Metabolic implications and safety of dolutegravir use in pregnancy

Valeriya Dontsova[1], MSc, Haneesha Mohan[1], PhD, Camille Blanco[1], BSc, Jennifer Jao[2], MD, Prof Nicholas DE Greene[3], PhD, Prof Andrew J Copp[3], DPhil, Rebecca Zash[4], MD, Lena Serghides[1,5,6], PhD.

[1]Toronto General Hospital Research Institute, University Health Network, Toronto, Canada

[2] Northwestern University Feinberg School of Medicine, Chicago, IL, USA; Department of Pediatrics, Division of Pediatric Infectious Diseases

[3]Developmental Biology and Cancer Department, UCL Great Ormond Street Institute of Child Health, University College London, London, UK

[4]Beth Israel Deaconess Medical Center, Boston, USA; Department of Medicine, Division of Infectious Disease

[5]Women's College Research Institute, Women's College Hospital, Toronto, Canada

[6]Department of Immunology and Institute of Medical Sciences, University of Toronto, Toronto, Canada

Corresponding Author:

Lena Serghides

101 College Street, 10-359

Toronto, Ontario, M5G 1L7, Canada

lena.serghides@utoronto.ca

Summary

Dolutegravir (DTG) is recommended for all people living with HIV because of its efficacy, high barrier to resistance, favourable safety and tolerance profile, and affordability. DTG has also been found to have the highest rates of viral suppression in pregnancy, therefore preventing perinatal HIV transmission. In view of these benefits, particularly for pregnant women, it is important to ask whether DTG is safe in pregnancy. DTG has been associated with metabolic complications including weight gain and rare events of hyperglycemia that could affect maternal, fetal, and postnatal health. Here we review the current clinical and experimentally-based literature on the implications of DTG usage for the pregnant female and for the developing embryo and fetus. Possible effects on folate status, energy metabolism, adipogenesis, and oxidative stress are considered. In many instances, insufficient data are available, pointing to the need for additional research in this important area of HIV treatment.

Search strategy and selection criteria

References for this review were identified through PubMed searches, authors' general knowledge of the field, and research papers from presenting authors at HIV conferences. Only papers written in English were included. PubMed searches were the following. Search 1: "dolutegravir.tw" AND ("pregnan*.ti" OR "conception.ti"). Search 2: "dolutegravir.tw" AND ("hyperglyc*.ti" OR "diabet*.ti"). Search was performed for all papers up to October 2022.

1. Introduction

Infection with the Human Immunodeficiency Virus (HIV) poses a severe disease burden, having claimed 36.3 million lives and currently affecting nearly 40 million people around the world.[1] Reducing HIV viral load (VL) in people living with HIV (PLWH) to undetectable and therefore untransmissible (U=U) levels remains the most effective approach at reducing the incidence of HIV infection. The UNAIDS and WHO 95-95-95 goal aims for 95% of PLWH to be aware of their infection status, 95% of people diagnosed with HIV to receive treatment, and 95% of PWLH receiving treatment to have undetectable VLs.[2] Perinatal transmission of HIV remains a serious concern for women of childbearing age living with HIV. The risk of transmission is highest at delivery and during breastfeeding, especially with detectable viremia in pregnancy, preterm delivery, and late initiation of treatment in pregnancy.[3,4] Combination antiretroviral therapy (ART) remains the most reliable treatment option for HIV infection and has been shown to effectively suppress VL, prevent the development of AIDS, and minimize the risk of HIV transmission.[3–5] Pregnant women initiating ART before conception had a 0·03% rate of vertical HIV transmission; the rate drops to 0·0% in the women that additionally had an undetectable VL at conception.[4] As ART has transformed HIV infection into a manageable chronic illness, increased attention has been directed towards optimizing current regimens and understanding chronic HIV-related comorbidities associated with HIV infection and ART, with the goal of improving quality of life for PLWH over the long term.[6]

Dolutegravir (DTG)-containing regimens with varied nucleoside reverse-transcriptase inhibitor (NRTI) backbones have recently become the preferred regimens worldwide and are

63 the WHO-recommended first-line therapy for all PLWH.[3,5,7,8] DTG-based regimens are
64 significantly more affordable compared to other first-line ART regimens, making them
65 favourable in low-middle income countries.[5] DTG also has a high barrier to HIV resistance as
66 DTG-resistant mutations have been shown to reduce HIV fitness.[9] Significantly lower rates of
67 viral resistance have been reported with DTG use and DTG is successfully used as salvage
68 treatment in virological failures.[9,10] In clinical trials, DTG-based regimens had the same or
69 improved efficacy as protease inhibitors (PIs), non-nucleoside reverse transcriptase inhibitors
70 (NNRTIs), and other integrase strand transfer inhibitors (INSTIs).[11–15]

71 In the context of pregnancy, the Safety and Pharmacokinetics of Dolutegravir in Pregnant HIV
72 Mothers and Their Neonates (DolPHIN-1,-2), and International Maternal Pediatric Adolescent
73 AIDS Clinical Trials (IMPAACT) network P2010 protocol (VESTED) randomized controlled
74 trials (RCT), showed that DTG was associated with more rapid and effective viral
75 suppression, compared to efavirenz (EFV), making DTG especially useful in pregnancy
76 wherein rapid viral suppression is essential in preventing perinatal transmission.[16–18] The
77 Pediatric HIV/AIDS Cohort Study Surveillance and Monitoring for ART Toxicities
78 (SMARTT) reported better viral suppression in women receiving DTG-based ART compared
79 to non-DTG ART regimens, without any differences in fetal outcomes.[19]

80 DTG-based treatments are associated with lower toxicity, fewer drug-drug interactions than
81 other ARV classes (PI and NNRTI), and a good tolerability profile, all of which improve the
82 quality of life and regimen adherence among PLWH.[11,20] However, DTG use has been
83 associated with metabolic complications in non-pregnant adults, such as weight gain and rare

events of hyperglycemia.[21,22] In pregnancy, DTG appears to be generally well tolerated; however there are limited studies available that examine metabolic parameters or post-natal outcomes.[16,17,23–26] Here we review available data on DTG safety in pregnancy, as well as clinical and experimentally-derived data of the metabolic effects of DTG. We further discuss how these DTG metabolic effects could impact fetal and maternal health.

2. Safety of DTG use in pregnancy

2.1. Clinical study findings on DTG and pregnancy and birth outcomes

Initial clinical surveillance studies, did not report an association between DTG-based ART and adverse birth outcomes (See Table 1 for a summary of studies).[25,26] In the DolPHIN-2 open-label RCT, recruiting from South Africa and Uganda,, DTG was associated with greater pregnancy and postpartum (puerperium) adverse events compared to the EFV arm, however this was not replicated in other studies (Table 1).[17,18,27] Analysis of data obtained in the Tsepamo study did not show differences in severe pregnancy outcomes, such as preterm birth, small for gestational age, or fetal demise, but an increased occurrence of maternal hypertension and increased intrapartum weight gain in women receiving a DTG versus EFV-based regimen was reported.[28,29] Furthermore, DTG was associated with fewer severe adverse birth outcomes in women with lower BMI.[28] In the VESTED trial DTG was associated with improved gestational weight gain and either similar or lower levels of adverse birth outcomes compared to EFV.[17]

2.2. Clinical study findings on DTG and NTDs

In 2018 the Tsepamo surveillance study in Botswana reported four newborns with NTDs among offspring of 426 women starting DTG at conception (0·94% [95%CI 0·37, 2·4]), compared to a 0·12% [0·07, 0·21] incidence with non-DTG based ART treatment and 0·09% [0·07, 0·12] incidence among offspring of women without HIV.[30] A 2019 follow-up to the Tsepamo study reported a decrease from the initial report in NTDs prevalence amongst DTG treated pregnant women to 0·3% [0·13, 0·69] of 1683 deliveries.[31] More anterior body wall defects (omphalocele and gastroschisis) were also reported in those receiving a DTG-based regimen from conception.[31] In a further update in 2022, 10 NTDs were reported in 9,460 women taking a DTG-based regimen from conception, for a rate of 0·11% [0·06, 0·19], compared to 0.08% [0·04, 0·14] on EFV and 0.07% [0·05, 0·08] HIV-negative pregnancies. This brought the rate of NTDs in the DTG group to the same level that of other ARVs and women without HIV.[32]

Additional studies have reported on DTG and NTDs. Although none of these studies have the sample size of the Tsepamo study, all reported no significant differences in rates of congenital defects (including NTDs) between DTG and other ARVs.[33–36] A summary of their findings can be found in Table 1.

