Citation:

J. Banerjee, T. S. Leung, and N. Aladangady, "Blood transfusion in preterm infants improves intestinal tissue oxygenation without alteration in blood flow," *Vox Sang,* vol. 111, pp. 399-408, Nov 2016.

Title Page

Blood transfusion in preterm infants improves intestinal tissue oxygenation without alteration in blood flow

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Running title: Transfusion and intestinal perfusion

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Statement of financial support: The study was part funded by Garfield Weston Foundation,

Hamamatsu Photonic KK, Japan and HCA International.

Disclosures: The authors declare that they have no competing interests.

Category of the study: Clinical observational study

Word count: 3149 words

Abstract

Background and Objective: The objective of the study was to investigate the splanchnic blood flow velocity and oximetry response to blood transfusion in preterm infants according to postnatal age.

Materials and Methods: Preterm infants receiving blood transfusion were recruited to three groups: 1 to 7 (group 1; n=20), 8 to 28 (group 2; n=21) & ≥29 days of life (group 3; n=18). Superior mesenteric artery (SMA) peak systolic (PSV) and diastolic velocities were measured 30-60 minutes pre and post-transfusion using Doppler ultrasound scan. Splanchnic Tissue Haemoglobin Index (sTHI), Tissue Oxygenation Index (sTOI) and Fractional Tissue Oxygen Extraction (sFTOE) were measured from 15-20 minutes before to post-transfusion using near infra-red spectroscopy (NIRS).

Results: The mean pre-transfusion Hb in Group 1, 2 and 3 was 11, 10 and 9g/dl respectively. The mean (SD) pre-transfusion SMA PSV in Group 1, 2 and 3 was 0.63 (0.32), 0.81 (0.33) and 0.97 (0.40) m/sec respectively, and this did not change significantly following transfusion. The mean (SD) pre-transfusion sTOI in Group 1, 2 and 3 was 36.7 (19.3), 44.6 (10.4) and 41.3 (10.4)% respectively. The sTHI and sTOI increased ($p<0.01$), and sFTOE decreased ($p<0.01$) following transfusion in all groups. On multivariate analysis, changes in SMA PSV and sTOI following blood transfusion were not associated with PDA, feeding, pre-transfusion Hb and mean blood pressure.

Conclusion: Pre-transfusion baseline splanchnic tissue oximetry and blood flow velocity varied with postnatal age. Blood transfusion improved intestinal tissue oxygenation without altering mesenteric blood flow velocity irrespective of postnatal ages.

Abbreviations:

APH – antepartum haemorrhage, sTOI – splanchnic tissue oxygenation index, sTHI – splanchnic tissue haemoglobin index, $SrSO_2$ – splanchnic regional oxygen saturation, sFTOE – splanchnic fractional tissue oxygen extraction, GA – gestational age, Hb – haemoglobin, IUGR – intra-uterine growth restriction, PDA – patent ductus arteriosus, PET – pre-eclamptic toxaemia, PSV – peak systolic velocity, SMA – superior mesenteric artery

Introduction

Red blood cell transfusion is thought to optimise the balance between tissue oxygen delivery and consumption and is a common practice in neonatal units[1]. Apart from independent association with mortality in preterm infants[2,3], retrospective reports have suggested association between blood transfusion and necrotising enterocolitis (NEC)[4-6]. This has led to the examination of the authenticity of transfusion associated NEC[7]. A systematic review of eleven case control studies and one cohort study has suggested the association of recent exposure of blood transfusion with NEC[8]. A small case series (n=4) study using near infra-red spectroscopy (NIRS) has shown abnormal splanchnic tissue oxygenation patterns possibly indicating mesenteric ischaemia in infants who developed NEC following transfusion[9]. Three randomised controlled trials examining the benefits of transfusion using high or low haemoglobin thresholds have informed on the development of NEC following blood transfusion[10-12]. Though not significant, NEC was found to be less common in infants in the liberal transfusion group (pooled OR 1.67; CI 0.82, 3.38)[13], thereby contradicting the association between blood transfusion and NEC. The association between blood transfusion and development of NEC still remains uncertain and open to conjecture.

Doppler ultrasound scan has been used to assess splanchnic perfusion in preterm infants [14]. Superior mesenteric artery (SMA) blood flow velocity measured by Doppler ultrasound scan has been used by researchers to investigate splanchnic blood flow[15,16]. Tissue oximetry measurements such as oxy/deoxy-hemoglobin (HbO_2/HHb) concentrations, tissue hemoglobin index (THI) and tissue oxygenation index (TOI) or regional tissue oxygen saturation (rSO_2) can be measured using Near-Infrared Spectroscopy (NIRS) [17], and fractional tissue oxygen extraction (FTOE) can be calculated using measured peripheral arterial saturation (SaO_2) and regional tissue oxygenation [18]. Researchers have used these

measurements as a biomarker to recognise the need for blood transfusion in newborn infants[14].

