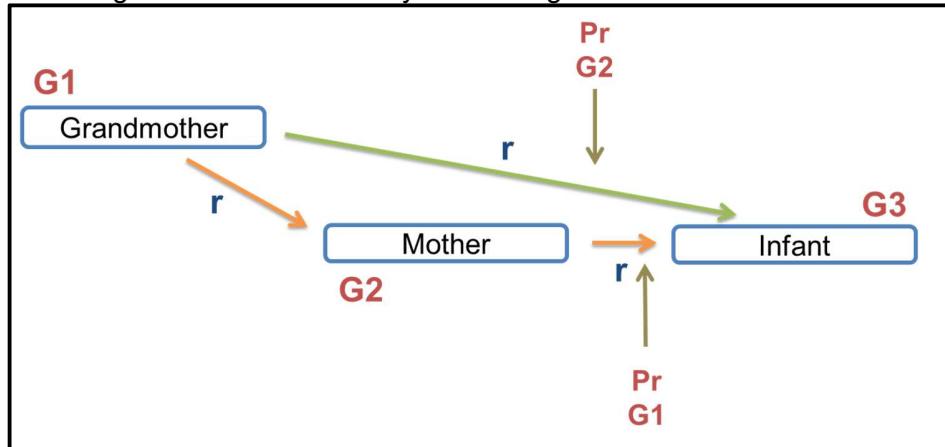
Second, I describe the intergenerational changes in growth status among the generations. Then, I use linear regression analysis to test relationships between linear growth variables (height, trunk and leg length) across the generations. Moreover, I explore maternal and grandmaternal associations with the infants' linear growth. Finally, I analyse the associations of maternal education with offspring growth.

8.3 Statistical analysis

First, I present the information provided by the grandmother and the mother about specific living conditions (family size, number of rooms used to sleep, type of water to drink, availability of a toilet at home, constructions materials of the house, household assets, and parents' occupation) they experienced during their childhood (5 to 10 yrs of age). I tested for differences between mothers and grandmothers using t-test statistic.

My approach to explore intergenerational associations in linear growth variables was based on three levels of analysis. First, bivariate correlations (r) were used to determine the strength of associations of the growth parameters between grandmothers and mothers, mothers and infants and grandmothers and infants. Second, partial correlation (Pr) coefficients were calculated to estimate the proportion of variance of infant's growth indicators attributed to maternal predictors after the variance of grandmaternal predictors were accounted for, and vice-versa (**Figure 8.1**).

Figure 8.1 Levels of analysis of intergenerational associations



G: Generation; r: bivariate correlation; Pr: partial correlation

Third, linear regression models were used to assess the magnitude of the associations between the grandmother's, mother's and infant's height/length and its components. Given the strong associations observed between grandmother's height and mother's height and to be able to independently assess the contribution of each towards the children's height and its components, I derived a maternal height residual, obtained by regressing maternal height z-score against grandmother's height z-score and predicting the residuals. The mother's height residual showed a strong association with mother's height z-score ($r = 0.88$, $p < 0.01$) but no association with the grandmother's height ($r = 0.00$, $p = 0.99$).

To assess the associations between social parameters and grandmother's, mother's and infant's height and its components I used correlation analysis. Finally, I assessed the independent contribution of the grandmother's biological and social variables on the mothers' height and its components using multiple regression analysis. The model was validated by confirming that it met residual assumptions such as normality, variance homogeneity, and independence. Noncollinearity between explicative variables was corroborated by calculating the inflation factors for variance between them.

8.4 Recalled early life conditions

8.4.1 Mothers and grandmothers early life conditions (five to ten years old)

First, mothers and grandmothers were asked about the number of people living in their home during their childhood and the number of siblings present. Forty-six per cent of the mothers recalled having a family size between two to four members and 46% of the grandmothers, between seven to fourteen members. Regarding the number of siblings, 87.5% of mothers recalled having between one and two siblings and 60.4% of grandmothers recalled having between five and fifteen siblings. Descriptive statistics of these variables are presented in **Table 8.1**.

Table 8.1 Mothers' and grandmothers' family size and number of siblings

	Mothers (n=48)				Grandmothers (n=48)			
	Mean (±)	Median (IQR)	Min	Max	Mean (±)	Median (IQR)	Min	Max
Family size	4.6(1.4)	5(1)	2	8	6.7(2.4)	6 (3)	3	14
Number of siblings	1.8 (0.7)	2(1)	1	4	4.9 (2.8)	5 (3)	0	15

Mothers significantly had fewer family members living with them during their childhood, experienced less overcrowding and had fewer siblings (**Table 8.2**).

Table 8.2 Mothers' and grandmothers' family size and overcrowding index experienced during their childhood

	Differences*					
	Mothers		Grandmothers			
	mean	SD	mean	SD	mean	95% CI
Family size	4.6	1.4	6.7	2.4	-2.1**	-2.83;-1.34
Number of siblings	1.8	0.7	4.9	2.8	-3.1**	-3.96;-2.25
Overcrowding index ^a	2.3	1.1	3.7	2.6	-1.4**	-2.17; -0.70

* Grandmother minus Mother

** p<0.01

^a Overcrowding index is defined as the number of people sleeping per room.

Significant differences were found regarding the living conditions of the grandmothers and their daughters (Table 8.3). Most of the studied mothers belonged to families where both mother and father preformed a paid job, while 63% of the grandmothers grew up in families in which the father was the main provider. Regarding the characteristics of the house, mothers grew up in households with better conditions than their mothers did, and all had access to better services (Table 8.3).

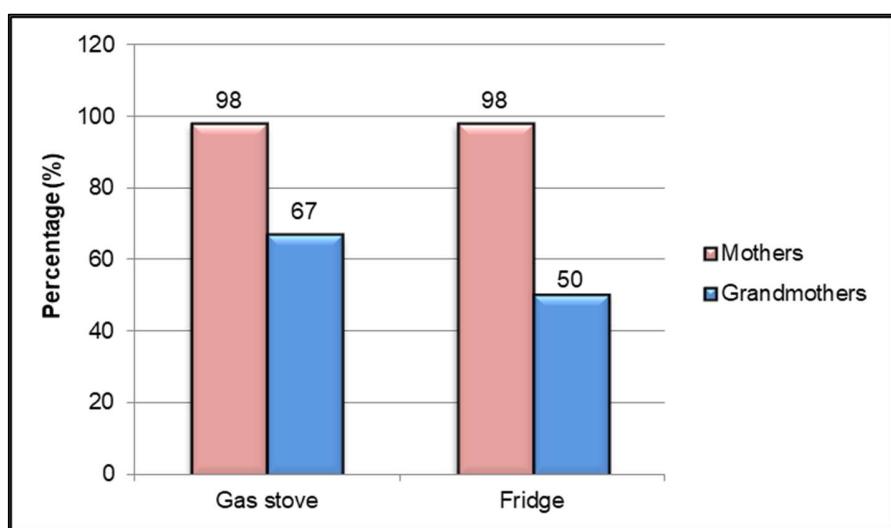
Table 8.3 Living conditions and sociodemographic that mothers and grandmothers experienced during their childhood

Variable	Mothers (n=48)	Grandmothers (n=48)	Differences
<u>Mothers' employment status (%)</u>			
Employees	62.5	17.4	
Employer/Self-employed	12.6	19.6	$\chi^2_{(2)}=20.4, p<0.01$
Housewife	25.0	63.0	
<u>Fathers' employment status (%)</u>			
Employees	70.0	52.1	
Self-employed	15.0	27.1	$\chi^2_{(2)}=3.42, p=0.18$
Employer	15.0	20.8	
<u>Access to piped potable water (%)</u>			
With access	100	56.2	
Wells/Rainwater	---	43.8	$\chi^2_{(1)}=26.8, p<0.01$
<u>Toilet access (%)</u>			
With sinkhole	93.7	60.4	
Latrine*/Backyards	6.3	39.5	$\chi^2_{(1)}=15.1, p<0.01$
<u>House floor (%)</u>			
Durable floor (with cement)	90.0	57.4	
Dirt floor	10.0	42.5	$\chi^2_{(1)}=12.6, p<0.01$
<u>Ceilings (%)</u>			
With blocks and with/without concrete	89.6	58.4	
Other (wood, sheet metal, cardboard)	10.4	41.7	$\chi^2_{(1)}=12.2, p<0.01$
<u>Walls (%)</u>			
With blocks and with/without concrete	100	52.1	
Other (wood, sheet metal, cardboard)	---	47.9	$\chi^2_{(1)}=18.6, p<0.01$

*Latrine refers to a facility used as a toilet, generally a trench (without a bowl) built in the backyard for familial use.