Despite the disappearance of the initial NTD signal, the etiology of the increased rates of NTDs is unknown. The initial signal could have been a matter of chance due to small sample size. Alternatively, there may have been other risk factors present during the 2018-2019 years

125 that had a combined effect with DTG on the emergence of NTDs, such as lower population
126 folate levels or other environmental exposures. However, this remains speculative. In light of
127 the most recent data from Botswana, DTG remains a preferred regimen for its superior
128 efficacy in preventing HIV related mortality and transmission in women of childbearing
129 potential.[37,38]

130

131 **2.3. DTG effects in animal and in vitro reproductive studies**

132 In pregnancy, DTG crosses the placenta and fetal exposure can be significant due to slow fetal
133 metabolism of the drug.[16,26] In reproductive toxicology studies performed in rats and rabbits,
134 supratherapeutic DTG was not associated with fetotoxicity or higher risk for congenital
135 defects (see Table 2).[39] However, in a large fetotoxicity study performed in C57BL/6J mice
136 fed a folate-sufficient diet, a small (was 0·47% (N=150 litters)) but significant increase in the
137 incidence of NTDs was observed at the therapeutic DTG dose but surprisingly not at the
138 supratherapeutic DTG dose (both delivered with therapeutic tenofovir (TDF)/emtricitabine
139 (FTC) backbone).[40] Mice receiving the therapeutic DTG regimen also had increased rates of
140 microphthalmia, bleeding defects, and edema.[40] Supratherapeutic DTG-only exposure from
141 conception in C3H/HeJ mice resulted in one NTD (exencephaly) in 109 embryos from 17
142 litters, along with evidence of neuronal damage and neuroinflammation in the pups of DTG-
143 treated dams (Table 2).[41] DTG-exposure of rat embryos cultured through the period of
144 neurogenesis did not demonstrate teratogenicity, although the design of the study was brought
145 into question, particularly the sample size, DTG penetrance of the embryo, and potential for

DTG metabolite teratogenicity (see Table 3).[42,43] In a cell culture model, DTG has been shown to affect morphogenesis and survival of murine pluripotent and human embryonic stem cells, along with transcript changes of developmental regulator genes (Table 3).[41,44,45] From these studies, it appears that DTG is essentially safe for use in human pregnancy although it may have the potential to affect some aspects of embryonic development. Although adverse developmental effects were observed in cell culture, they were rarer and milder in the in vivo models and largely absent in clinical studies, potentially due to compensation by whole organism homeostatic mechanisms.

3. DTG metabolic effects and implications for DTG safety in pregnancy

Inadequate nutrition and poor metabolic health increase the risk of adverse pregnancy outcomes and congenital defects, poor maternal health outcomes, and contribute to metabolic programming that increases the lifelong risk of poor metabolic health in neonates.[46,47] Studies conducted to characterize DTG's effects on metabolic pathways are summarized below, and implications for maternal–fetal health and avenues for future studies are discussed.

3.1. DTG-associated changes to folate metabolism

Interest in investigating the association between DTG and folate increased following the original report of a higher prevalence of NTDs.[30] A comparison of serum folate levels in women participating in the ADVANCE trial found that folate levels increased in non-pregnant

166 women taking DTG administered with TAF/FTC over 12 weeks, folate levels remained stable
167 in women taking TDF/FTC/DTG, and decreased in women taking TDF/FTC/EFV.[48] In the 26
168 women who became pregnant during the study, folate levels increased slightly in those taking
169 TAF/FTC/DTG or TDF/FTC/DTG and decreased slightly in those taking TDF/FTC/EFV for
170 24 weeks, however, the pregnancy cohort was severely limited in sample size.[48]

171 Animal and in vitro studies examining the effects on dietary folate and folate transport indicate
172 mild effects of DTG at therapeutic plasma levels (see Figure 1).[49–51] DTG has been shown to
173 be a partial antagonist of folate receptor 1 (FOLR1/FRα) in placental cell lines.[49] In the same
174 study folic acid supplementation was able to rescue early DTG-related (100 μM) toxicity in
175 zebrafish.[49] Data from the Zamek-Gliszczynski M et al. study support in vitro DTG
176 antagonism of FOLR1, however, extrapolating quantified in vitro DTG inhibition to in vivo
177 conditions, this effect was deemed not clinically relevant at therapeutic dosage of DTG.[50] In
178 DTG-treated placental explants and placenta cell lines, and in placentas isolated from DTG-
179 treated mice, variable changes to the gene and protein expression of the folate transport and
180 metabolism pathway were observed. DTG treatment of placental cell lines was associated with
181 a modest reduction in expression of reduced folate carrier (RFC) and proton-coupled folate
182 transporter (PCFT), along with a decrease in their transport function.[51] In the Mohan H et al.
183 fetotoxicity study, supratherapeutic DTG treatment was associated with lower rates of fetal
184 anomalies than the therapeutic DTG dose, and concurrently higher levels of fetal folate (fetal
185 folate levels in the therapeutic dose were similar to control), suggesting either potential
186 compensation by increased folate uptake or biphasic effects of DTG on a system interacting

187 with or affecting the folate pathway.[40]

188 It remains unclear whether the dysregulation of folate transport or metabolism by DTG has a
189 clinical impact on human pregnancies. Neither clinical or animal data suggests that DTG
190 reduces folate levels, but there is some evidence suggesting a diminished response to folate
191 through FOLR1 inhibition and reduced folate transport across the placenta following DTG
192 treatment. Therefore, folate insufficiency in pregnancy may exaggerate the effects of DTG and
193 proper folate supplementation should have a protective effect against adverse events. Although
194 fetal folate levels and active placental folate transport cannot be quantified in human
195 pregnancies, studies examining the interaction of maternal folate levels and placental nutrient
196 transporters in women receiving DTG-based ART may provide insight into the etiology of
197 adverse perinatal events.

198

199 **3.2. Metabolic effects of DTG use in clinical studies**

200 Maternal obesity, hyperglycemia, prediabetes/type 2 diabetes mellitus (T2DM), and metabolic
201 syndrome (defined as having three or more of increased waist circumference, blood pressure,
202 plasma triglycerides, fasted blood glucose, and decreased HDL-cholesterol), increase the risk
203 for adverse events in pregnancy and contribute to fetal metabolic programming towards
204 increased risk for poor metabolic health.[46,47]

205 DTG has been associated with weight gain, rare new-onset hyperglycemia, and some studies
206 report an increased risk for diabetes mellitus and metabolic syndrome, described in more detail

below.[21,22,52,532] However, few studies have addressed DTG metabolic effects in pregnancy, and results of clinical trials generally suggest improved pregnancy outcomes for DTG compared to other ARVs, primarily EFV.[23,26] Larger-scale studies are needed to corroborate these results. In addition, studies comparing DTG metabolic effects in pregnancy to those of people without HIV are lacking. DTG-associated transient changes to fasted blood glucose in non-pregnant female mice have been reported.[54] However animal studies investigating maternal glucose homeostasis in pregnancy are yet to be carried out.

3.2.1. DTG-associated weight gain

DTG-based regimens are associated with greater weight gain and in the long-term may contribute to other metabolic complications.[21,53] In both naive and ART-experienced patients, DTG-based regimens are associated with greater weight gain than NNRTI-, PI-, and some other INSTI-based regimens.[15,21,53,55–63] The degree of weight gain varies dramatically in relation to the backbone formulation, demographics, and baseline characteristics of the study participants, with TAF-containing NRTI backbones, female sex, older age, and black race being independently associated with greater risk for treatment-associated weight gain.[15,21,58,64–66] In PLWH with advanced viremia and immune suppression (CD4+ T-cell count <200 cells/mm^3), initiation of ART leads to weight gain as part of the 'return-to-health' phenomenon. Indeed, weight gain among treatment-naive individuals initiating DTG-based therapy is greater than in treatment-experienced PLWH switching to a DTG-based regimen.[56,60,62,64,66–69] Furthermore, poor virological control, adverse events, and slower rate of

228 viral suppression in PI and NNRTI drugs are often cited as reasons for smaller magnitude and
229 rate of weight gain in comparison to DTG-based regimens. However, although some NNRTIs
230 are associated with a slower rate of viral suppression in select studies, the INSTI elvitegravir
231 (EVG) exhibits a similar viral suppression rate to DTG and is associated with similar weight
232 gain to NNRTIs.[59] Furthermore, increased weight gain continues to be an issue in the long-
233 term, as shown in 5 year follow-up studies by Ando N et al. and Bourgi K et al.[58,63] In most
234 retrospective studies, the inclusion criteria include successful viral suppression and high CD4+
235 cell count prior to INSTI-treatment as a way to control for the return-to-health effect.
236 Randomized ART-switch and double-blind RCTs corroborate the weight-gain effects of
237 DTG.[21,61]

238 In pregnancy, women receiving DTG-based ART experienced greater intrapartum weight gain
239 than EFV-based ART, however it was still below the recommended weekly weight for a
240 healthy pregnancy.[17,27–29] Sufficient weight gain during pregnancy reduces the risk of preterm
241 birth, and small, and very small for gestational age neonates; DTG-based regimens therefore
242 appear more favorable.[70]

243

244 **3.2.2. DTG-associated hyperglycemia, T2DM, and metabolic syndrome**

245 Currently, only short-term prospective, case-report, and cross-sectional study data exist on the
246 effects of DTG on metabolic health, partly due to the recent implementation of DTG as a first-
247 line treatment. There is also a significant degree of discrepancy between various studies, with
248 some citing DTG-associated improvement to metabolic parameters[71,72] while others report