Neonatal circulation goes through complex adaptive mechanisms during the first few weeks of life, and hence the effect of transfusion on intestinal circulation could be variable in different postnatal ages. Tissue oximetry and blood flow response to blood transfusion in various postnatal ages may provide the vital information about pathogenic mechanism of developing transfusion associated NEC. The aim of this study was to investigate whether splanchnic blood flow and oximetry response to blood transfusion in preterm infants varies with postnatal age using Doppler ultrasound scan and NIRS.

Methods

The study was conducted at Homerton University Hospital, a tertiary neonatal unit in London, UK. Preterm infants receiving blood transfusion for clinical indication were eligible. A pragmatic sample size of 60 infants was selected with an aim to recruit 20 infants each to postnatal age groups of 1 to 7 (Group 1), 8 to 28 (Group 2) and ≥29 (Group 3) days of life [18]. In accordance with the British Committee for Standards in Haematology guideline [19], blood transfusion was indicated during the first week, second to fourth week and after fourth week of life if Hb (Hct) is less than 12g/dl (0.36), 10g/dl (0.30) and 8g/dl (0.25) respectively, based on results from arterial, venous or capillary blood sample. The decision for transfusion was made by attending clinical team depending on infant's Hb, ventilation status and/or oxygen requirement (FiO_2 >0.35). Fifteen ml/kg of leukocyte depleted, cytomegalovirus negative, Sickle cell negative, plasma reduced and cross matched packed red blood cell (haematocrit 50-70%) was transfused over a period of 3 hours through an intravenous cannula; the ongoing feeding regime was uninterrupted during the transfusion.

Intestinal Doppler ultrasound scan measurements

The superior mesenteric artery (SMA) peak systolic and diastolic velocities were measured 30-60 minutes before and after blood transfusion using a 7 MHz Doppler probe with range-gated pulsed wave Doppler ultrasound scanner (Logic P6, GE Healthcare, US). The measurements were performed by a single operator (JB) to minimise intra-operator variability (intra-class correlation coefficient for SMA PSV 0.77, p=0.02 and mean difference 0.02). A mean of five cardiac cycles was taken for Doppler measurements.

Intestinal or splanchnic oxygenation measurements

Intestinal or splanchnic tissue haemoglobin index (sTHI) is a measurement of total haemoglobin content in the tissue illuminated by Near Infra-Red (NIR) light. Splanchnic or intestinal tissue oxygenation index (sTOI) is an estimation of oxygenated haemoglobin in the tissue in comparison to total haemoglobin expressed in percentage[20]. Splanchnic fractional tissue oxygen extraction (sFTOE) is calculated as follows: sFTOE = (SaO_2 – sTOI)/SaO_2 and is expressed in percentage. It signifies the interaction between tissue oxygen availability and extraction [18]. The sTHI in arbitrary units and sTOI in percentage were measured using a NIRS device (NIRO 300, Hamamatsu Photonics K.K., Japan) with a sample acquisition rate of 6Hz (samples/sec). The NIRS probe was placed over the hypogastrium in the midline and held in place with a single use tourniquet (Vygon 'Vene K' Quick Release, Vygon UK Ltd.). The NIRS measurements were started from 15-20 min before transfusion, and continuously measured until 15-20 min post blood transfusion. Simultaneously oxygen saturation (SaO_2) was also measured.

The study was approved by the National Research Ethics Committee (REC no.12/LO/0527) and was adopted as an NIHR portfolio study (NIHR Study ID 13594). Informed written parental consent was obtained.

Data analysis

A mean for 15 minute epochs of NIRS oximetry measurements were determined for each infant using mathematical software Matlab (Math works, USA) during the following time periods: T1 - 15 to 20 minutes before the start of the blood transfusion, T2 - 1 hour into blood transfusion, T3 - 2 hour into blood transfusion and T4 - 15 to 20 minutes post blood transfusion. The mean of these epochs were then compared using repeated measures ANOVA with Bonferroni correction. The pre and post-transfusion values of all other measurements were compared using paired (two-tailed) t-test. The measurements between the feeding groups, and between gestational and postnatal age matched infants with and without PDA were also compared. A multivariate analysis was performed to explore the association between the Doppler flow and NIRS measurements with gestational age, postnatal age, pre-transfusion Hb, mean blood pressure (MBP), feeding volume and presence of PDA. A p value of <0.05 was considered significant. The data was analysed using SPSS 22.0 software (IBM Corp., USA).