Concerning the ownership of household assets that could be considered important for their health, women were asked if during their childhood the family owned a fridge and a gas-cooking stove. Differences were found between grandmothers and their daughters (**Figure 8.2**), with the mothers' families probably having greater economic capacity to purchase these assets.

Figure 8.2 Differences between mothers and grandmothers in the ownership of household assets



Differences were found between mothers and grandmothers:
 Fridge: $\chi^2_{(1)} = 28.6$, $p < 0.01$ and Stove: $\chi^2_{(1)} = 16.1$, $p < 0.01$.

Moreover, mothers and grandmothers were asked about their access to medical services during their childhood. All the mothers and 90% of the grandmothers recalled that when they were five years old and fell sick, their parents or other relative took them to the doctor. They did not remember if their families had access to public health services, however, they were sure about receiving medical care during their childhood. According to the results, a high percentage of the mothers had access to this service and this represented a higher level of access compared to the grandmothers (Fisher's exact $p = 0.03$).

Finally, mothers and grandmothers were asked about their total years of education and if they performed a paid job during their childhood. The average total duration of education of the grandmothers was $11.5 (\pm 4.2)$ years, which was lower in comparison to their daughters ($\tilde{x} = 16.2 \pm 2.4$) ($z = -5.47$, $p < 0.001$, Wilcoxon rank sum). Only five per

cent of the mothers and 14% of the grandmothers reported performing a paid job when they were between 5 and 13 years of age. They worked in supermarkets, cleaning houses, as nannies, or making tortillas for sale.

Overall, I observed a strong change in the social factors between grandmothers and mothers, whereby during their respective childhoods, the mothers had completed more education, experienced less overcrowding, had fewer co-living siblings and had better household conditions.

8.5 Intergenerational changes in growth conditions among generations

8.5.1 General description of the data

Table 8.4 presents the main characteristics of the 48 triads included in this analysis. There are clear patterns observed in this table. There are marked differences in the biological characteristics between grandmothers and mothers, whereby mothers are taller and have longer trunks and legs. Moreover, when compared the mean z-scores of the growth variables between the three generations, show significant differences. Infants present better growth conditions than mothers and grandmothers (**Table 8.4-Differences 3G**).

Furthermore, mothers had their first pregnancy on average at an older age compared to their own mothers. Interestingly, no differences were found between mothers and grandmothers in the relative contribution of height components (sitting height, and leg length) to the overall height and the ratio between trunk height and leg length. In this sample, for both mothers and grandmothers, trunk height provided a greater contribution than leg length. This indicates a significant secular trend in growth between these two generations.

Table 8.4 Biological characteristics of the sample

	Children		Mothers		Grandmothers		Differences Grandmother – Mother		Differences 3G [#]
	mean	SDI	mean	SD	mean	SD	mean	95% CI	
Age (years)	1.9	0.2	30.0	4.8	56.9	6.9	-27.0**	-28.4; -25.6	--
Age at 1st pregnancy (years)	--	--	27.1	5.0	23.4	3.2	3.67**	2.24; 5.09	--
Height/Length (cm)	85.7	3.5	155.3	5.7	149.6	5.9	5.70**	3.96; 7.43	--
Height/Length (z-score)	-0.2	0.9	-1.2	0.9	-2.1	0.9	0.87**	0.61; 1.14	$\chi^2_{(2)} = 64.7$ p<0.01
Trunk length (cm)	52.0	1.8	82.5	2.7	79.5	3.1	2.94**	2.02; 3.87	--
Trunk length (z-score)	-0.3	0.7	-1.8	0.7	-2.6	0.8	0.77**	0.53; 1.01	$F = 118.6$ p<0.01
Leg length (cm)	33.8	2.1	72.7	4.3	70.0	3.7	2.73**	1.45; 4.02	--
Leg length (z-score)	-0.4	0.9	-1.2	1.0	-1.9	0.9	0.68**	0.36; 0.99	$F = 29.7$ p<0.01
Trunk length/leg length ratio	1.5	0.1	1.1	0.1	1.1	0.1	0.00	-0.02; 0.02	--
Relative trunk length index	60.6	1.2	53.2	1.4	53.2	1.2	-0.04	-0.50; 0.42	--
Relative leg length index	39.4	1.2	46.8	1.4	46.8	1.2	0.04	-0.42; 0.50	--