249 increased risk for T2DM, metabolic syndrome, and hyperglycemia.[22,52] In an observational
250 prospective study interrogating changes to insulin sensitivity and circulating lipids following a
251 switch from ritonavir-boosted PI to a DTG-containing regimen in patients with stable
252 virological control, DTG was associated with lower IL-6, triglycerides, LDL and total
253 cholesterol, leptin, insulin, and HOMA-IR index.[71] In ART-naive patients, initiating a DTG-
254 based regimen was associated with lower rates of new-onset diabetes at 0·91%, in comparison
255 to those starting a NNRTI- or a PI-containing regimen, 1·37% and 1·50% respectively.[72] Hsu
256 R et al. reported no increased risk of prediabetes or diabetes mellitus in ART naive and
257 experienced patients on different INSTIs, however being on ART was associated with higher
258 incidence of T2DM than the general population, 9–13 versus 6·7 per 1000 person-years.[73] A
259 cross-sectional study examining risk factors (including ART regimen, NRTI backbone, VL,
260 BMI, sex, and lymphocyte count) for developing metabolic syndrome in PLWH receiving
261 ART for ≥6 months in Zambia, reported that DTG-based regimens, compared to PI- and
262 NNRTI-based therapy, were independently associated with doubling of the risk for metabolic
263 syndrome.[52] A national survey of HIV clinicians' perspectives on DTG use for PLWH in
264 Uganda in terms of tolerability and effectiveness, reported favorable outcomes for patients
265 initiating or switching to DTG. However, it was noted that hyperglycemia, insomnia, and
266 decreased libido were some of the side-effects associated with DTG treatment.[74] Case reports
267 of hyperglycemia following initiation of DTG have appeared throughout the literature, where
268 hyperglycemia occurred in patients with normal BMI, experiencing weight gain or loss, and
269 without prior history of insulin resistance.[75–77] Discontinuation of INSTI-based therapy
270 normalized glycemic control in the presented cases and the patients no longer needed anti-

diabetic medication.[75,78] A large-scale surveillance study performed in Uganda reported a greater incidence of new-onset hyperglycemia in PLWH switching to, or initiating, DTG-based regimens than in patients receiving non-DTG-based regimens, 0·47% vs. 0·03% respectively.[22] Furthermore, no association of hyperglycemia with weight gain was observed, as in most cases of hyperglycemia the patients had lost weight.[22] A caveat to the study was that a greater proportion of individuals in the DTG arm were male, older, and on ART for more than 5 years, all of which are risk factors for hyperglycemia.[22] A recent study interrogating adverse-drug events in treatment-experienced and naive patients observed hyperglycemia with an incidence of 2·4% within 13 to 62 weeks of DTG-based regimen initiation.[79] Furthermore, the SPRING-1, SPRING-2, SAILING, SINGLE, and FLAMINGO clinical trials, which assessed the efficacy of DTG, reported hyperglycemia among its adverse drug events; hyperglycemia also appears as an adverse drug event leading to DTG-discontinuation.[79–84]

To date, only one study reports on the incidence of gestational diabetes in patients receiving DTG-based treatment, wherein DTG-based ART was associated with a lower risk for gestational diabetes compared to EFV-based ART.[23] No change to insulin sensitivity was observed in exposed uninfected infants born to women receiving a DTG- versus an EFV-based ART.[24] In the follow-up to the IMPAACT 2010 VESTED study, no differences in maternal or fetal HbA1c between FTC/TDF/DTG, FTC/TAF/DTG, or FTC/TDF/EFV were found.[85]

Taken together, these studies show that DTG is associated with metabolic changes in non-pregnant adults. There remains a gap in knowledge of whether the observed effects in non-

pregnant individuals are replicated in pregnancy and relevant to perinatal outcomes.

3.3. Animal and in vitro studies examining DTG-associated metabolic changes

DTG-associated weight gain and hyperglycemia observed in clinical studies may result from drug induced changes to energy homeostasis at the hypothalamic, tissue, and/or cellular levels. Animal models and in vitro studies using human samples have shown distinct alterations to adipose tissue function and insulin sensitivity, as well as changes to mitochondrial function and oxidative metabolism associated with DTG.[86–90] Studies conducted to characterize DTG's effects on these pathways are summarized below and shown in Figure 1.

3.3.1. DTG effect on the melanocortin system

DTG-associated weight gain and metabolic perturbations may be a symptom of a change to energy homeostasis regulation by the hypothalamus. Many hormones are involved in the regulation of satiety/hunger and energy expenditure and deviations from the physiological baseline in pregnancy may lead to fetal programming affecting metabolic health.

In the Tivicay (DTG) product monograph, DTG was shown to reduce alpha-melanocortin stimulating hormone (α-MSH) binding of melanocortin 4 receptor (MC4R) by 65% at the clinical DTG Cmax. This may shift the anorexigenic/orexigenic balance towards increased orexigenic neural tone, thereby increasing appetite and reducing post-prandial satiety resulting in increased food intake without altering energy expenditure – leading to weight gain.[91] A

potential role of MC4R in development has been sparsely documented, and has not yet been thoroughly studied.[92] Analysis of MC4R binding by various INSTIs revealed a capacity for MC4R antagonism by bictegravir (BIC), cabotegravir (CAB), EVG, raltegravir (RAL), and DTG, with EC50 >100 fold beyond the unbound plasma Cmax for each individual drug.[91] It would be ideal to conduct further studies examining DTG effects on hormones involved in regulation of satiety/hunger and energy expenditure, such as α-MSH, thyroid hormones, cortisol, and leptin.

3.3.2. DTG effects on adipocytes

Data from in vitro and animal studies suggest the DTG is associated with adipose tissue changes that could contribute to a mechanistic understanding of the clinically observed weight gain. White adipose tissue (WAT) has roles in both energy storage and endocrine signaling through adipokine secretion, whereas brown adipose tissue (BAT) contributes to energy consumption through oxidizing free fatty acids and generating non-shivering thermogenesis.[89] White adipocytes secrete leptin, an anorexigenic pro-satiety peptide, and adiponectin, which improves insulin sensitivity. In perturbed metabolic states, such as insulin resistance, WAT tends towards hypertrophy and fibrosis, alongside plasma hyperlipidemia.[87] Cold exposure, fasting, and beta-adrenergic stimulation promote BAT activation and WAT beiging, which are associated with better metabolic outcomes.[89]

Treatment with DTG has been shown to cause changes to adipose tissue composition, function, and signaling.[87,89,90] In simian noninfected subcutaneous and visceral adipose tissue,

TDF/FTC/DTG treatment induced adipose tissue fibrosis and hypertrophy, with increased mRNA expression of the adipogenic peroxisome proliferator-activated receptor gamma (PPAR-γ) and CCAAT/enhancer-binding protein alpha (CEBPα), and decreased mRNA expression of adiponectin.[87] In obese PLWH, increased adipose tissue fibrosis was seen in those treated with INSTI-based rather than non-INSTI-based treatment.[87] In cultured proliferating human adipocyte stem cells and mature adipocytes, standalone DTG treatment at Cmax was associated with mitochondrial dysfunction, increased fibrotic markers, lipid accumulation, and lipogenesis, and decreased leptin and adiponectin secretion, and insulin sensitivity.[87] These findings were replicated by Pickering R et al., wherein DTG reduced leptin and adiponectin signaling in cultured subcutaneous adipocytes, while increasing pro-adipogenic and pro-fibrotic PPAR-γ and collagen-6 transcripts without altering total triacylglycerol (TAG) content in both subcutaneous and visceral cultured adipocytes.[93] Long-term 2-year treatment with TDF/FTC/DTG of SIV-infected macaques was associated with a maintained pro-fibrotic, adipogenic phenotype of subcutaneous and visceral WAT.[90] Interestingly, the emergent WAT phenotype of increased lipogenesis, decreased lipolysis, and insulin resistance seen in the SIV-infected macaques treated with DTG does not co-occur under healthy conditions or under the typical progression of obesity, T2DM, and metabolic syndrome.[87] Characterization of oxidative BAT in cell culture and in vivo models with short-term (<2 weeks) DTG exposure demonstrates a reduction in thermogenesis, adipogenesis, BAT-specific markers, uncoupling protein 1 (UCP1) expression, and insulin sensitivity.[89,90]

If the DTG-associated adipose tissue changes discussed lead to clinically observable changes

354 to circulating adipokines like leptin, body-composition, and whole-body energy expenditure,
355 this may in part explain the weight gain. Therefore, it would be useful to monitor these
356 parameters in patients receiving DTG-based ART. It will be also important to study DTG
357 effects on leptin levels in the context of pregnancy, as leptin is produced by the placenta, and
358 its production is altered in several pathologic conditions including preeclampsia and
359 gestational diabetes.[94,95]

360

361 **3.3.3. Oxidative stress and metal ion chelating associated with DTG exposure**

362 At a cellular level, the decreases in BAT oxidative capacity and WAT insulin sensitivity may
363 stem from altered cellular metabolism – initiating or resulting in oxidative stress. Oxidative
364 stress in the context of pregnancy can negatively impact fetal development, and oxidative
365 stress is common in many pathways leading to congenital defects.[96]

366 George JW et al. report a reduction to mitochondrial REDOX reactions and ATP production,
367 alongside increased glycolysis, in HeLa cells after 24-hour DTG exposure.[88] In erythrocytes,
368 48-hour DTG incubation increased reactive oxygen species (ROS) production, surface
369 ceramide and phosphatidyl serine, and cytosolic [Ca2+], indicating cellular oxidative stress.[86]

370 DTG's inhibitory action on the viral integrase is in part due to cation chelation, which is
371 hypothesized to interfere with the host's own enzymes.[41,49,97] In the Bade AN et al. study,
372 DTG was found to be a broad-spectrum MMP inhibitor by binding the Zn++ ion bound by this
373 class of enzymes.[41] MMPs have essential roles in neural crest migration, synapse

development, axonal guidance, and angiogenesis in the embryo and contribute to uterine vascular remodeling by the cytotrophoblasts in the development of the placenta.[41,98]

The cation chelating property of DTG may extend to other metal-binding enzymes such as superoxide dismutases (Mn-, Zn-, and Cu-SOD), resulting in increased cellular ROS, although these effects have not yet been tested. Oxidative stress at the level of the placenta may result in lower fetal weight as reported in the Mohan H et al. study, however this effect was not observed clinically.[40,99] To test whether these molecular effects have a systemic effect on development, experimental studies on placental function correlated to fetal outcomes ought to be conducted. The metal ion chelating property of DTG is an interesting mechanism to consider further, as it would affect a broad spectrum of pathways that could contribute to the variety of effects observed with DTG in in vitro and model studies. Further the degree of such insult would be modified by dietary factors and could explain clinically observed outcomes. Well-designed studies would be needed to assess this clinically.