Results

Infant characteristics

Fifty nine infants were studied; infant and maternal characteristics at birth and on the day of transfusion are presented in **Table 1**. The median pre-transfusion haemoglobin was higher in Group 1 compared to Group 2 ($p=0.03$, 95% CI 0.07 to 1.56) and Group 3 ($p<0.001$, 95%

CI 1.25 to 2.88). Thirty-two infants had PDA on echocardiography on the day of transfusion, of these only six were >14 days of postnatal age; otherwise normal cardiac morphology. Three infants in group 1, two in group 2 and one in group 3 were on single inotropic support (Dopamine) for hypotension, the dose remained unchanged for the duration of the measurements. Ten infants in group 1 were unfed, rest were receiving hourly bolus nasogastric feeds, and all except one (preterm formula) were on maternal expressed breast milk (MEBM). Two infants in group 2 were unfed, two were receiving 2 hourly and rest were on hourly bolus feeds with MEBM. Two infants in group 3 were unfed, majority were fed with MEBM, one was fed formula, and all were on 1 to 2 hourly feed. Majority of the infants in Group 1 (1-7 days) were receiving antibiotics for presumed sepsis at the time of blood transfusion. Though 38% in Group 2 and 44% in Group 3 were receiving antibiotics none of these infants had culture positive sepsis. None of the infants in the study developed NEC during their stay in the neonatal unit. None of the infants developed any transfusion related complication.

The mean pre and post blood transfusion heart rate, respiratory rate, saturation (SaO_2) and blood pressure are presented in **Table 2**. There was no significant change in HR, RR and SaO_2 following blood transfusion in all three groups of infants. Systolic blood pressure (BP) increased significantly in Group 1 infants, where as diastolic and mean BP increased significantly in all three groups following blood transfusion **(Table 2)**.

Doppler measurements

The mean pre-transfusion baseline superior mesenteric artery (SMA) peak systolic velocity (PSV) showed an increasing trend with postnatal age (group 2 vs. group 1, p=0.09) and was significantly higher in group 3 infants compared to group 1 (p<0.01; CI 0.6, 0.1). The SMA

diastolic velocity was similar between the postnatal groups. The mean pre-transfusion SMA PSV decreased following blood transfusion in all the three groups but this was not significant (**Table 3**). The SMA diastolic velocity remained unaltered following transfusion in all the three postnatal age groups.

Doppler measurements and feeds

Comparing Doppler measurement parameters in relation to feeds, the pre-transfusion SMA PSV was found to be significantly higher in infants who were fed >50% of total fluid volume (n=32) compared to those received feed <50% of total fluid volume (n=27) (0.91±0.35 vs. 0.71±0.35 m/sec; p<0.01). The pre-transfusion SMA PSV was higher in infants fed >50% of total fluid volume but not significant on multivariate analysis using confounding factors such as PDA, gestational age and chronological age. The SMA PSV decreased following transfusion but this was not significant in both the feeding groups (**Figure 1**). The pre-transfusion SMA diastolic velocity was similar in both feeding groups (p=0.89) and showed no significant change post-transfusion (p=0.79).

SMA Doppler measurements in infants with PDA

SMA Doppler measurements of infants with PDA (n=11, mean gestational age=25 wks & mean postnatal age=16 days) were compared to gestational age (mean=26 wks) and postnatal age (mean=17 days) matched infants with closed PDA (n=11). The pre-transfusion baseline mean SMA PSV was significantly higher in those infants with closed PDA (0.94±0.4 vs. 0.68±0.3 m/s, p = 0.006, CI 0.07, 0.45). The pre-transfusion SMA diastolic velocity was similar between the two PDA groups. The SMA PSV remained unchanged following transfusion in both PDA (p=0.29, CI -0.05,0.15) and closed-PDA group (p=0.19, CI -

0.04,0.20). The SMA diastolic velocity also remained unchanged post-transfusion in the PDA groups.

NIRS measurements

Intestinal or splanchnic tissue haemoglobin index (sTHI)

The sTHI levels increased consistently during transfusion in all three groups and the pattern of increase was identical except in the first hour of transfusion in Group 3 (**Figure 2**). While the maximal increase in sTHI happened later in Group 3, the percentage increase maximised post-transfusion in all the three groups ($p<0.001$; **Table 4**).

Intestinal or splanchnic tissue oxygenation Index (sTOI)

The mean pre-transfusion sTOI was significantly higher in group 2 infants compared to group 1 (44.6 vs. 36.7 %; p=0.03, 95% CI -0.6, -15.2). The mean pre-transfusion sTOI showed an increasing trend over time but it did not reach statistical significance until the end of transfusion (**Table 4, Figure 2**). The baseline sTOI increased by 42%, 29% and 30% following transfusion in group 1, group 2 and group 3 infants respectively.

Intestinal or splanchnic fractional tissue oxygen extraction (sFTOE)

The mean pre-transfusion sFTOE was significantly lower in group 2 compared to group 1 (64.7 % vs. 51.4%, p=0.02, CI 1.1, 17.6). The mean pre-transfusion sFTOE decreased significantly post-transfusion in all the three groups (**Table 4**).