#Differences 3G: Grandmother – Mother – Infant

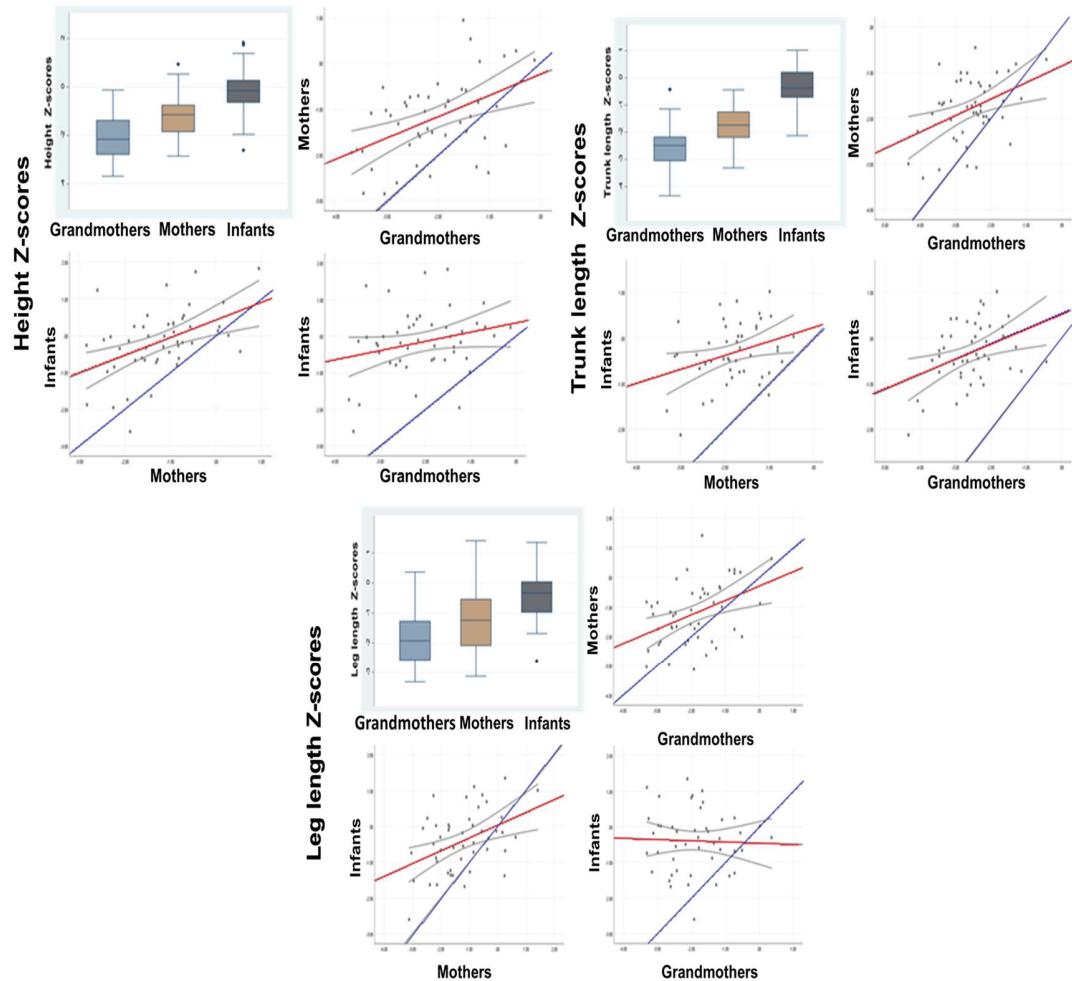
* p<0.05

** p<0.01

^a Overcrowding index is defined as the number of people sleeping per room.

Overall, the trend of the growth parameters between the generations (grandmothers-mothers, mothers-infants, and grandmothers-infant) was positive, indicating, in general, that taller grandmothers had taller daughters who have taller children (Figure 8.3).

Figure 8.3 Secular changes and linear associations of: a) z-score for height, b) z-score for trunk length, and c) z-score for leg length by generation



Secular changes in height and body proportions between the generations were first analysed through box plot graphs, to compare group mean values. Then, I analysed linear associations of growth parameters across the grandmother/mother/infant triad using scatterplots with regression lines. The blue line is the line of identity and the red line is the linear regression slope with 95% confidence intervals. Overall, the secular trends were positive, showing that mothers have better growth conditions than their mothers, and infants, show better conditions than their mothers. Note that all data are converted to z-scores, however the children's mean z-scores are not directly comparable with those of the mother and grandmother as the reference population is not the same.

From **section 9.1** and **9.2** of this chapter, it is clear that there was a strong change in the social and biological factors between grandmother and mothers. There is evidence from many studies that components of maternal phenotype (physiological and behavioural) impact offspring outcomes, such as birth weight and adult height. Moreover, there is evidence that living conditions and social inequalities shape health outcomes.

In this sample, grandmothers experienced poorer living conditions during their own childhood that are reflected in their poor adult health outcomes (short stature). As these conditions can perpetuate across generations, I was expecting to find poor nutritional condition of the mothers, due to their own mothers' exposure to adversity. However, this was not the case and it is possible that social improvements led to positive biological effects across generations.

In the following sections I analysed potential biological and social predictors of the secular trends in the daughter's and infant's height. Regarding social factors, I focused on grandmother's years of education.

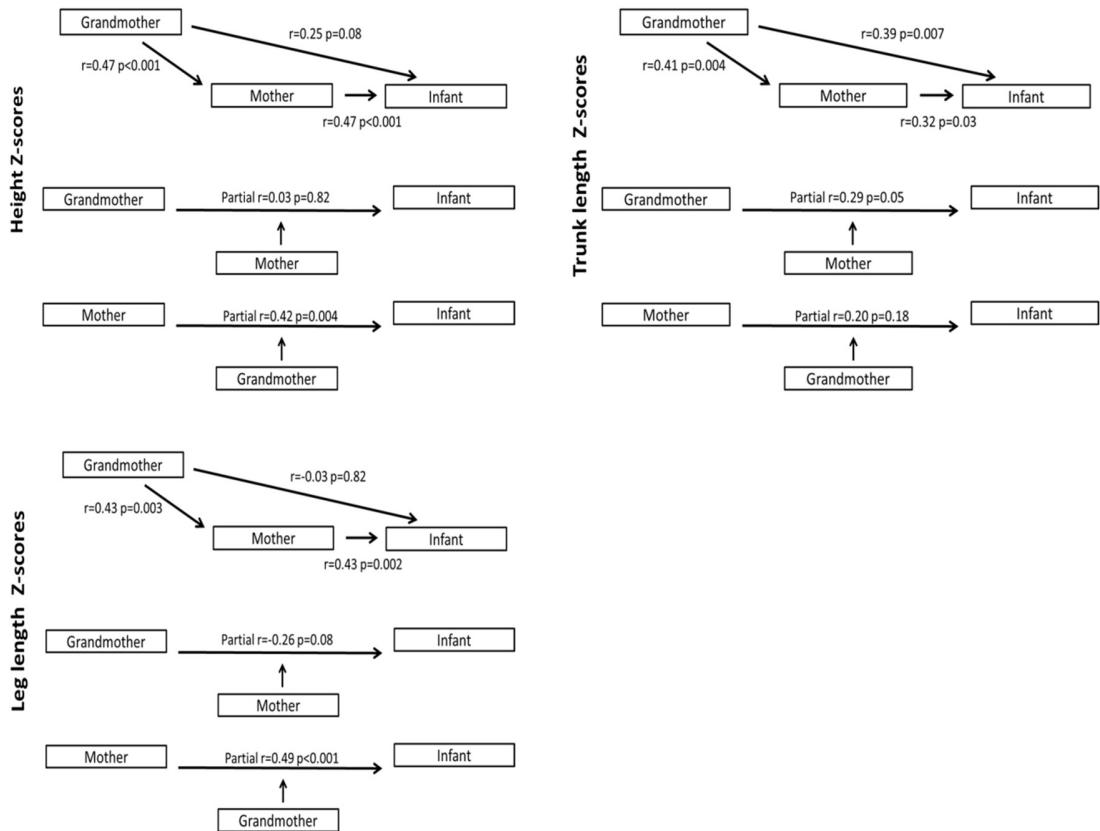
8.5.2 Maternal contribution to height and its components (trunk and leg lengths)

8.5.2.1 Correlations of the growth parameters among the triad

Bivariate correlations were obtained to determine the strength of associations of the growth parameters between grandmothers and mothers, mothers and infants and grandmothers and infants, and partial correlation coefficients were calculated to estimate the proportion of variance of infant's growth indicators attributed to maternal predictors after the variance of grandmaternal predictors was accounted for, and vice-versa.

Presented in **Figure 8.4** are bivariate and partial correlation coefficients of the growth parameters across all three generations. Bivariate correlations between grandmothers and mothers and mothers and infants were strong and significant. Between grandmothers and infants, grandmothers' trunk length was the stronger predictor of infants' trunk length. After accounting for grandmothers' growth predictors, maternal overall height and leg length were the strongest predictors of infants' height and leg length, respectively. As with the simple bivariate correlations, grandmothers' trunk length still remained as the stronger predictor of infants' trunk length after accounting maternal variance.

Figure 8.4 Bivariate and partial correlations in growth parameters between generations



Bivariate correlations were used to determine the strength of associations of the growth parameters between grandmothers and mothers, mothers and infants and grandmothers and infants. Partial correlation coefficients were then calculated, to estimate the proportion of variance of infant's growth indicators attributed to maternal phenotype after the variance of grandmaternal phenotype was accounted for, and vice-versa. The 'r' is the value of the bivariate correlation and the 'Pr', the value of the partial correlation. Overall, bivariate correlations between grandmothers and mothers and mothers and infants were strong, positive and significant. The partial correlations showed that maternal overall height and leg length were the strongest predictors of infants' height and leg length, after accounting for the grandmaternal variance.