4. Conclusions

The global HIV pandemic presents a severe healthcare burden, which can be successfully managed by ART. DTG-based ART is a preferred treatment option in both resource-rich and resource-limited settings because of its efficacy, high barrier to resistance, favourable safety and tolerance profile, and affordability. DTG-associated changes to maternal physiology such as weight change, hyperglycemia, and folate metabolism, along with changes to adipose tissue, oxidative stress, and potential interference with metal-binding enzymes may affect fetal

development and influence metabolic health in the child. However, it remains unclear the degree to which the reported cellular changes impact physiology and whether targeting these pathways in treatment would improve the DTG-specific side effects observed clinically. Furthermore, despite increasing evidence of DTG-associated metabolic changes in non-pregnant adults, there have not been similar reports in pregnancy, and their connection to fetal development has not yet been studied. Studies investigating maternal metabolic health, such as weight and adipose change, plasma lipid profile, adipokine levels, glucose homeostasis correlating to pregnancy outcomes and long-term fetal health are warranted.

Specifically, addressing the following questions would provide great insight: Does DTG affect maternal metabolic health? Do maternal metabolic health changes resulting from DTG treatment affect pregnancy outcomes and fetal metabolic health? How does maternal nutritional status interact with DTG in influencing birth outcomes? In clinical practice, it is pertinent to increase focus on monitoring maternal health and metabolic alterations occurring as a result of DTG treatment. Further, given the scale at which ART is being used in pregnancy it is important that systematic monitoring of adverse events and pregnancy/birth outcomes is implemented, as even small changes in risk have the potential to translate into many pregnancies and babies affected. In the absence of a mechanistic understanding, adequate nutrition and folic acid supplementation should be encouraged.

Conflict of Interest Statement

The authors declare no conflicts of interest.

Funding statement

This work was supported by funds from the Eunice Kennedy Shriver National Institutes of Child Health & Human Development of the National Institutes of Health, award # R01HD104553. The funder played no role in the writing of the manuscript or the decision to submit it for publication.

Contributors

VD and LS conceptualized the manuscript with input from all co-authors. VD generated the original draft with input from LS. HM, CB, JJ, NDEG, AJC, and RZ reviewed and edited the manuscript. All authors approved the final draft.

427 **Table 1: Summary of clinical pregnancy studies including a DTG-based regimen**

Study Type, Location, Date	Regimen	N	Outcomes
Surveillance study, Botswana, Aug 2014 – May 2018[30]	DTG-based ART from conception	426	DTG from conception group had four cases (0·94%) of NTDs: encephalocele, myelomeningocele, iniencephaly, and anencephaly. NTD rates were similar between the HIV-negative (0·09%) and other ART groups (0·12%)
	DTG-based ART started in pregnancy	2812	
	Other ART	11,300	
	HIV-negative	66,057	
Surveillance study, Brazil, Jan 2015 – May 2018[34]	DTG-based ART	382	No increased risk for adverse peripartum outcomes associated with DTG exposure reported. 2/490 (0·18%) of NTDs in DTG treated women reported in study update Feb 2019.
	EFV-based ART	1,045	
Surveillance study, Botswana, Oct 2018 – Mar 2019[33]	DTG-based ART	152	One NTD in DTG from conception arm (0·66%). Two in HIV-negative pregnancies (0·09%).
	Other ART	544	
	HIV-negative	2328	
Surveillance study, United States, Puerto Rico, Swiss cohort, 2007 – Jan 2020[19]	DTG-based ART	120	DTG-based regimen was associated with a mildly higher risk of preterm births associated. 1 case of syndactyly, 2 cases of polydactyly in DTG group.
	Atazanavir/r-based ART	464	
	Darunavir/r-based ART	185	
	Rilpivirine-based ART	243	
	RAL-based ART	86	
	Elvitegravir/c-based ART	159	
Database analysis, Antiretroviral pregnancy registry, Jan 1989 – Jul 2022, APR 2022.[35]	DTG-based ART	1362	The reported rates of congenital anomalies associated with DTG were 3·45% in APR, with one NTD case of anencephaly.
Surveillance study, Botswana,	DTG-based ART	1729	No increased risk of adverse birth outcomes on DTG was reported.

Aug 2014 – Aug 2016[25]	EFV-based ART	4593	
Surveillance study, Botswana, Aug 2014-Apr 2020[28]	DTG-based ART	2,450*	Surveillance study showed that DTG regimen had the same or better peripartum outcomes as compared to other ART regimens in all maternal weight classes.
	EFV-based ART	7,459*	
	Other ART	6,492*	
	NVP-based ART	4,695*	
	LPV/r-based ART	841*	
Open-label RCT, South Africa, Uganda, Jan 2018 – Aug 2018[27]	TDF/FTC/DTG	135	DTG group showed slightly higher (24%) severe adverse events than EFV (18%) group.
	TDF/XTC/EFV	133	
Open-label RCT (72-week follow-up) South Africa, Uganda, Jan 2018 – Aug 2018[18]	TDF/FTC/DTG	135	Greater proportion of adverse pregnancy events were found in DTG (22%) than EFV (11%) arm.
	TDF/FTC/EFV or 3TC	133	
Open-label RCT, Zimbabwe, South Africa, Uganda, Brazil, Botswana, Tanzania, Thailand, United States, India, Jan 2018 – Feb 2019[17]	TDF/FTC/DTG	204	DTG group had lower rates of preterm birth (6%) compared to EFV group (12%). TAF/FTC/DTG had higher gestational weight gain (0·378kg/week) versus TDF/FTC/DTG (0·319kg/week) and TDF/FTC/EFV (0·291kg/week).
	TAF/FTC/DTG	201	
	TDF/FTC/EFV	200	
Surveillance study, Botswana, Aug 2014 – Mar 2019[29]	DTG (TDF/FTC 98.8%)	621	DTG group had 0·35 kg/week weight gain over 18-36 weeks gestation. EFV group had 0·31 and HIV-negative group had 0·44 kg/week weight gain.
	EFV (TDF/FTC 99.8%)	757	
	HIV-negative	11,280	
RCT, Botswana, Aug 2016 – May 2019[24]	Mothers receiving TDF/XTC/DTG	182	No difference in insulin sensitivity in exposed uninfected infants born to women taking DTG versus those taking EFV in pregnancy.
	Mothers receiving TDF/XTC/DTG	124	
Prospective surveillance study, Gaborone,	DTG-based ART	197	Lower rates of GDM were observed in DTG-treated (6·1%) vs EFV-treated
	EFV-based ART	126	

Botswana, Aug 2016 – May 2019[23]			(13·5%) women. Both rates were comparable to HIV-negative group (7·4%).
	HIV-negative	163	
Database analysis, France, 2012 – 2016[100]	DTG-based ART	49	Higher birth defect rates in DTG arm at 4·1% versus RAL (1·3%) and Elvitegravir (1·4%).
	RAL-based ART	240	
	Elvitegravir-based ART	70	
Retrospective analysis, United States, 2015-2018[101]	DTG-based ART	66	No side effects on DTG treatment were reported, with 2 cases of birth defects: a congenital heart abnormality and a nonimmune hydrops fetalis.
Retrospective analysis, Sweden, 2014 – Aug 2017[102]	DTG-based ART	36	DTG-based regimen showed no difference in adverse pregnancy events from that of general population.

428 *varied N for different outcomes
429 Abbreviations: ART, antiretroviral therapy; DTG, dolutegravir; EFV, efavirenz; FTC, emtricitabine;
430 LPV, lopinavir; RAL, raltegravir; TAF, tenofovir alafenamide; TDF, tenofovir disoproxil fumarate;
431 XTC, either 3TC (lamivudine) or FTC; /c, cobicistat booster; /r, ritonavir boosted.