NIRS measurements and feeds

The pre-transfusion sTHI was similar between the infants receiving >50% feeds compared to those receiving <50% feeds (38.2±13.8 vs. 31.8±7.9, CI -0.2,12.8; p=0.06). Similarly, the pre-transfusion sTOI was comparable between the two groups of infants (43.1±8.7% vs. 39.3±13%, p=0.23, CI -2.5,10.1). The sTOI and sTHI increased and sFTOE decreased significantly post-transfusion in both feeding groups (p<0.001).

NIRS measurements in infants with PDA and those with closed PDA

The baseline mean pre-transfusion sTOI was similar in the PDA group (44.7±10.2 %) compared to the closed-PDA group (43.1±10.1%; p=0.73, CI -7.6,10.7). The sTOI and sTHI increased significantly in all the time points at 1 hour (T2), 2 hours (T3) and post-transfusion (T4) when compared to baseline pre-transfusion values in both the groups with and without PDA (**Figure 3**). The sFTOE decreased in both groups post-transfusion.

Multivariate analysis

The pre-transfusion SMA PSV and sTOI in the postnatal age group infants was not significantly associated with confounding factors such as gestational age, birth weight, pre-transfusion Hb, volume of feed, PDA and mean blood pressure. The pre-transfusion SMA PSV and sTOI as well as the degree of changes following blood transfusion was not associated with the confounding factors in gestational and chronological age matched infants with and without PDA. The changes in SMA PSV and sTOI following blood transfusion in the postnatal age group infants was also not influenced by gestational age, birth weight, feeding volume, pre-transfusion Hb, PDA and mean blood pressure on multivariate analysis.

Discussion

We have shown that blood transfusion in preterm infants increased splanchnic TOI and THI and decreased FTOE during the first week (group 1), 8th to 28th day (group 2) and ≥28 days (group 3) of life thereby demonstrating a true improvement in the balance between tissue oxygen delivery and extraction post-transfusion. These changes were not influenced by gestational age, birth weight, pre-transfusion Hb, volume of feeds, PDA and blood pressure. This finding indicates that blood transfusion improves gut tissue oxygenation balance irrespective of postnatal age. Bailey et al also reported a significant increase in $srSO_2$ in preterm infants more than seven days of age (n=30; mean postnatal age 31.7±16.2 days) from a baseline 41.3±2.2% to 48.2±2.5% following transfusion[21]. Dani et al studied $srSO_2$ changes in preterm infants (n=15; mean postnatal age 32±23 days) and noted similar changes (pre-transfusion 54±12% to 70±8% post-transfusion)[18]. Mintzer et al reported an increase in splanchnic regional oxygenation ($srSO_2$) and decrease in fractional tissue oxygen extraction (FTOE) following transfusion by studying 10 preterm infants during the first week of life, and speculated that these NIRS parameters could be used to evaluate the relationship between oxygen delivery and consumption[22].

The pre-transfusion SMA PSV in our study was significantly higher in older postnatal age group infants and this may reflect maturational change and could also be due to higher feed intake as reported by Havranek et al[23]. However, on multivariate analysis the pre-transfusion SMA PSV was not associated with feeding volume. Similar to Dani et al [18] we noticed a decreasing trend but no significant change in SMA flow velocity post-transfusion in all groups. Though not comparable to our measurements, Nelle et al reported a significant decrease in coeliac artery flow velocity following blood transfusion in clinically stable preterm infants (mean gestational age 29 ± 5 weeks and mean postnatal age 48 ± 21 days)[24].

In concurrence to previously reported studies [23] we have shown that blood flow in the splanchnic circulation was higher in the pre-transfusion state in the predominantly fed infants. There was a significant increase in sTOI and decrease in sFTOE following transfusion in both the groups irrespective of the amount of feeds; this indicates improvement in the balance between tissue oxygen delivery and extraction following blood transfusion irrespective of the amount of feeds. Studies have shown that normal post-prandial rise in SMA PSV gets attenuated immediately following transfusion but this normalises by 24 to 48 hours following transfusion[25,26]. These and other retrospective studies[27,28] have implicated increased risk of NEC following transfusion in infants receiving feeds during transfusion, but no infant developed features of NEC following transfusion in our study.

We have demonstrated that splanchnic tissue oxygenation increases and fractional tissue oxygen extraction decreases without altering mesenteric blood flow velocities following blood transfusion irrespective of the presence of PDA. Studies have reported attenuated intestinal blood flow response to feeding in infants with large PDA (PDA to Left pulmonary artery ratio >1)[29] thereby speculating haemodynamically significant PDA to be an important risk factor for NEC. A small study of preterm infants with significant PDA receiving blood transfusion have shown attenuated mesenteric blood flow at least 4 hours after transfusion[30]. This is the first report on the interaction of PDA on the effect of blood transfusion on gut circulation by simultaneously measured splanchnic tissue oximetry and blood flow.