Finally, the persistent negative association of the grandmothers' LLZ on the infants' LLZ, led me to consider the negative maternal effects on postnatal growth. Published quantitative genetic models (Ezard et al., 2014; Prizak et al., 2014; Kuijper and Hoyle, 2015) have shown that the maternal effects on the population fitness are sensitive to environmental changes. In stable environments, negative effects maximized fitness by minimizing the phenotypic variance. However, the contributions of two generations in stable environments are not uniform. According to Prizak et al (2014), when parental effects are positive, grandparental effects operate in an antagonistic way to constrain the phenotypic variance and ensure fitness.

8.5.2.2 Height and height component relationships between three generations

Table 8.5 presents the relationships, obtained using linear regression analysis, between height and height components in the triad. I observed that within one generation, i.e. between grandmother and mother or between mother and child, maternal height was strongly and positively associated with the height of her offspring and its components. These associations were less evident across a span of two generations, namely between grandmother and grandchild, where only the grandchild's trunk height was significantly associated with the grandmother's height.

I substituted maternal height with its components to assess their independent contribution. I observed that maternal leg length was positively and significantly associated with the offspring's height and leg length, ie in the next generation. I also observed a significant association of maternal trunk height with offspring trunk height within one generation, between grandmothers and mothers. I did not observe any significant associations between grandmother's height components and their grandchild's height or height components.

Table 8.5 Linear regression analysis between height and height components in the triad

	Height			Grandmother			Leg Length			mean VIF
	β	95% CI	r^2	β	95% CI	β	95% CI	r^2		
Mother Total Height	0.46**	0.20; 0.72	20.8%		0.14	-0.20; 0.48	0.40**	0.11; 0.70	20.6%	1.31
Mother Trunk Height	0.32**	0.11; 0.53	15.7%		0.30*	0.02; 0.57	0.12	-0.12; 0.36	15.2%	1.31
Mother Leg Length	0.41*	0.08; 0.73	10.5%		-0.09	-0.51; 0.33	0.53**	0.17; 0.88	14.8%	1.31
Mother										
	Height			Trunk Height			Leg Length			mean VIF
	β	95% CI	r^2	β	95% CI	β	95% CI	r^2		
Child Total Length	0.48**	0.22; 0.75	20.8%		0.15	-0.21; 0.52	0.37**	0.13; 0.61	18.9%	1.10
Child Trunk length	0.33**	0.12; 0.53	17.2%		0.23	-0.05; 0.50	0.18	-0.01; 0.36	13.8%	1.11
Child Leg Length	0.36*	0.09; 0.63	11.8%		-0.05	-0.41; 0.30	0.37**	0.13; 0.61	15.4%	1.11
Grandmother										
	Height			Trunk Height			Leg Length			mean VIF
	β	95% CI	r^2	β	95% CI	β	95% CI	r^2		
Child Total Length	0.25	-0.04; 0.55	4.24%		0.21	-0.18; 0.60	0.11	-0.23; 0.44	2.36%	1.31
Child Trunk length	0.31**	0.11; 0.52	16.4%		0.24	-0.02; 0.51	0.15	-0.08; 0.38	14.9%	1.31
Child Leg Length	-0.04	-0.33; 0.25	0.15%		-0.02	-0.41; 0.36	-0.02	-0.35; 0.31	0.15%	1.31

Highlighted in red are shown the outcomes variables and in blue, the predictors. Each line represents an independent linear regression.

* p<0.05

** p<0.01

8.5.2.3 Mother and grandmother combined contribution to children's height and height components

Table 8.6 shows the combined mother and grandmother associations with the child's height. Overall, the mother's height was consistently and independently associated with the child's length and its components. In contrast, I observed that the grandmother's height was independently associated with the child's height and trunk length, but not leg length, but only the association with child's trunk length reached significance.

Table 8.6 Maternal contribution to infants' height and body segments

	Grandmother Height		Mother Height residuals ^a		r^2	mean VIF
	β	95% CI	β	95% CI		
Child Total Length	0.25	-0.02; 0.52	0.36**	0.12; 0.60	19.1%	1.00
Child Trunk Length	0.32**	0.12; 0.51	0.18*	0.01; 0.35	22.6%	1.00
Child Leg Length	-0.04	-0.30; 0.23	0.36**	0.13; 0.60	15.0%	1.00

* p<0.05

** p<0.01

^a Mother height residuals are defined as the standardized residuals that result from the regression of grandmothers' height against mothers' height, both as z-score values. These residuals represent the part of the mother's height that is not explained by the grandmother's height. Each line represents an independent linear regression

8.6 Social determinants of height and its components

In **Table 8.7** I describe the associations of height and its components with indicators of wealth during childhood and the duration of education, within one generation i.e. between grandmother and mother or between mother and child, and over two generations, i.e. between grandmother and grandchild. I observed that more years of education was positively associated with overall height and trunk height across one generation and across two generations; however only those associations observed between grandmothers and mothers reached statistical significance. Moreover, education duration was positively associated with leg length within mothers.

The grandmothers' number of siblings was inversely associated with the height and height components of their offspring, but only the height and leg length associations reached significance. The mothers' and grandmothers' number of siblings did not seem to be associated with infants' growth parameters.

The grandmother's experience of overcrowding during childhood was negatively associated with height and trunk height of their offspring and grandchildren, but only the trunk height association for their daughters reached significance. The mother's experience of overcrowding in childhood did not seem to be associated with overall height or trunk height and leg length of the children.

Table 8.7 Contribution of social variables to the overall height and its components within generations

Mother	Child		
	Height	Trunk Height	Leg Length
Years of education	0.15	0.22	0.00
Number of siblings	-0.13	-0.20	-0.03
Overcrowding index	-0.01	-0.10	0.10
Mother			
Grandmother	Height	Trunk Height	Leg Length
Years of education	0.40**	0.36*	0.31*
Number of siblings	-0.33*	-0.18	-0.35*
Overcrowding index	-0.25	-0.48**	-0.01
Child			
Grandmother	Height	Trunk Height	Leg Length
Years of education	0.11	0.20	0.01
Number of siblings	-0.07	0.02	-0.11
Overcrowding index	-0.22	-0.30	-0.04

Highlighted in red are shown the outcomes variables and in blue, the predictors. Each line represents an independent bivariate correlation.

* p<0.05

** p<0.01

8.6.1 Independent contribution of education

Grandmother years-of-education maintained a constant significant association across the daughters' growth variables (height and its components), with higher association with total height. Hence, I was interesting in explore if the significant association with education remained after accounting for grandmothers height. **Table 8.8** presents the results of the independent associations of grandmothers' height and years of education with their daughter's height and its components. Overall, a greater number of years in education showed a small but positive association with the mother's height and its components, but only reached significance for overall height.

Table 8.8 Contribution of grandmothers education to their daughters overall height and its components

	Grandmother					
	Height		Education		r^2	mean VIF
	β	95% CI	β	95% CI		
Mother Total Height	0.39**	0.14; 65.4	0.06*	0.01; 0.12	28.7%	1.06
Mother Trunk Height	0.29**	0.09; 0.49	0.04	0.00; 0.08	23.4%	1.06
Mother Leg Length	0.34*	0.00; 0.67	0.06	-0.01; 0.13	13.6%	1.06

* p<0.05

** p<0.01

Each line represents an independent linear regression

8.7 Summary points

- I. I observed a strong change in the social factors between grandmothers and mothers, whereby during their respective childhoods, the mothers had completed more education, experienced less overcrowding, had fewer co-living siblings and had better household conditions.
- II. There are marked differences in the biological characteristics between grandmothers and mothers, whereby mothers are taller and have longer trunks and legs. Infants present better growth conditions than mothers and grandmothers.
- III. In this sample, for both mothers and grandmothers, trunk height provided a greater contribution than leg length. This indicates a significant secular trend in growth between these two generations.