Table 2: Summary of in vivo reproductive studies with DTG

Animal model, study	Regimen	Dosage (mg/kg)	Treatment start	N (Litters)	Results
C57BL/6J female mice[40]	1xDTG (DTG/FTC/TDF) equivalent to human therapeutic drug levels	2·5/33·3/50	GD 0.5	150	Five cases of NTDs were observed in the therapeutic 1xDTG dosage only. Two had exencephaly, two had spina bifida, and one had potential anencephaly.
	5xDTG (DTG/FTC/TDF) reaching supratherapeutic levels	12·5/33·3/50		111	
	Control, water	-		91	
C3H/HeJ female mice[41]	DTG	50	GD 0·5	17	Exencephaly in one fetus in DTG arm but N insufficient for statistical power.
	Control, vehicle	-		9	
Sprague-Dawley female rats[39]	DTG	5	GD 6	22	No significant differences in external abnormalities. Meningocele/absent eye bulge at 1000mg/kg dose but N insufficient for statistical power.
	DTG	50		21	
	DTG	100		27	
	DTG	300		27	
	DTG	1000		47	
	Control	-		49	
Japanese white female rabbits[39]	DTG	40	GD 6	19	No significant differences in external abnormalities observed. One cranioschisis at 40mg/kg dose but N insufficient for statistical power.
	DTG	100		3	
	DTG	200		18	
	DTG	300		5	
	DTG	1000		24	
	Control	-		24	

Abbreviations: DTG, dolutegravir; FTC, emtricitabine; GD, gestational day; TDF, tenofovir disoproxil fumarate.

Table 3: Summary of In vitro developmental toxicology studies with DTG

Culture model, study	DTG dosage (μM)*	N	Results
Murine P19C5 pluripotent stem cells and human embryonic stem cells H9[45]	0·25, 0·5, 1, 2, 4	46-48 aggregates per condition for morphogenesis	DTG was associated with impaired stem cell morphogenesis and changes to developmental regulator genes in a dose-dependent manner in both P19C5 and H9 cells.
CA1S human embryonic stem cells[46]	8·32	5 replicates	DTG was associated with reduced expression of pluripotency markers in CA1S cells.
H9 human embryonic stem cells[46]	8·32	6 replicates	DTG was associated with increased rates of apoptosis in H9 cells.
Sprague Dawley GD9 embryo culture[43]	12·6	16 embryos	DTG did not affect embryo lethality, visceral yolk sac, somite number, or embryo size.
	22·2	16 embryos	
Zebrafish embryo culture[52]	100	2-4 experimental replicates	DTG was associated with developmental toxicity post-fertilization.

*Cmax for DTG in non-pregnant adults is reported as 701–11·56 μM.[103]

Figure Legends

Figure 1. Summary of the observed effects associated with DTG from animal and *in vitro* studies. DTG effects on folate metabolism[40,49–51], cellular energy homeostasis[86,88], adipocyte function[86,89,90,93], and matrix metalloproteinases[41] are shown (anticlockwise from top left). Dotted line indicates variable effects reported. Red arrows indicated increase/higher; blue arrows indicate decrease/lower, black arrow indicates unchanged. FRa, folate receptor 1; RFC, reduced folate carrier; PCFT; proton-coupled folate transporter; MMP, matrix metalloproteinases; PPARγ, peroxisome proliferator-activated receptor gamma; CEBPα, CCAAT/enhancer-binding protein alpha; UCP1, uncoupling protein 1; ROS, reactive oxygen species.

References

1. Pandey A, Galvani AP. The global burden of HIV and prospects for control. *Lancet HIV*. 2019; **6**: e809–11.

2. UNAIDS. Understanding Fast-Track Targets. Accelerating action to end the AIDS epidemic by 2030. *UNAIDS*; 2015.

3. Panel on Treatment of HIV During Pregnancy and Prevention of Perinatal Transmission. Recommendations for the Use of Antiretroviral Drugs During Pregnancy and Interventions to Reduce Perinatal HIV Transmission in the United States [Internet]. Department of Health and Human Services; Available from: https://clinicalinfo.hiv.gov/en/guidelines/perinatal.

4. Sibiude J, Le Chenadec J, Mandelbrot L, Hoctin A, Dollfus C, Faye A, et al. Update of Perinatal Human Immunodeficiency Virus Type 1 Transmission in France: Zero Transmission for 5482 Mothers on Continuous Antiretroviral Therapy From Conception and With Undetectable Viral Load at Delivery. *Clin Infect Dis*. 2023; **76**: e590–8.

5. Consolidated guidelines on HIV prevention, testing, treatment, service delivery and monitoring: recommendations for a public health approach. Geneva, Switzerland: World Health Organization; 2021.

6. Kumar S, Samaras K. The Impact of Weight Gain During HIV Treatment on Risk of Pre-diabetes, Diabetes Mellitus, Cardiovascular Disease, and Mortality. *Front*

Endocrinol. 2018; **9**: 705.

7. Panel on Antiretroviral Guidelines for Adults and Adolescents. Guidelines for the Use of Antiretroviral Agents in Adults and Adolescents with HIV. Department of Health and Human Services. [Internet]. 2021. Available from: https://clinicalinfo.hiv.gov/en/guidelines/adult-and-adolescent-arv.

8. Ryom L, De Miguel R, Cotter AG, Podlekareva D, Beguelin C, Waalewijn H, et al. Major revision version 11.0 of the European AIDS Clinical Society Guidelines 2021. *HIV Med*. 2022; **23**: 849–58.

9. Llibre JM, Pulido F, García F, García Deltoro M, Blanco JL, Delgado R. Genetic barrier to resistance for dolutegravir. *AIDS Rev*. 2015; **17**: 56–64.

10. Kouamou V, Manasa J, Katzenstein D, McGregor AM, Ndhlovu CE, Makadzange AT. Drug resistance and optimizing dolutegravir regimens for adolescents and young adults failing antiretroviral therapy. *AIDS*. 2019; **33**: 1729–37.

11. Kanters S, Vitoria M, Doherty M, Socias ME, Ford N, Forrest JI, et al. Comparative efficacy and safety of first-line antiretroviral therapy for the treatment of HIV infection: a systematic review and network meta-analysis. *Lancet HIV*. 2016; **3**: e510–20.

12. NAMSAL ANRS 12313 Study Group, Kouanfack C, Mpoudi-Etame M, Omgba Bassega P, Eymard-Duvernay S, Leroy S, et al. Dolutegravir-Based or Low-Dose Efavirenz-Based Regimen for the Treatment of HIV-1. *N Engl J Med*. 2019; **381**: 816–

491 26.

492 13. Venter WDF, Sokhela S, Simmons B, Moorhouse M, Fairlie L, Mashabane N, et al.
493 Dolutegravir with emtricitabine and tenofovir alafenamide or tenofovir disoproxil
494 fumarate versus efavirenz, emtricitabine, and tenofovir disoproxil fumarate for initial
495 treatment of HIV-1 infection (ADVANCE): week 96 results from a randomised, phase
496 3, non-inferiority trial. *Lancet HIV*. 2020; **7**: e666–76.

497 14. Calmy A, Tovar Sanchez T, Kouanfack C, Mpoudi-Etame M, Leroy S, Perrineau S, et
498 al. Dolutegravir-based and low-dose efavirenz-based regimen for the initial treatment of
499 HIV-1 infection (NAMSAL): week 96 results from a two-group, multicentre,
500 randomised, open label, phase 3 non-inferiority trial in Cameroon. *Lancet HIV*. 2020; **7**:
501 e677–87.

502 15. Venter WDF, Moorhouse M, Sokhela S, Fairlie L, Mashabane N, Masenya M, et al.
503 Dolutegravir plus Two Different Prodrugs of Tenofovir to Treat HIV. *N Engl J Med*.
504 2019; **381**: 803–15.

505 16. Waitt C, Orrell C, Walimbwa S, Singh Y, Kintu K, Simmons B, et al. Safety and
506 pharmacokinetics of dolutegravir in pregnant mothers with HIV infection and their
507 neonates: A randomised trial (DolPHIN-1 study). PLoS Med. 2019;16:e1002895.

508 17. Lockman S, Brummel SS, Ziemba L, Stranix-Chibanda L, McCarthy K, Coletti A, et al.
509 Efficacy and safety of dolutegravir with emtricitabine and tenofovir alafenamide

fumarate or tenofovir disoproxil fumarate, and efavirenz, emtricitabine, and tenofovir disoproxil fumarate HIV antiretroviral therapy regimens started in pregnancy (IMPAACT 2010/VESTED): a multicentre, open-label, randomised, controlled, phase 3 trial. *Lancet*. 2021; **397**: 1276–92.

18. Kintu K, Malaba TR, Nakibuka J, Papamichael C, Colbers A, Byrne K, et al. Dolutegravir versus efavirenz in women starting HIV therapy in late pregnancy (DolPHIN-2): an open-label, randomised controlled trial. *Lancet HIV*. 2020;**7**: e332–9.

19. Patel K, Huo Y, Jao J, Powis KM, Williams PL, Kacanek D, et al. Dolutegravir in Pregnancy as Compared with Current HIV Regimens in the United States. *N Engl J Med.* 2022; **387**: 799–809.

20. Kandel CE, Walmsley SL. Dolutegravir - a review of the pharmacology, efficacy, and safety in the treatment of HIV. *Drug Des Devel Ther*. 2015; **9**: 3547–55.

21. Sax PE, Erlandson KM, Lake JE, Mccomsey GA, Orkin C, Esser S, et al. Weight Gain Following Initiation of Antiretroviral Therapy: Risk Factors in Randomized Comparative Clinical Trials. *Clin Infect Dis*. 2020; **71**: 1379–89.

22. Lamorde M, Atwiine M, Owarwo NC, Ddungu A, Laker EO, Mubiru F, et al. Dolutegravir-associated hyperglycaemia in patients with HIV. *Lancet HIV*. 2020; **7**: e461–2.