Though results from the current study indicate that blood transfusion improves oxygen delivery to the splanchnic tissues its clinical implications should be interpreted carefully. None of these infants were severely anaemic (**Table 1**) for a prolonged period and hence the splanchnic tissue may not have been ischaemic pre-transfusion protecting them from NEC. Pooled data from the retrospective studies have shown that the infants who developed NEC

following transfusion compared to those with classical NEC had lower pre-transfusion haematocrit (26% vs. 32%) [31]. Singh et al have also reported low haematocrit was an independent risk factor for development of NEC[6] which corresponds with the reduced risk of NEC in infants in the liberal haemoglobin threshold group indicated in the randomised controlled trials [13].

The attending clinical team decided on transfusion based on measured Hb and clinical condition of the infant, hence, we cannot exclude selection bias. Infants in the group one were studied while receiving their first blood transfusion. Infants in the second and third group may have received blood transfusions in the past. The exact number of transfusions previously received, age of infant at transfusion and age of donor red cells were not collected. The mean pre-transfusion Hb of the infants studied was 10g/dl. Recently, severe chronic anaemia was found to be associated with NEC [32], and hence our study findings may not be applicable for infants managed by restrictive blood transfusion practice. We aimed to recruit 20 infants to each group; managed to recruit 20 to group 1 and 2 but 18 infants to group 3 (≥29 days of life). This is unlikely to influence the study findings. Compared to other reported studies that combined infants of various postnatal ages [18,21], we have recruited infants into three postnatal age groups, to minimise the impact of adaptive physiological haemodynamic changes on the study findings. Fewer babies were ventilated in group 3 compared to group 1 and 2. However, the pre and post transfusion blood gas pH and pCO_2 were similar in all three groups of infants studied, and hence ventilation status is unlikely to have impact on the study findings. Six infants were receiving Dopamine (5mcg/kg/min) but this is unlikely to influence the study findings as the dosage of Dopamine infusion remained unchanged for the duration of the measurements. The splanchnic oxygenation measurements of seven infants were excluded from the analysis due to motion artefacts, which is comparable to other reported NIRS studies [33,34]. We measured the splanchnic tissue oxygenation upto 20 minutes following transfusion to facilitate post-transfusion

Doppler measurements. Other researchers have measured splanchnic tissue oxygenation up to 1[18] and 12[21] hours post-transfusion, and reported persistence of increased tissue oxygenation state following transfusion in more stable preterm infants.

We have demonstrated that blood transfusion improves intestinal tissue perfusion without altering mesenteric blood flow velocity irrespective of postnatal age, volume of feeds and presence of PDA. Pre-transfusion splanchnic tissue oxygenation and mesenteric blood flow varied with postnatal age. NIRS may be a useful non-invasive bedside monitoring tool to detect early signs of compromised oxygenation-extraction balance in mesenteric tissue. The effects of severe chronic anaemia and blood transfusion on gut perfusion in appropriately grown as well as growth restricted preterm infants need to be investigated.

Acknowledgements:

Babies and parents who participated in the study

Doctors and Nurses Neonatal unit, Homerton University Hospital

Mr Darius Khatibi, Medical Technical Office, Homerton University Hospital

Dr Kyriakos Iliadis – Consultant Paediatric Radiologist, Homerton University Hospital for helping with setting up and training of Doppler measurements

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Contributorship - Dr J Banerjee and Dr Narendra Aladangady conceived of the study, and with Dr Terence Leung prepared the study design, protocol and ethics application and received hospital R&D approval. JB and NA consented patients; JB performed the Doppler scans and NIRS measurements and data collection. JB conducted statistical analysis supported by NA and TSL. First draft of the manuscript was prepared by JB and was reviewed and approved by all authors.

Figure legends:

Figure 1. Blood transfusion and changes in SMA peak systolic velocity (PSV) in relation to percentage of feeds compared to total fluid volume.

* p<0.01 Comparison between baseline pre-transfusion measurements

Figure 2. Blood transfusion and changes in splanchnic tissue oxygenation (sTOI) and splanchnic tissue haemoglobin index (sTHI).

Group 1: birth to 7th day, Group 2: 8 – 28 days, Group 3: ≥29 days of postnatal age

Figure 3. Blood transfusion and Splanchnic Tissue Oxygenation Index (sTOI) and Tissue Haemoglobin Index (sTHI) in relation to PDA

List of Tables:

Table 1. Maternal and infant characteristics at birth and on the day of blood transfusion