- IV. Overall, the trend of the growth parameters between the generations (grandmothers-mothers, mothers-infants, and grandmothers-infant) was positive, indicating, in general, that taller grandmothers had taller daughters who have taller children.
- V. Bivariate correlations between grandmothers and mothers and mothers and infants were strong and significant.
- VI. After accounting for grandmothers' growth predictors, maternal overall height and leg length were the strongest predictors of infants' height and leg length, respectively.
- VII. The persistent negative association of the grandmothers' LLZ on the infants' LLZ, led me to consider the negative maternal effects on postnatal growth.
- VIII. Maternal height was strongly and positively associated with the height of her offspring and its components.
- IX. I did not observe any significant associations between grandmother's height components and their grandchild's height or height components.
- X. Overall, the mother's height was consistently and independently associated with the child's length and its components.
- XI. Grandmother years-of-education maintained a constant significant association across the daughters' growth variables (height and its components), with higher association with total height.

8.8 Conclusions

Due to the poor social conditions experienced by the grandmothers during childhood and the poor nutritional conditions reflected in their short adult stature, my results could suggest that:

- I. Improvements in social status could compensate for the lower maternal physical investment due to poor nutritional status.
- II. Better social experience of grandmothers was correlated with better nutritional status in their daughters.

The analysis provided in this chapter showed that although maternal investment can be constrained due to poor growth conditions, improvements in women's status, through education and employment, could improve the well-being of their children.

Overall, health status, characterized by height, and social status, characterized by education were correlated. In this analysis, the grandmother's height and her years of study were positively associated with her daughter's height which, in turn, was the maternal growth indicator that was positively correlated to infants' height.

In infants, no statistically significant associations with maternal social factors were found. However, the high correlation between mothers and infants height and the fact that physical growth is a strong marker of social status indicates that maternal status is reflected in the offspring current conditions. Probably the significance at the statistical level of this association could be seen at older ages.

Overall, by showing through this analysis that mothers are taller than grandmothers, and that this is in part the result of socio-economic improvements, I think I am able to demonstrate that there is the possibility for the population to become taller in association with better living conditions and that could be mediated by maternal capital investment during the first 1000 days of life.

Therefore, although I do not have any data to predict what will happen with the growth of the infants from my sample, this analysis makes the point that environmental conditions can improve across generations, with positive impacts on growth conditions. Hence, the prevalence of stunting in my sample is unlikely to be solely attributed to genetic factors.

CHAPTER 9

GENERAL DISCUSSION AND CONCLUSION

9.1 Introduction

In this chapter, I combine the main findings of my research project to produce an overview of possible pathways or mechanisms by which the grandmother and other members of the maternal social support network influence maternal and childhood outcomes. Next, I describe the strengths and the limitations of the study and finally, I provide a conclusion for my research project.

9.2 Summary of the findings

Contrary to primary hypothesis, the findings reported in [Chapter 6](#) have shown that the presence of and support from the maternal grandmother did not show a statistical effect on their grandchildren's growth outcomes. The study findings also did not support the secondary hypothesis: women's perception of stress, resilience and self-efficacy about motherhood and infant temperament did not differ among the groups. Concerning the cortisol concentrations in saliva, no differences were found in infants, and in mothers, those in the non-grandmother group unexpectedly presented lower levels of cortisol in saliva.

[Table 9.1](#) provides an overview of my findings. Overall, there is not enough evidence to state that in this sample having a maternal grandmother was advantageous in comparison to the group without grandmaternal support. Hence, my results do not support previous findings that suggest that maternal grandmothers might be the principal allomother with beneficial effects on children's nutritional status (Gibson and Mace, 2005; Cunningham et al., 2010; Meehan et al., 2014; Sheppard and Sear, 2016).

Moreover, the results did not provide evidence of advantageous effects of the grandmother on women's and children's physiological and behavioural markers of stress, resilience, self-efficacy and temperament.

However, this also does not constitute evidence that maternal grandmothers are unimportant in the life of their daughters and grandchildren. Hence, in the following sections, I present a summary of the findings stratified by grandmaternal groups and explain the possible reasons why I did not find differences between the groups, as well as the possible pathways through which grandmothers could have beneficial effects.

Table 9.1 Overview of the study findings

No	Hypothesis	Outcome measures	Results
I	Primary hypothesis: children whose mothers receive support from the grandmother are taller and have greater lean mass	Infants length and body composition	No statistical differences among the groups in length-for-age (mean=0.1, 95% CI=-0.4; 0.6). Body composition: Results are not available
II	Secondary hypothesis:	Outcome measures	Results
i	Women who received help by their mothers: d) are less stressed e) have lower levels of cortisol f) have lower fat mass and higher lean mass	Maternal stress scores using a self-administered scale Saliva cortisol levels Body composition	No statistical differences were found among the groups ($z = -0.78$, $p=0.43$) Mothers that belong to the non-grandmother group presented lower levels of cortisol in saliva ($z = -1.88$, $p=0.059$) Results are not available
ii	Women helped by their mothers: c) show higher levels of resilience d) show higher levels of satisfaction and self-efficacy about motherhood	Maternal resilience scores using a self-administered scale Maternal satisfaction and self-efficacy scores using a self-administered scale	No statistical differences were found among the groups. Overall, women showed high levels of resilience (median=3.4, IQR=3.2-3.7) with higher scores in strength and self-confidence (median=3.6, IQR=3.2-3.7), family support (median=3.4, IQR=3.2-3.7) and social support factors (median=3.4, IQR=3.2-3.7). No statistical differences were found among the groups ($z = -0.81$, $p=0.41$, Wilcoxon rank sum). Overall, women show high levels of self-efficacy and maternal satisfaction (median=7.0, IQR=2.0).
iii	Infants who receive care from their grandmothers: c) are less stressed d) have lower levels of cortisol	Infant temperament using a self-administered questionnaire filled by the mother Saliva cortisol levels	No statistical differences were found by grandmaternal group in the three temperament factors (Negative affectivity: $z = 0.96$, $p=0.34$; Surgency/Extraversion: $z = 1.72$, $p=0.08$ and Regulatory capacity: $z = -0.07$, $p=0.95$). Overall, according to the mothers, infants showed a higher frequency of temperament behaviours related to extraversion and regulatory capacity. No statistical differences among the groups ($z = -0.60$, $p=0.56$)

9.2.1 Mothers and infants with grandmaternal support

Although I was not able to find clear benefits from the presence of the maternal grandmother on children's nutritional conditions, as I stated before, this does not constitute evidence of the potential unimportance of maternal grandmothers. First, the patterns of contact frequency in the sample reveal that support from the grandmother is sought and preferred by mothers and this could tell us about the close relationship between these family members and the benefits of this relationship through the provision of emotional support and advice that mothers reported receiving from them. I am aware that this pattern was established as a selection criterion, however, this was established according to the information that the population gave me during the pilot studies, ie, it was a criterion based on reality. These findings are consistent with two studies carried out in Mexico in which it was reported that maternal grandmothers are the preferred caregivers (Partidas, 2004; Gutiérrez-Carbajal et al., 2019).

Moreover, among my results, I found that grandmothers are a source of support when mothers needed help in decision-making at home and acted as a source of information during pregnancy, delivery and first days of life of their grandchildren. Women reported that during the *cuarentena* (postpartum period), their mothers helped them with their babies and supported them with household chores, which is consistent with a previous study that stated that the family represents a source of instrumental and emotional support during this period (Clark, 2001).