23. Mmasa KN, Powis K, Sun S, Makhema J, Mmalane M, Kgole S, et al. Gestational

diabetes in women living with HIV in Botswana: lower rates with dolutegravir- than with efavirenz-based antiretroviral therapy. *HIV Med*. 2021; **22**: 715–22.

24. Jao J, Sun S, Bonner LB, Legbedze J, Mmasa KN, Makhema J, et al. Lower Insulin Sensitivity in Newborns With In Utero HIV and Antiretroviral Exposure Who Are Uninfected in Botswana. *J Infect Dis*. 2022; **226**: 2002–9.

25. Zash R, Jacobson DL, Diseko M, Mayondi G, Mmalane M, Essex M, et al. Comparative safety of dolutegravir-based or efavirenz-based antiretroviral treatment started during pregnancy in Botswana: an observational study. *Lancet Glob Health*. 2018; **6**: e804–10.

26. Hill A, Clayden P, Thorne C, Christie R, Zash R. Safety and pharmacokinetics of dolutegravir in HIV-positive pregnant women: a systematic review. *J Virus Erad*. 2018; **4**: 66–71.

27. Malaba TR, Nakatudde I, Kintu K, Colbers A, Chen T, Reynolds H, et al. 72 weeks post-partum follow-up of dolutegravir versus efavirenz initiated in late pregnancy (DolPHIN-2): an open-label, randomised controlled study. *Lancet HIV*. 2022; **9**: e534–43.

28. Zash R, Caniglia EC, Diseko M, Mayondi G, Mabuta J, Luckett R, et al. Maternal weight and birth outcomes among women on antiretroviral treatment from conception in a birth surveillance study in Botswana. *J Int AIDS Soc*. 2021; **24**: e25763.

29. Caniglia EC, Shapiro R, Diseko M, Wylie BJ, Zera C, Davey S, et al. Weight gain

during pregnancy among women initiating dolutegravir in Botswana. *EClinicalMedicine*. 2020; **29–30**: 100615.

30. Zash R, Makhema J, Shapiro RL. Neural-Tube Defects with Dolutegravir Treatment from the Time of Conception. *N Engl J Med.* 2018; **379**: 979–81.

31. Zash R, Holmes L, Diseko M, Jacobson DL, Brummel S, Mayondi G, et al. Neural-Tube Defects and Antiretroviral Treatment Regimens in Botswana. *N Engl J Med*. 2019; **381**: 827–40.

32. Zash R, Holmes LB, Diseko M, Jacobson DL, Mayondi G, Mabuta J, et al. Update on neural tube defects with antiretroviral exposure in the Tsepamo Study, Botswana. In 2022. Available from: https://programme.aids2022.org/Abstract/Abstract/?abstractid=12759

33. Raesima MM, Ogbuabo CM, Thomas V, Forhan SE, Gokatweng G, Dintwa E, et al. Dolutegravir Use at Conception - Additional Surveillance Data from Botswana. *N Engl J Med*. 2019; **381**: 885–7.

34. Pereira GFM, Kim A, Jalil EM, Fernandes Fonseca F, Shepherd BE, Veloso VG, et al. Dolutegravir and pregnancy outcomes in women on antiretroviral therapy in Brazil: a retrospective national cohort study. *Lancet HIV*. 2021; **8**: e33–41.

35. Antiretroviral Pregnancy Registry Steering Committee. Antiretroviral Pregnancy Registry Interim Report for 1 January 1989 through 31 July 2022. [Internet].

Wilmington, NC: Registry Coordinating Center; 2022. Available from: www.APRegistry.com

36. Money D, Lee T, O'Brien C, Brophy J, Bitnun A, Kakkar F, et al. Congenital anomalies following antenatal exposure to dolutegravir: a Canadian surveillance study. BJOG Int J Obstet Gynaecol. 2019;126:1338–45.

37. Phillips AN, Venter F, Havlir D, Pozniak A, Kuritzkes D, Wensing A, et al. Risks and benefits of dolutegravir-based antiretroviral drug regimens in sub-Saharan Africa: a modelling study. Lancet HIV. 2019;6:e116–27.

38. Dugdale CM, Ciaranello AL, Bekker LG, Stern ME, Myer L, Wood R, et al. Risks and Benefits of Dolutegravir- and Efavirenz-Based Strategies for South African Women With HIV of Child-Bearing Potential: A Modeling Study. *Ann Intern Med.* 2019; **170**: 614–25.

39. Stanislaus DJ, Posobiec LM, Laffan SB, Solomon HM, Ziejewski MK, Romach EH. Absence of developmental and reproductive toxicity in animals exposed to dolutegravir. *Birth Defects Res*. 2020; **112**: 245–61.

40. Mohan H, Lenis MG, Laurette EY, Tejada O, Sanghvi T, Leung KY, et al. Dolutegravir in pregnant mice is associated with increased rates of fetal defects at therapeutic but not at supratherapeutic levels. *EBioMedicine*. 2021; **63**: 103167.

41. Bade AN, McMillan JM, Liu Y, Edagwa BJ, Gendelman HE. Dolutegravir Inhibition of

586 Matrix Metalloproteinases Affects Mouse Neurodevelopment. *Mol Neurobiol.* 2021; **58**:
587 5703–21.

588 42. Posobiec LM, Chapman SP, Murzyn SF, Rendemonti JE, Stanislaus DJ, Romach EH.
589 No developmental toxicity observed with dolutegravir in rat whole embryo culture.
590 *Birth Defects Res*. 2021; **113**: 1190–7.

591 43. Copp AJ, Greene NDE, Jao J, Zash R, Mohan H, Dontsova V, et al. Dolutegravir and
592 rat whole embryo culture. *Birth Defects Res*. 2022; **114**: 23–4.

593 44. Kirkwood-Johnson L, Katayama N, Marikawa Y. Dolutegravir Impairs Stem Cell-
594 Based 3D Morphogenesis Models in a Manner Dependent on Dose and Timing of
595 Exposure: An Implication for Its Developmental Toxicity. *Toxicol Sci Off J Soc*
596 *Toxicol*. 2021; **184**: 191–203.

597 45. Smith MSR, Mohan H, Ajaykumar A, Hsieh AYY, Martineau L, Patel R, et al. Second-
598 Generation Human Immunodeficiency Virus Integrase Inhibitors Induce Differentiation
599 Dysregulation and Exert Toxic Effects in Human Embryonic Stem Cell and Mouse
600 Models. *J Infect Dis*. 2022; **226**: 1992–2001.

601 46. King JC. Maternal obesity, metabolism, and pregnancy outcomes. *Annu Rev Nutr.* 2006;
602 **26**: 271–91.

603 47. Stothard KJ, Tennant PWG, Bell R, Rankin J. Maternal overweight and obesity and the
604 risk of congenital anomalies: a systematic review and meta-analysis. *JAMA*. 2009; **301**:

636–50.

48. Chandiwana NC, Chersich M, Venter WDF, Akpomiemie G, Hill A, Simmons B, et al. Unexpected interactions between dolutegravir and folate: randomized trial evidence from South Africa. *AIDS*. 2021; **35**: 205–11.

49. Cabrera RM, Souder JP, Steele JW, Yeo L, Tukeman G, Gorelick DA, et al. The antagonism of folate receptor by dolutegravir: developmental toxicity reduction by supplemental folic acid. *AIDS*. 2019; **33**: 1967–76.

50. Zamek-Gliszczynski MJ, Zhang X, Mudunuru J, Du Y, Chen JL, Taskar KS, et al. Clinical Extrapolation of the Effects of Dolutegravir and Other HIV Integrase Inhibitors on Folate Transport Pathways. *Drug Metab Dispos Biol Fate Chem*. 2019; **47**: 890–8.

51. Gilmore JC, Hoque MT, Dai W, Mohan H, Dunk C, Serghides L, et al. Interaction between dolutegravir and folate transporters and receptor in human and rodent placenta. *EBioMedicine*. 2022; **75**: 103771.

52. Hamooya BM, Mulenga LB, Masenga SK, Fwemba I, Chirwa L, Siwingwa M, et al. Metabolic syndrome in Zambian adults with human immunodeficiency virus on antiretroviral therapy: Prevalence and associated factors. *Medicine* (Baltimore). 2021; **100**: e25236.

53. McCann K, Shah S, Hindley L, Hill A, Qavi A, Simmons B, et al. Implications of weight gain with newer anti-retrovirals: 10-year predictions of cardiovascular disease

and diabetes. *AIDS*. 2021; **35**: 1657–65.

54. Dontsova V, Mohan H, Dunk CE, Jao J, Greene NDE, Copp AJ, et al. Effect of Dolutegravir on Glucose Homeostasis in Female Mice. In 2022. Available from: https://seatoskymeeting.eventsair.com/QuickEventWebsitePortal/31st-annual-canadian-conference-on-hivaids-research-cahr-2022/cahr-2022-virtual-agenda/Agenda/AgendaItemDetail?id=4275e976-13d1-227f-8f54-39fefc85e70d

55. Griesel R, Kawuma AN, Wasmann R, Sokhela S, Akpomiemie G, Venter WDF, et al. Concentration-response relationships of dolutegravir and efavirenz with weight change after starting antiretroviral therapy. *Br J Clin Pharmacol*. 2022; **88**: 883–93.