Characteristics	Group 1 (1 – 7 ds) n = 20	Group 2 (8 – 28 ds) n = 21	Group 3 (>28 ds) n = 18
Gestational age (completed weeks)*	26 (23 – 27)	25 (23 – 30)	26 (24 – 34)
Birth weight (grams)*	763 (600 – 1180)	740 (600 – 1240)	793 (520 – 1746)
Chronological age (days)*	5 (1 – 7)	14 (8 – 27)	45 (29 – 93)
Haemoglobin at birth (g/dl)*	14.5 (9.8 – 20.7)	14.7 (10.0 – 17.4)	15.3 (10 – 18.9)
Maternal PET†	3 (15)	5 (24)	4 (22)
IUGR†	3 (15)	5 (24)	4 (22)
Chorioamnionitis†	9 (45)	8 (38)	8 (44)
Antepartum haemorrhage†	6 (30)	8 (38)	4 (22)
Antenatal steroids†	17 (85)	20 (95)	16 (89)
Weight at transfusion (grams)*	774 (700 – 1180)	805 (680 – 1250)	1125 (887 – 2045)
Pre-transfusion Hb (g/dl)*	11.0 (8.5 – 13.1)	10.3 (7.7 – 12.2)	9.2 (7 – 10.9)
Total fluids (ml/kg/d)*	150 (90 – 180)	150 (100 – 180)	165 (100 – 180)
Total feeds (ml/kg/d)*	18 (0 – 70)	120 (0 – 180)	155 (0 – 180)
Invasive/Non-invasive ventilation/nasal cannula oxygen or breathing in air†	13 (65)/7(35)	13 (62)/7 (33)/1 (5)	6 (33)/9 (50)/3 (17)
Presence of PDA†	19 (95)	12 (57)	1 (6)
Presumed sepsis on antibiotics†	19 (95)	8 (38)	8 (44)

* Median (Range), † Number (percentage)

Table 2. Blood transfusion (BT) and physiological parameters

Vital and laboratory parameters Mean (SD)	Group 1 (1 – 7 days) n = 20			Group 2 (8 – 28 days) n = 21			Group 3 (>28 days) n = 18		
	Pre-BT	**Post-BT**	**p value**	**Pre-BT**	**Post-BT**	**p value**	**Pre-BT**	**Post-BT**	**p value**
Heart rate (bpm)	159.1 (8.8)	157.1 (15.1)	0.67	153 (13.4)	153 (14.9)	0.99	150.0 (11.7)	149.4 (13.0)	0.90
Respiratory rate (bpm)	53.2 (12.3)	50 (11.7)	0.13	48.5 (10.2)	48.4 (8.3)	0.91	52.8 (13.9)	52.1 (11.4)	0.73
Arterial saturation (SaO2)%	93.2 (2.9)	93.2 (2.5)	0.96	91.9 (3.5)	92.3 (4.0)	0.67	93.0 (3.8)	93.2 (4.1)	0.88
Systolic BP (mm of Hg)	46.7 (6.6)	51.6 (4.9)	<0.01	54.9 (9.6)	57.7 (11.7)	0.07	62.2 (14.0)	63.7 (12.1)	0.45
Diastolic BP (mm of Hg)	24.3 (3.1)	30.7 (4.7)	<0.01	31.4 (5.4)	35.8 (8.3)	<0.01	31.3 (6.0)	36.2 (6.6)	0.01
Mean BP (mm of Hg)	32.7 (3.7)	37.9 (3.7)	<0.01	39.9 (6.3)	43.4 (8.1)	0.02	43.2 (7.9)	46.2 (6.6)	0.02

Table 3. Blood transfusion (BT) and Superior Mesenteric Artery (SMA) Doppler blood flow parameters in postnatal age groups

Blood flow parameters **Mean (SD)**	**Group 1 (1 – 7 days)** **n = 20**			**Group 2 (8 – 28 days)** **n = 21**			**Group 3 (>28 days)** **n = 18**		
	Pre-BT	**Post-BT**	**p value**	**Pre-BT**	**Post-BT**	**p value**	**Pre-BT**	**Post-BT**	**p value**
SMA peak systolic velocity (m/sec)	0.63 (0.32)	0.59 (0.23)	0.51	0.81 (0.33)	0.73 (0.24)	0.22	0.97 (0.40)	0.88 (0.32)	0.32
SMA diastolic velocity (m/sec)	0.12 (0.05)	0.12 (0.04)	0.65	0.13 (0.04)	0.12 (0.04)	0.45	0.13 (0.04)	0.12 (0.02)	0.37

Table 4. Blood transfusion (BT) and splanchnic NIRS parameters in postnatal age groups

Cerebral oximetry parameters **Mean (SD)**	**Group 1 (1 – 7 days)** **n = 17†**			**Group 2 (8 – 28 days)** **n = 20††**			**Group 3 (>28 days)** **n = 15†††**		
	Pre-BT	**Post-BT**	**p value**	**Pre-BT**	**Post-BT**	**p value**	**Pre-BT**	**Post-BT**	**p value**
Splanchnic tissue haemoglobin index (sTHI) (percentage increase from baseline) %	Zeroed baseline	39.4	0.001	Zeroed baseline	45.4	0.001	Zeroed baseline	47.5	0.001
Splanchnic tissue oxygenation index (sTOI) %	36.7 (19.3)	52.1 (20.8)	0.01	44.6 (10.4)	57.6 (14.3)	0.01	41.3 (10.4)	53.8 (16.5)	0.01
Splanchnic fractional tissue oxygen extraction (sFTOE)%	60.7 (13.4)	44.4 (20.3)	0.004	51.4 (11.5)	37.0 (14.9)	0.005	55.6 (11.8)	42.7 (15.1)	0.0004