Nevertheless, not all social relationships are completely benign, and this includes the relationship with the maternal grandmother. Counter-intuitively, women who did not have grandmaternal support showed a more favourable attitude towards breastfeeding and breastfed their children for longer. Overall, advice and concerns of the grandmothers about breastfeeding may reflect cultural beliefs and practices that do not protect breastfeeding, as stated in other populations (El Safy et al. 2016, summary available at http://journals.lww.com/jpgn/Citation/2016/05001/ESPGHAN_49th_ANNUAL_MEETIN_G_of_the_European.1.aspx). Additionally, grandmothers could identify breastfeeding as a barrier to bonding with their grandchildren and as an obstacle to their daughters getting adequate rest (Grassley and Eschiti, 2008) and hence, they could perceive formula feeding as key to overcoming these barriers.

Although I need more detail data about grandmother's beliefs and attitudes about breastfeeding, as well as information about their own breastfeeding experiences to further develop and interpretation of this finding, the lack of adequate knowledge and misinformation from the grandmother could be considered potential factors that influence the breastfeeding patterns of their daughters. Hence, this could be a key group for potential interventions considering that previous reports have found that counselling sessions with the grandmother and their encouragement and enthusiasm about breastfeeding positively influence feeding patterns (Mahoney and James, 2000; Ingram et al., 2003; Nunes et al., 2011).

Concerning the opposite results in the cortisol saliva concentrations, there are potential factors that must be taken into account to understand why the presence of the grandmother could potentially increase stress in their daughters. First, the quality of the relationship between the partners and grandmothers might be affected, especially in those cases in which the grandmother cohabited with the family. Second, a conflict between mothers and grandmothers over different styles of parenting could elevate stress in the mothers. During the interviews, some women showed concern that the grandmother was spoiling the child, which made me think that children who are cared for by the grandmothers might behave worse and show lengthy periods of whining or tantrums that potentially may cause stress to the mothers. Nevertheless, according to the infant temperament evaluation made by the mothers, the children showed positive behaviours and did not differ between the two groups.

Therefore, other factors, apart from the maternal grandmother, could be relevant. First, cortisol levels could vary depending on the events and concerns that women experience in their day-to-day lives and the activities related to each day (Flinn and England, 1997; Nepomnaschy and Flinn, 2009). Second, I studied a group of women that are working mothers that, despite having support from the partner, are the primary caretakers of the children and the house, which could represent an energetic demand and a source of stress. This result is consistent with other studies carried out in Mexico and with the report from the last Mexican National Survey on Time Use (ENUT 2014), that showed that although men are showing more participation, Mexican women continue having the major responsibility of housework and childcare (Casique, 2000, 2008, ENUT 2014 available at: <https://www.inegi.org.mx/programas/enut/2014/>).

Although the physiological measurement of stress in mothers showed a potential negative effect of the grandmother's presence and support, the behavioural markers told another story. Overall, women perceived low levels of stress, perceived themselves as capable of overcoming adversity (resilience) and showed high levels of maternal self-efficacy and satisfaction. My findings not support the hypothesis that the presence of the maternal grandmother is a determinant of women's levels of perceived stress, resilience and self-efficacy. However, as an important part of the maternal family support system, according to the results in **Chapter 5**, they could promote the skills and self-esteem needed to overcome stressful events and have a positive impact on the way their daughters perceived themselves as mothers. This is consistent with the literature regarding how the family, as an important source of support, can promote the capacity to overcome adversity (Seccombe, 2000) and exert a protective function against stress and depression primarily through the mediation of self-efficacy (Cutrona and Troutman, 1986; Farkas-Klein, 2008; Farkas and Valdés, 2010).

Moreover, these positive behaviours were negatively correlated with women's stress perception, which is also consistent with the literature reporting that resilient individuals can successfully adjust to their environments despite challenging circumstances (Masten et al., 1990; Suárez, 1996), and that women with greater parenting self-efficacy experience fewer symptoms of psychological distress (Cutrona and Troutman, 1986; Halpern and Mclean, 1997; Haslam et al., 2006).

Regarding infants behaviour, women perceived that their children showed more positive behaviours (related to regulatory capacity), which was related to the women's high perception of resilience and self-efficacy as mothers. Considering this finding, although some mothers were worried about the grandmothers' permissiveness of children's behaviour, the way women respond to child behaviours could be making the difference. In this sample, women with positive psychological resources (resilience and self-efficacy) probably were able to develop the abilities to have beneficial interactions with their children, ie, they could successfully manage negative behaviours in their children, which may be causing mothers to perceive their children as having good temperament, even if this is not the case.

However, in contrast to their daughters, a higher percentage of grandmothers used food and drinks to relieve distress behaviours in the child. This could be a key issue to address, considering that the strategies used could be associated with later inadequate eating patterns and body composition outcomes.

9.2.2 Mothers and infants without grandmaternal support

While the women of the grandmother's group were engaged in more contact with their mothers and were provided with emotional support and advice from them, women without this regular interaction were able to find other sources of support within their own family. Interestingly, they found closeness with other female relatives (aunts, cousins, sisters, stepmothers, mothers-in-law and sisters-in-law) that could be compensating for the lack of frequent contact with, or total absence of, the maternal grandmother. This finding is consistent with kinship studies that found that in the absence of grandmothers, other female relatives can assume a maternal role (Thornton, 1991; Davidson, 1997; Bastida, 2001; Ellingson and Sotirin, 2006).

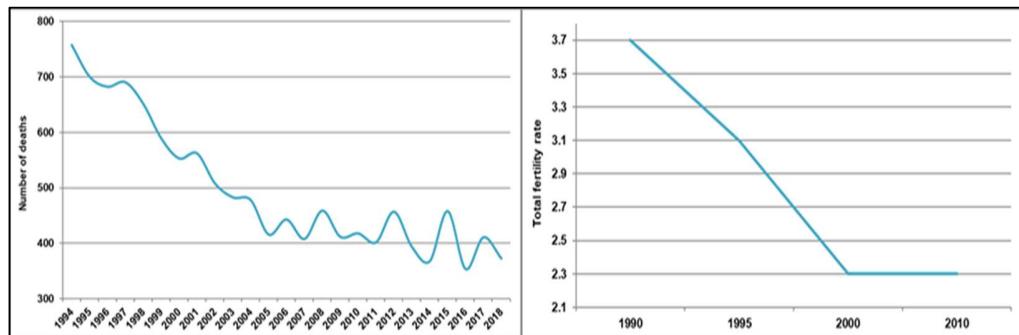
The previous finding could potentially explain the lack of differences in nutritional outcomes among the groups. There is evidence that the presence of aunts is associated with increased infant survival (Heath 2003) and that maternal kin may benefit child height (Shepard and Sear 2016). Moreover, Meehan's (2014) findings revealed that social networks can buffer the absence of an important allomother, with positive effects on children's weight and height, especially in a matrilocal context. Hence, the investment of other female relatives, especially maternal relatives, appears to be a beneficial strategy for this group.

Clearly, my sample differs in either having or not the support of the maternal grandmother. Since the outcomes of the two groups were nearly identical, and since women in the grandmother group identified their mother as a primary source of support, it is very unlikely that the grandmothers provide no useful help. Instead, the most likely explanation for my null findings is that women without grandmaternal support substituted the absence of their mothers with other sources that could provide similar help. My research provides data about this strategy and the potential benefits of expanding social networks.

However, it could be important to consider that other characteristics of the Yucatecan population could be associated with the lack of differences between the groups. First, the population from which my sample comes is undergoing significant changes in terms of mortality and fertility rates (**Figure 9.1**) as well as experiencing epidemiologic and nutrition transitions.