56. Esber AL, Chang D, Iroezindu M, Bahemana E, Kibuuka H, Owuoth J, et al. Weight gain during the dolutegravir transition in the African Cohort Study. *J Int AIDS Soc*. 2022; **25**: e25899.

57. Norwood J, Turner M, Bofill C, Rebeiro P, Shepherd B, Bebawy S, et al. Brief Report: Weight Gain in Persons With HIV Switched From Efavirenz-Based to Integrase Strand Transfer Inhibitor-Based Regimens. *J Acquir Immune Defic Syndr*. 2017; **76**: 527–31.

58. Bourgi K, Jenkins CA, Rebeiro PF, Palella F, Moore RD, Altoff KN, et al. Weight gain among treatment-naïve persons with HIV starting integrase inhibitors compared to non-nucleoside reverse transcriptase inhibitors or protease inhibitors in a large observational cohort in the United States and Canada. *J Int AIDS Soc*. 2020; **23**: e25484.

643 59. Bourgi K, Rebeiro PF, Turner M, Castilho JL, Hulgan T, Raffanti SP, et al. Greater
644 Weight Gain in Treatment-naive Persons Starting Dolutegravir-based Antiretroviral
645 Therapy. *Clin Infect Dis*. 2020; **70**: 1267–74.

646 60. Calza L, Colangeli V, Borderi M, Bon I, Borioni A, Volpato F, et al. Weight gain in
647 antiretroviral therapy-naive HIV-1-infected patients starting a regimen including an
648 integrase strand transfer inhibitor or darunavir/ritonavir. *Infection*. 2020; **48**: 213–21.

649 61. Ibrahim F, Samarawickrama A, Hamzah L, Vincent R, Gilleece Y, Waters L, et al.
650 Bone mineral density, kidney function, weight gain and insulin resistance in women
651 who switch from TDF/FTC/NNRTI to ABC/3TC/DTG. *HIV Med*. 2021; **22**: 83–91.

652 62. Ruderman SA, Crane HM, Nance RM, Whitney BM, Harding BN, Mayer KH, et al.
653 Brief Report: Weight Gain Following ART Initiation in ART-Naïve People Living With
654 HIV in the Current Treatment Era. *J Acquir Immune Defic Syndr*. 2021; **86**: 339–43.

655 63. Ando N, Nishijima T, Mizushima D, Inaba Y, Kawasaki Y, Kikuchi Y, et al. Long-term
656 weight gain after initiating combination antiretroviral therapy in treatment-naïve Asian
657 people living with human immunodeficiency virus. *Int J Infect Dis*. 2021; **110**: 21–8.

658 64. Taramasso L, Bonfanti P, Ricci E, Orofino G, Squillace N, Menzaghi B, et al. Factors
659 Associated With Weight Gain in People Treated With Dolutegravir. *Open Forum Infect*
660 *Dis.* 2020; **7**: ofaa195.

661 65. Taramasso L, Ricci E, Menzaghi B, Orofino G, Passerini S, Madeddu G, et al. Weight

Gain: A Possible Side Effect of All Antiretrovirals. *Open Forum Infect Dis*. 2017; **4**: ofx239.

66. Lake JE, Wu K, Bares SH, Debroy P, Godfrey C, Koethe JR, et al. Risk Factors for Weight Gain Following Switch to Integrase Inhibitor-Based Antiretroviral Therapy. *Clin Infect Dis*. 2020; **71**: e471–7.

67. Rizzardo S, Lanzafame M, Lattuada E, Luise D, Vincenzi M, Tacconelli E, et al. Dolutegravir monotherapy and body weight gain in antiretroviral naïve patients. *AIDS*. 2019; **33**: 1673–4.

68. Goldberg RN, Kania AT, Michienzi SM, Patel M, Badowski ME. Weight Gain in Incarcerated Individuals Living With HIV After Switching to Integrase Strand Inhibitor-Based Therapy. *J Int Assoc Provid AIDS Care*. 2021; **20**: 2325958221996860.

69. Calza L, Borderi M, Colangeli V, Miani T, Nuti B, Bon I, et al. Weight gain in treatment-naive HIV-1 infected patients starting abacavir/lamivudine/dolutegravir or tenofovir alafenamide/emtricitabine/bictegravir. *AIDS*. 2022; **36**: 153–5.

70. Hoffman RM, Ziemba L, Chinula L, João E, Stringer JSA, Fairlie L, et al. Antepartum weight gain and adverse pregnancy outcomes: a mediation analysis. In 2023. Available from: https://www.croiconference.org/wp-content/uploads/sites/2/posters/2023/Weight_mediation_Analysis_Poster_12Feb23_Final-133209583712785646.pdf

71. Calza L, Colangeli V, Borderi M, Coladonato S, Tazza B, Bon I, et al. Improvement in insulin sensitivity and serum leptin concentration after the switch from a ritonavir-boosted PI to raltegravir or dolutegravir in non-diabetic HIV-infected patients. *J Antimicrob Chemother*. 2019; **74**: 731–8.

72. Ursenbach A, Max V, Maurel M, Bani-Sadr F, Gagneux-Brunon A, Garraffo R, et al. Incidence of diabetes in HIV-infected patients treated with first-line integrase strand transfer inhibitors: a French multicentre retrospective study. *J Antimicrob Chemother*. 2020; **75**: 3344–8.

73. Hsu R, Brunet L, Fusco JS, Mounzer K, Vannappagari V, Henegar CE, et al. Incident type 2 diabetes mellitus after initiation of common HIV antiretroviral drugs. *AIDS*. 2021; **35**: 81–90.

74. Zakumumpa H, Kiguba R, Ndagije HB, Ategeka G, Ssanyu JN, Kitutu FE. Patient experiences of sexual dysfunction after transition to dolutegravir-based HIV treatment in mid-Western Uganda: a qualitative study. *BMC Infect Dis*. 2022; **22**: 692.

75. McLaughlin M, Walsh S, Galvin S. Dolutegravir-induced hyperglycaemia in a patient living with HIV. *J Antimicrob Chemother*. 2018; **73**: 258–60.

76. Hailu W, Tesfaye T, Tadesse A. Hyperglycemia After Dolutegravir-Based Antiretroviral Therapy. *Int Med Case Rep J*. 2021; **14**: 503–7.

77. Hirigo AT, Gutema S, Eifa A, Ketema W. Experience of dolutegravir-based

antiretroviral treatment and risks of diabetes mellitus. *SAGE Open Med Case Rep*. 2022; **10**: 2050313X221079444.

78. Fong PS, Flynn DM, Evans CD, Korthuis PT. Integrase strand transfer inhibitor-associated diabetes mellitus: A case report. *Int J STD AIDS*. 2017; **28**: 626–8.

79. Namulindwa A, Wasswa JH, Muyindike W, Tamukong R, Oloro J. Prevalence and factors associated with adverse drug events among patients on dolutegravir-based regimen at the Immune Suppression Syndrome Clinic of Mbarara Regional Referral Hospital, Uganda: a mixed design study. *AIDS Res Ther*. 2022; **19**: 18.

80. van Lunzen J, Maggiolo F, Arribas JR, Rakhmanova A, Yeni P, Young B, et al. Once daily dolutegravir (S/GSK1349572) in combination therapy in antiretroviral-naive adults with HIV: planned interim 48 week results from SPRING-1, a dose-ranging, randomised, phase 2b trial. *Lancet Infect Dis*. 2012; **12**: 111–8.

81. Cahn P, Pozniak AL, Mingrone H, Shuldyakov A, Brites C, Andrade-Villanueva JF, et al. Dolutegravir versus raltegravir in antiretroviral-experienced, integrase-inhibitor-naive adults with HIV: week 48 results from the randomised, double-blind, non-inferiority SAILING study. *Lancet*. 2013; **382**: 700–8.

82. Walmsley SL, Antela A, Clumeck N, Duiculescu D, Eberhard A, Gutiérrez F, et al. Dolutegravir plus abacavir-lamivudine for the treatment of HIV-1 infection. *N Engl J Med*. 2013; **369**: 1807–18.

83. Raffi F, Jaeger H, Quiros-Roldan E, Albrecht H, Belonosova E, Gatell JM, et al. Once-daily dolutegravir versus twice-daily raltegravir in antiretroviral-naive adults with HIV-1 infection (SPRING-2 study): 96 week results from a randomised, double-blind, non-inferiority trial. *Lancet Infect Dis*. 2013; **13**: 927–35.

84. Clotet B, Feinberg J, van Lunzen J, Khuong-Josses MA, Antinori A, Dumitru I, et al. Once-daily dolutegravir versus darunavir plus ritonavir in antiretroviral-naive adults with HIV-1 infection (FLAMINGO): 48 week results from the randomised open-label phase 3b study. *Lancet*. 2014; **383**: 2222–31.

85. Chinula L, Goldberg E, McCathy K, Krotje C, Knowles K, Mathad J, et al. Pregnancy Hemoglobin A1c and glucose with DTG vs EFV, TDF vs TAF: IMPAACT 2010. In 2022. Available from: https://www.croiconference.org/abstract/pregnancy-hemoglobin-a1c-and-glucose-with-dtg-vs-efv-tdf-vs-taf-impaact-2010/

86. Al Mamun Bhuyan A, Signoretto E, Bissinger R, Lang F. Enhanced Eryptosis Following Exposure to Dolutegravir. *Cell Physiol Biochem Int J Exp Cell Physiol Biochem Pharmacol*. 2016; **39**: 639–50.