† 3 infants, †† 1 infant and ††† 3 infants excluded from this analysis due to motion artefacts

Figure 1. Blood transfusion and Superior mesenteric artery (SMA) peak systolic velocity (PSV)
Group 1: birth to 7th day, Group 2: 8 – 28 days, Group 3: ≥29 days of postnatal age

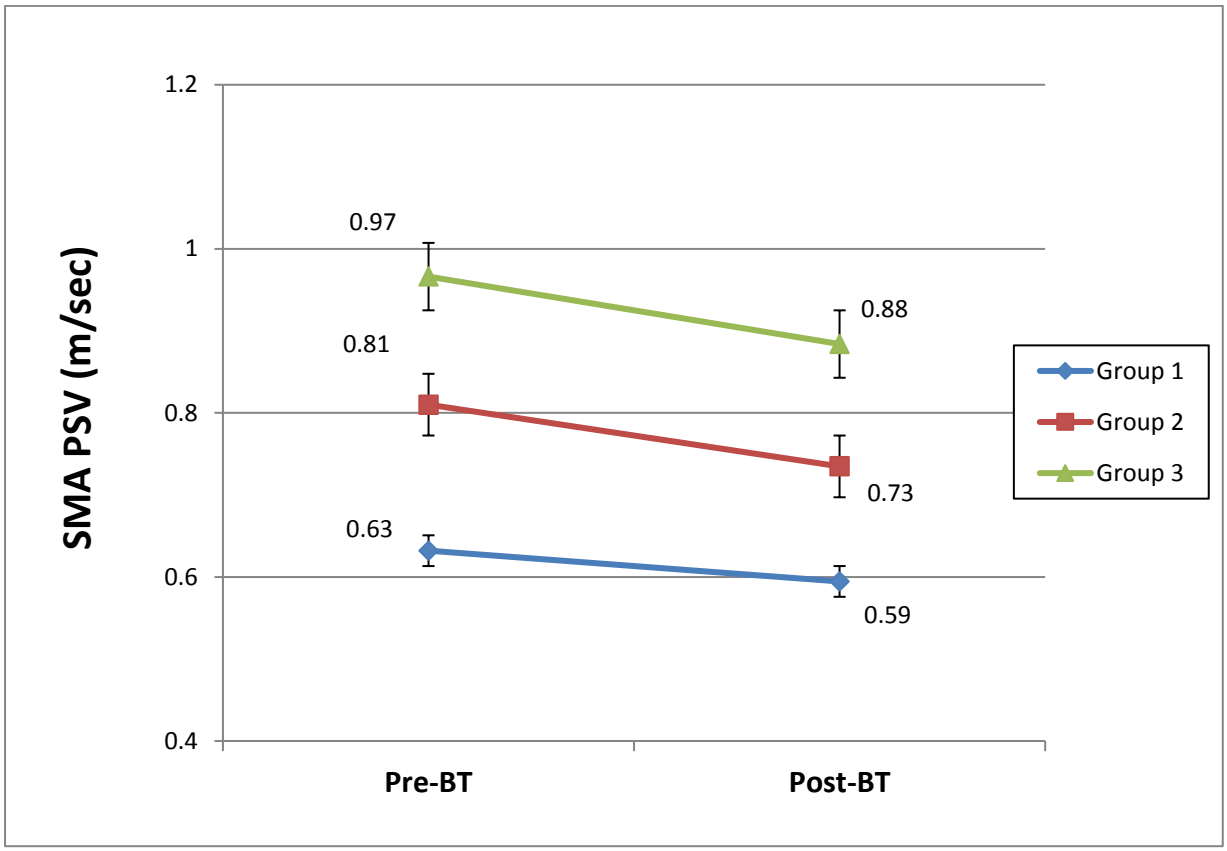
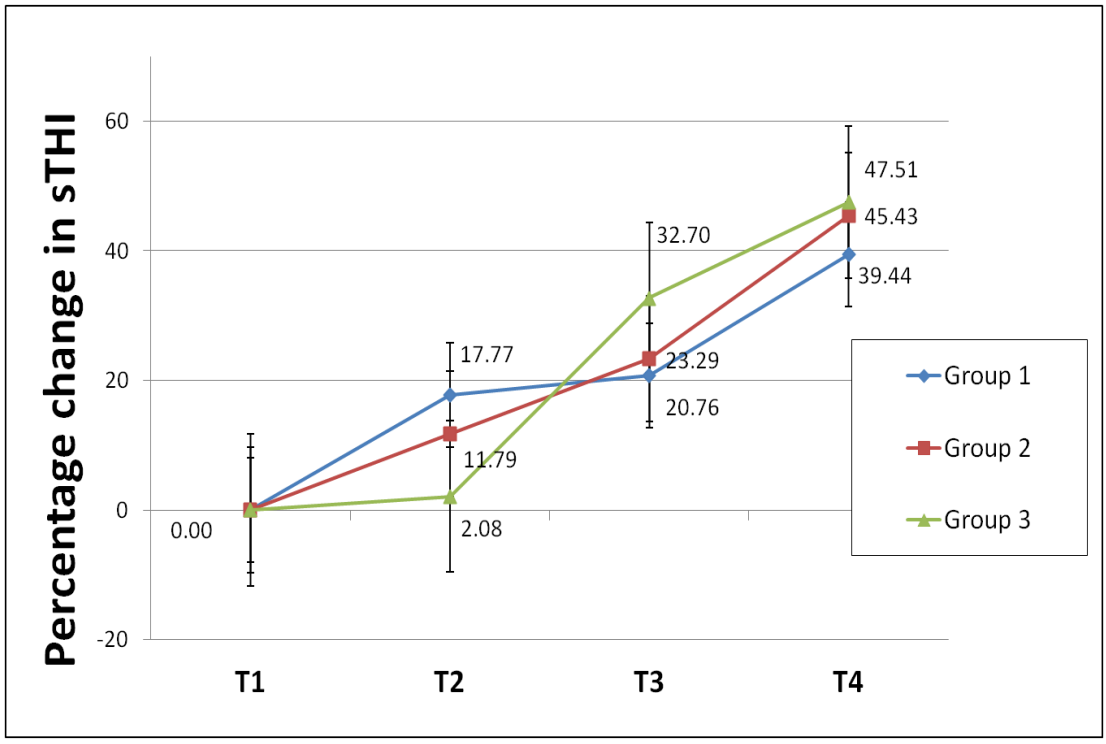