In my sample, differences were observed between the grandmother and mother generations in terms of number of siblings. Grandmothers reported having a mean of five siblings (± 3) while mothers reported having two siblings (± 0.7). Although one of the inclusion criteria of the study was that women were first time mothers, I gathered information about family planning and 57% of women reported that they planned to become pregnant after agreeing with their partners and feeling that they were experiencing a 'stable stage' in their lives. More information would be needed to explain the differences in the number of siblings between the generations as well as data about the potential factors related to family planning that can be associated with this result.

Figure 9.1 Yucatan infant mortality rate (1994-2018) and total fertility rates (1990-2010)



Source: National Institute of Statistics and Geography (INEGI)

Moreover, contemporary Yucatan children experience a lower frequency of infectious diseases than in previous decades. Fifty per-cent of the mothers reported that their children became ill from an infectious disease between one and two times in the last month before the first home visit. The type of infection is presented in **Table 9.2** and the most frequent were the respiratory tract infections.

Table 9.2 Frequency of infectious diseases in infants of the sample

	With GM (n=28)	Without GM (n=17)	Total (n=45)
Vomiting and diarrhoea	17.8	11.8	15.5
Respiratory tract infections	71.4	70.6	71.1
Not specify	10.7	17.6	13.3

This information is relevant considering that the activation of the immune system demands the use of energy and if the exposure to infections is frequent, the demand for energy will be continuous, leading to potential growth deficits. However, in my sample, the frequency (1-2 times in a month) could be considered low which could indicate that children are less exposed to infectious agents probably to the adequate levels of sanitation experienced at their homes (access to basic services and good household conditions, see Table 4.4). If the energy cost of infections is lower, then the potential benefits of GM support for promoting infant growth might also be reduced.

Considering the previous information, I am exploring the effects of the grandmothers in an ecological context that is very different from the contexts in which positive and significant effects have previously been found between grandmothers presence and child growth and nutrition (Gibson and Mace, 2005; Sheppard and Sear, 2016). This means that other health indicators should also be explored, for example extending to the overweight component of nutritional status.

In my study, for example, there was evidence of the ‘double burden of malnutrition’ (Varela-Silva et al., 2012). This phenomenon, where individuals may show markers of either undernutrition or overweight or both, is seen mainly in developing countries, in which the population is undergoing nutritional transition. Data indicate that this transition is occurring in the Yucatecan population, leading to an increase in the consumption of high fat-sugar diets (Azcorra et al., 2013; Bogin et al., 2014) along with a reduction in energy expenditure (Bogin et al., 2014).

In this sense, it could be harder to detect grandmaternal effects in a population experiencing both under- and over-nutrition so I need to be very careful when choosing indicators to evaluate nutritional conditions in my sample.

In my results, I presented BMI values to evaluate the risk of excess weight in my sample. However, BMI has three relevant limitations; first, this index is not independent of stature. Second, people of the same height can have different BMI values according to their body proportions (eg relative leg length) and third, BMI is not a reliable estimator of body composition (amount of lean tissue and fat tissue) (Garn et al., 1986). Moreover, previous studies have shown that the height denominator of BMI could be important as well as the weight nominator. For example, Wells and Cole (2014) reported that taller and faster-growing children have elevated cardiovascular risk markers such as insulin resistance, independently of their adiposity.

These BMI limitations were considered when my study was designed and therefore, to obtain more accurate body composition data, I used isotope measurements to calculate total body water (TBW). It was necessary to obtain saliva samples from the mother and urine samples from the child before and after they received an oral dose of deuterium oxide. The analysis of the samples is still in progress and scheduled to be completed by February 2020.

Accurate data on body composition could provide an important new way to define the health conditions in this transitional population, and to explore the potential benefits or negative effects of grandmother support.

Regarding attitudes about breastfeeding, I do not have information to demonstrate that women from the non-grandmother group did not receive advice from their mothers. It is possible that despite the distance (in those cases where the mother lived in another city) women communicated with their mothers and received advice and knowledge from them. However, considering the less favourable results in the duration of exclusive breastfeeding from the other group, their favourable attitude towards breastfeeding could be related to other factors.

First, women without the support of the grandmother could have a higher disposition to exclusively breastfeed their babies for the recommended time. There is evidence showing that women who have the intention (being highly motivated and confident) to exclusively breastfeed, score higher on the IFFAS scale (Ishak et al., 2014). In addition, they might also benefit from accurate information provided by other female relatives. Data about

perceptions of other female family members towards breastfeeding practices could be useful to collect in future studies.

In comparison with the other group, mothers without the support of the grandmothers showed lower concentrations of cortisol. As with the grandmother group, I need to consider that this difference may be due to certain events and concerns experienced during the week and everyday activities (Flinn and England, 1997, 2003; Nepomnaschy and Flinn, 2009). However, in contrast to the other group, the less frequent contact with the mother could potentially not expose them to negative interactions between the grandmother and the partner, nor to conflicts related to different styles of parenting. Moreover, their partners invested more time in childcare during the week, which could relieve some stress in this group of mothers.

Women and infants from this group also showed a high level of positive behaviours, which could be associated with the maternal social structure based on family ties, ie successful adjustment to the environment could be related to the support and advice provided by other family members, such as aunts and sisters. This is consistent with the literature which stated that social support and support provided by the family, can help to relieve the negative effects of stress and promote the abilities needed to overcome adversity (Seccombe, 2000) and with previous studies that reported the closer relationship between siblings when the relationship with a parent is poor (Voorpostel and Blieszner, 2008).

As I stated before, children's behaviour could be sensitive to interactions with different family caretakers (Belsky et al., 1996; Geary and Flinn, 2001; Geary et al., 2001). In my study, infants without frequent contact with their grandmothers showed positive behaviours, which could be related to positive interactions with other female family members and with their mothers through the successful management of their behaviour.

Therefore, the support provided by other members of the family could be protecting mothers and infants from the negative effects of stress and the potential disadvantages related to grandmother absence. Grandmothers are important; however, it seems that women developed the ability to find support from others when their mothers are not available.

Although I did not carry out in-depth interviews to explore in detail the quality of the women's social relationships, my data included indicators of positive aspects of relationships, such as emotional support (Umberson and Karas Montez, 2010), which could help to explain some of the results. Overall, my data suggested that grandmothers are an important source of emotional support and advice to women as well as their partners, while women from the other group relied more on their partners and other relatives, such as female siblings, mothers-in-law, and non-relatives such as friends when needed (see section 5.4.5). This indicates that women from this sample are not socially isolated, and this ability to establish different social bonds and obtain some specific resources, such as emotional support, from them can be related to the lack of difference between the groups in maternal and child health outcomes. In future studies, detailed information about the family environment and quality of social relationships could be relevant (Umberson and Karas Montez, 2010). Data about family cohesion and warmth, the absence of family discord, and having at least one adult who provides social support during childhood (Grossman and Tierney, 1998) are important factors to be considered.

Additionally, it could be important to consider that in future studies, data about family and kinship background could be valuable to contextualize the patterns of support provided by the grandmother. Throughout their history, the Yucatecan population has been characterized by particular patterns of family settlement and social organization. Historical, demographic and archaeological evidence suggests that the organizational structure mostly consisted of a group of large patrilocal families that functioned as a cooperative and production economic units that could live together in the same household (Bracamonte and Robleda, 1996). However, there is also evidence about the population experiencing both patrilocal and/or matrilocal residence patterns. For instance, it has been reported that among Mayan groups it was common for the son-in-law to 'serve' the bride's family for a set time, before or after the marriage. Once the husband had met this requirement, the couple could move to the house of the husband's father. Nevertheless, according to official reports, this 'service' of the son-in-law is no longer a common practice, and the population shows flexibility regarding residential patterns. For example, there is evidence for the possibility of arranging matrilocal or neolocal residence patterns before marriage (Rosado Rosado, available at: http://www.mayas.uady.mx/articulos/muger.html#_ednref1).