87. Gorwood J, Bourgeois C, Pourcher V, Pourcher G, Charlotte F, Mantecon M, et al. The Integrase Inhibitors Dolutegravir and Raltegravir Exert Proadipogenic and Profibrotic Effects and Induce Insulin Resistance in Human/Simian Adipose Tissue and Human Adipocytes. *Clin Infect Dis*. 2020; **71**: e549–60.

738 88. George JW, Mattingly JE, Roland NJ, Small CM, Lamberty BG, Fox HS, et al.
739 Physiologically Relevant Concentrations of Dolutegravir, Emtricitabine, and Efavirenz
740 Induce Distinct Metabolic Alterations in HeLa Epithelial and BV2 Microglial Cells.
741 *Front Immunol*. 2021; **12**: 639378.

742 89. Jung I, Tu-Sekine B, Jin S, Anokye-Danso F, Ahima RS, Brown TT, et al. Dolutegravir
743 Suppresses Thermogenesis via Disrupting UCP1 Expression and Mitochondrial
744 Function in Brown/Beige Adipocytes in preclinical models. *J Infect Dis*. 2022; jiac175.

745 90. Ngono Ayissi K, Gorwood J, Le Pelletier L, Bourgeois C, Beaupère C, Auclair M, et al.
746 Inhibition of Adipose Tissue Beiging by HIV Integrase Inhibitors, Dolutegravir and
747 Bictegravir, Is Associated with Adipocyte Hypertrophy, Hypoxia, Elevated Fibrosis,
748 and Insulin Resistance in Simian Adipose Tissue and Human Adipocytes. *Cells*. 2022;
749 **11**: 1841.

750 91. McMahon C, Trevaskis JL, Carter C, Holsapple K, White K, Das M, et al. Lack of an
751 association between clinical INSTI-related body weight gain and direct interference
752 with MC4 receptor (MC4R), a key central regulator of body weight. *PloS One*. 2020;
753 **15**: e0229617.

754 92. Xu H, Zhang H, Fang Y, Yang H, Chen Y, Zhang C, et al. Activation of the
755 Melanocortin-4 receptor signaling by α-MSH stimulates nerve-dependent mouse digit
756 regeneration. *Cell Regen*. 2021; **10**: 19.

757 93. Pickering R, Asundi A, Lin N. In vitro model to assess antiretroviral therapy on
758 adipocyte biology [CROI Abstract 514]. vCROI 2021 Abstr. Eb. 2021; : 196.

759 94. Pérez-Pérez A, Toro A, Vilariño-García T, Maymó J, Guadix P, Dueñas JL, et al. Leptin
760 action in normal and pathological pregnancies. *J Cell Mol Med*. 2018; **22**: 716–27.

761 95. Sagawa N, Yura S, Itoh H, Kakui K, Takemura M, Nuamah MA, et al. Possible role of
762 placental leptin in pregnancy: a review. *Endocrine*. 2002; **19**: 65–71.

763 96. Wells PG, McCallum GP, Chen CS, Henderson JT, Lee CJJ, Perstin J, et al. Oxidative
764 stress in developmental origins of disease: teratogenesis, neurodevelopmental deficits,
765 and cancer. *Toxicol Sci*. 2009; **108** :4–18.

766 97. Barreca ML, Iraci N, De Luca L, Chimirri A. Induced-fit docking approach provides
767 insight into the binding mode and mechanism of action of HIV-1 integrase inhibitors.
768 *ChemMedChem*. 2009; **4**: 1446–56.

769 98. Majali-Martinez A, Hiden U, Ghaffari-Tabrizi-Wizsy N, Lang U, Desoye G, Dieber-
770 Rotheneder M. Placental membrane-type metalloproteinases (MT-MMPs): Key players
771 in pregnancy. *Cell Adhes Migr*. 2016; **10**: 136–46.

772 99. Poston L, Raijmakers MTM. Trophoblast oxidative stress, antioxidants and pregnancy
773 outcome--a review. *Placenta*. 2004; **25** Suppl A: S72-78.

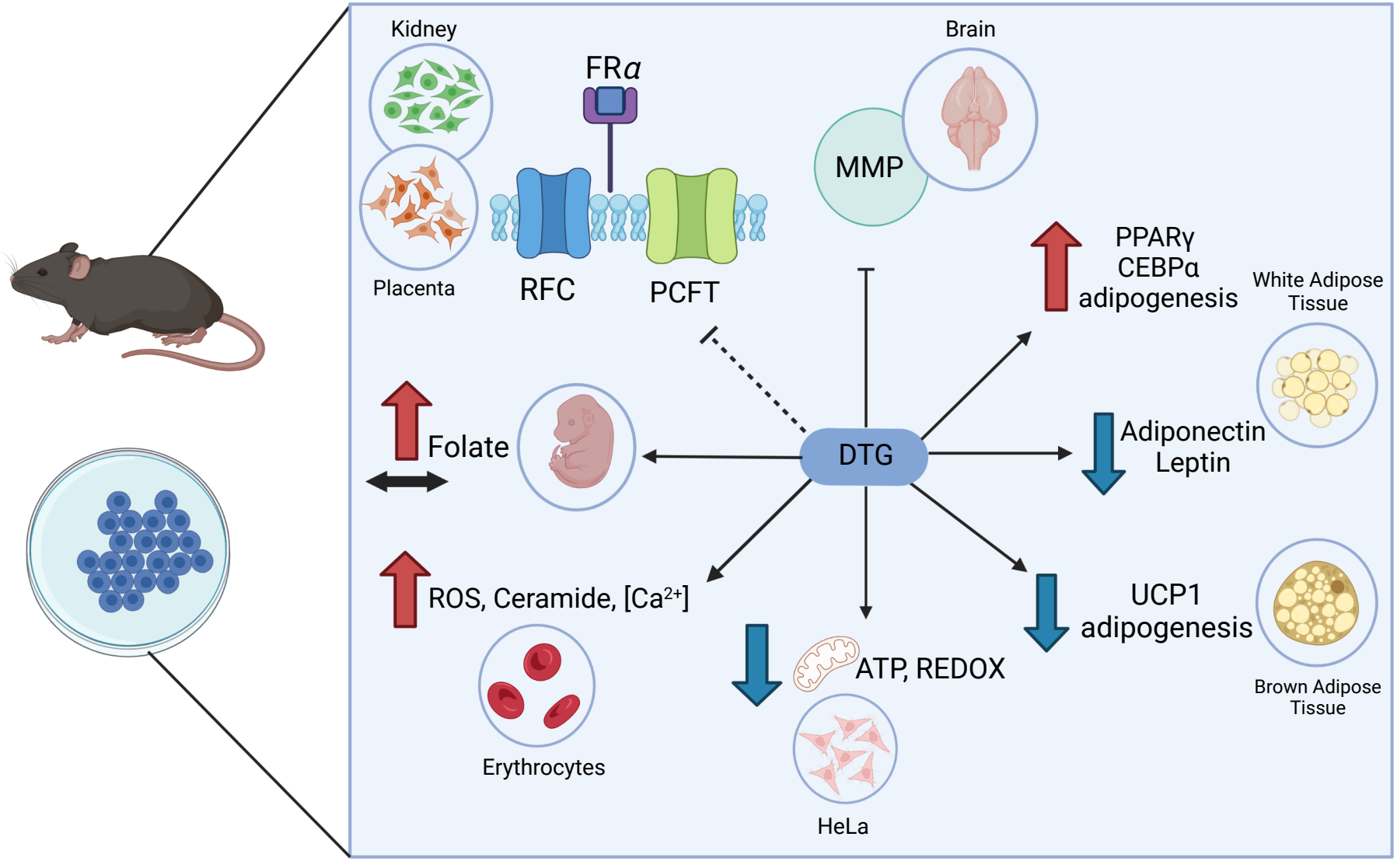
774 100. Chouchana L, Beeker N, Treluyer JM. Is There a Safety Signal for Dolutegravir and

Integrase Inhibitors During Pregnancy? *J Acquir Immune Defic Syndr*. 2019; **81**: 481–6.

101. Grayhack C, Sheth A, Kirby O, Davis J, Sibliss K, Nkwihoreze H, et al. Evaluating outcomes of mother-infant pairs using dolutegravir for HIV treatment during pregnancy. *AIDS*. 2018; **32**: 2017–21.

102. Bornhede R, Soeria-Atmadja S, Westling K, Pettersson K, Navér L. Dolutegravir in pregnancy-effects on HIV-positive women and their infants. *Eur J Clin Microbiol Infect Dis*. 2018; **37**: 495–500.

103. Kala S, Watson B, Zhang JG, Papp E, Guzman Lenis M, Dennehy M, et al. Improving the clinical relevance of a mouse pregnancy model of antiretroviral toxicity; a pharmacokinetic dosing-optimization study of current HIV antiretroviral regimens. *Antiviral Res*. 2018; **159**: 45–54.


Kidney
Brain
FRα
MMP
Placenta
RFC
PCFT
PPARγ
CEBPα
adipogenesis
White Adipose Tissue
Folate
DTG
Adiponectin
Leptin
ROS, Ceramide, [Ca2+]
UCP1
adipogenesis
ATP, REDOX
Brown Adipose Tissue
Erythrocytes
HeLa