Figure 2. Blood transfusion and changes in splanchnic tissue oxygenation (sTOI) and splanchnic tissue haemoglobin index (sTHI).

Group 1: birth to 7th day, Group 2: 8 – 28 days, Group 3: ≥29 days of postnatal age



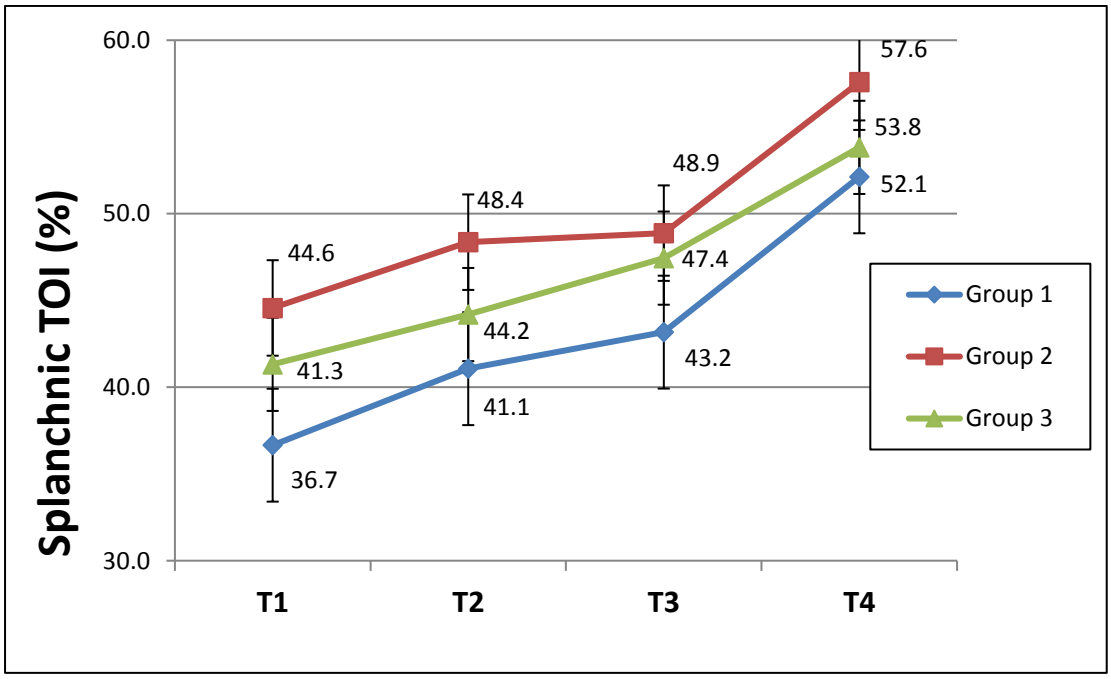


Figure 3. Blood transfusion and Splanchnic Tissue Oxygenation Index (sTOI) and Tissue Haemoglobin Index (sTHI) in relation to PDA