In my sample, data on family decisions about residential patterns were not systematically collected. From the 90 families studied, 78% were nuclear families residing separately from both the husband's and the wife's natal households. Twenty-eight per cent were extended families, in which at least one other paternal and/or maternal family member shared the home. In 14 of these 25 extended families, the maternal grandmother was one of the members and some mothers pointed out that the reason why the grandmother was living with them was that the partner spent six days per week working outside the city, and the grandmothers supported them by helping care for the child. On the other hand, none of the participants was living with the paternal grandmothers and only three women from the none-grandmother group reported receiving support from them. I did not collect data from these cases about the role of the paternal grandmother.

Unfortunately, the information described above is the only data available about family structure in this sample. Therefore, due to the flexibility on residential patterns and the importance of family arrangements it will be necessary for future studies to obtain data about these characteristics to know in more detail the role and influence of the grandmothers throughout the life of women and children. Moreover, this information could be useful to explain more about why mothers seek the support of the maternal grandmother in this context.

9.3 Strengths of the study

I provide below an overview of the strengths of the study:

- I. The sample is well-matched by socioeconomic conditions. The similarity in the socioeconomic background among the groups helped me to verify if the differences found among the groups were related to the grandmother's support and/or other factors not associated with socioeconomic factors.
- II. The established criteria for assigning the participants to either group were clearly defined.
- III. The sample is clearly standardized by parity, husband/partner presence and maternal diseases, such as depression, anxiety, and high blood pressure during pregnancy and delivery. Regarding the characteristics of the infants and grandmothers, the selection was also clear and systematic. Only first-born

children between 1.7 and 2.3 years and the biological mother of the mother were part of the study.

- IV. Obtaining data from 2 year-old children was challenging considering the difficulties of this age group. At this age children are starting to reaffirm their independence (i.e. what they like to do or not), hence, I needed to design effective ways to reassure their participation in all the data collection activities. The process was demanding but successful. As far as I know, few researchers tackle this difficult group hence, my data would be considered unusual.
- V. Regarding data collection, the study followed a systematic approach to obtain quality data. This approach was possible due to the extensive training and pilot studies carried out on all data collection aspects.
- VI. The study outcomes from both mother and child enabled me to analyse trends and associations among complex inter-connected factors that may influence early infant growth and maternal health. Particularly, I consider that the study portrayed the complexity of the social relationships characteristic of our species and enabled me to explore the potential pathways through which maternal investment in social capital could be associated with maternal and infant well-being.
- VII. There was a strong link between the framework of life history theory and the results of the study.
- VIII. As far as I know, my study is the first that obtained and described data regarding grandmother's support in the Mexican population and examined the association between this support and the health conditions of the mother-infant dyad.

9.4 Limitations of the study

Some of the limitations of the study have been discussed through the thesis; however, below I present an overview of the most important limitations:

- I. The study has a restricted sample size due to the various activities planned for data collection that needed participants to invest a certain amount of time, which some potential participants were not able to provide because of their daily activities. The sample size may have limited the statistical power to detect differences between the groups, though almost without exception, there was negligible evidence of any such difference. Moreover, since the two groups was

clearly well matched by socioeconomic conditions and had near-zero overlap regarding the key exposure, the frequency of grandmaternal support, increasing the sample is unlikely to help detect differences.

- II. Potential limitations need to be considered to explain the lack of associations between physiological and behavioural markers of stress. First, the lack of certainty that women followed rigorously the instructions for saliva collection. Second, the lack of information about participants' health conditions during the sampling. Third, the lack of information on the women's menstrual cycle, considering that cortisol levels vary according to the phase of the menstrual cycle. Finally, the lack of more than one saliva sample to calculate baseline cortisol levels. However, most of these limitations were due to time restrictions and by the time availability determined by the participants.
- III. Incomplete data for some outcomes due to 10 participants withdrawing from some aspects of the study. From the 80 dyads that decided to continue the data collection, I was not able to collect saliva samples from 9 children. Moreover, from the 52 recruited grandmothers I was not able to obtain anthropometry data from 4 women, and only 45 grandmothers completed the self-administered questionnaire.
- IV. Due to the lack of enough childcare centres to recruit women from different socioeconomic characteristics and incorporate a wide range of socioeconomic variability in the study, my sample is likely over-representative of women that use the service of specific childcare centres and who belong to a similar socioeconomic level. Moreover, using other recruitment methods (Facebook, radio and universities) could have increased any socioeconomic bias.

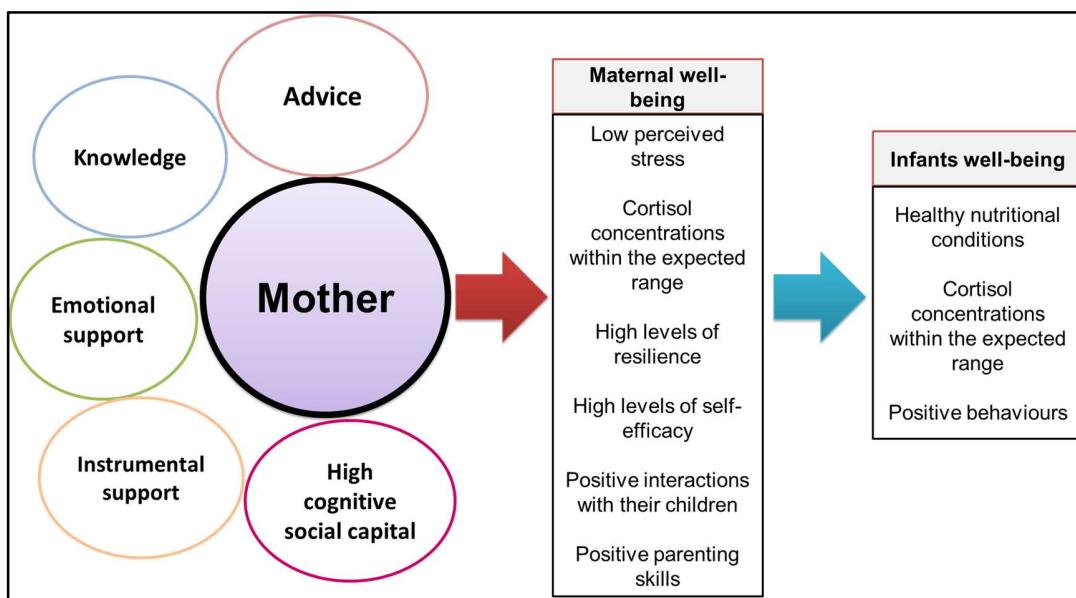
9.5 Conclusion

In the present study, the structure of mother's social networks is strongly based on family ties. Therefore, the lack of differences in the outcomes between the groups could be due to the support and advice provided by different family members, such as the maternal grandmother and other female relatives.

Through the support provided by the grandmother and other female relatives, women obtained knowledge, advice and emotional and instrumental support, factors that could

be influencing their high cognitive social capital perception, ie in their high levels of trust in these relationships (Figure 9.1). Interestingly women were able to use this capital to acquire physiological, behavioural and psychological resources that help them to bear the energetic and psychological demands of motherhood and enable them to provide a benign environment to raise their children, resulting in a healthy infant profile.

Figure 9.1 Perception and benefits of the maternal family support system



According to my results, it seems that the resources provided through the regular contact and close relationship with female relatives were beneficial to women stress response and behaviour, which, in turn, had a beneficial impact on children growth and temperament.

In conclusion, my findings suggest that mothers with greater psycho-social capital have a positive impact on their child's development. Mothers can find different sources of social support for rearing their children, and those lacking a grandmother are not at a systematic disadvantage.

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