Fetal Programming

Low Birth Weight Infants Do Not Have Capillary Rarefaction at Birth
Implications for Early Life Influence on Microcirculation

Rohan D’Souza, Rajendra P. Raghuraman, Preetha Nathan, Isaac T. Manyonda, Tarek F.T. Antonios

See Editorial Commentary, pp 768–769

Abstract—Low birth weight predicts adult essential hypertension and is linked to increased cardiovascular mortality in adult life. A reduction in capillary density (ie, rarefaction) is a hallmark of essential hypertension, and evidence suggests that rarefaction precedes the onset of the rise in blood pressure, because it is found in normotensive individuals at high risk of developing hypertension, suggesting that rarefaction is likely to be a primary structural abnormality. We hypothesized that low birth weight infants would have significant capillary rarefaction at birth. We studied 44 low birth weight infants born to normotensive mothers (33 were born preterm, birth weight: 1823±446 g; and 11 were born at term, birth weight: 2339±177 g) and compared them with 71 infants born at term with normal weight (birth weight: 3333±519 g). We used orthogonal polarized spectroscopy to measure basal (ie, functional) and maximal (ie, structural) skin capillary densities. Low birth weight infants, whether born preterm or at term, had significantly higher functional capillary density (mean difference of 10.5 capillaries per millimeter squared; 95% CI: 6.6–14.4 capillaries per millimeter squared; \(P<0.0001\)) and higher structural capillary density (mean difference of 11.1 capillaries per millimeter squared; 95% CI: 7.6–14.5 capillaries per millimeter squared; \(P<0.0001\)) when compared with normal weight term infants. We conclude that low birth weight infants born to normotensive mothers do not have capillary rarefaction at birth. These results contradict what might have been predicted from the concept of the intrauterine origins of adult disease and suggest that microcirculatory abnormalities observed in individuals of low birth weight occur in postnatal life rather than during their intrauterine existence. (Hypertension. 2011;58:847-851.)

Key Words: capillary rarefaction ■ microcirculation ■ hypertension ■ low birth weight

Microcirculatory abnormalities and impaired tissue perfusion have been implicated in the pathogenesis of essential hypertension, obesity, diabetes mellitus, and insulin resistance.¹ In essential hypertension in particular, a reduction in the spatial density of arterioles and capillaries (ie, rarefaction) appears to play a central role.² We have shown previously that much of the capillary rarefaction in essential hypertension is attributed to the structural (ie, anatomic) absence of capillaries.³ We have also shown significant capillary rarefaction in patients with borderline intermittent essential hypertension and in normotensive offspring of hypertensive parents, suggesting a familial predisposition in which capillary rarefaction represents a primary vascular abnormality that antedates the onset of sustained elevation of blood pressure (BP).⁴,⁵

There is now a strong body of evidence showing that low birth weight (LBW), that is, birth weight of <2.5 kg regardless of gestation or causation,⁶ is associated with structural and functional vascular abnormalities,⁷ the development of essential hypertension,⁸,⁹ diabetes mellitus,¹⁰ and increased cardiovascular mortality in adult life.¹¹ Based on these associations and our own findings that capillary rarefaction antedates the onset of sustained elevation of BP, we hypothesized that capillary rarefaction would be demonstrable in LBW infants at or soon after birth. To test this hypothesis we studied functional and structural capillary densities in LBW and in normal birth weight (NBW) infants born at term to normotensive mothers.

Methods

Participants
This single-center observational study was performed at St George’s Hospital and was approved by the Wandsworth Research Ethics

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From the Blood Pressure Unit and Department of Clinical Sciences (R.D., R.P.R., P.N., T.F.T.A.), St George’s, University of London, London, United Kingdom; Department of Obstetrics and Gynaecology (R.D., I.T.M.), St George’s Hospital National Health Service Trust, London, United Kingdom. I.T.M. and T.F.T.A. contributed equally to the study. R.D. recruited all of the subjects and, together with P.N., conducted the observations and maintained the database. R.P.R. helped in the analysis and wrote the first draft with R.D. All of the authors discussed the results and implications and commented on the article at all stages.
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Committee. Written informed consent was obtained from all of the parents. Pregnant mothers were recruited from antenatal clinics, the day assessment unit, antenatal and postnatal wards, and the neonatal unit. We recruited a total of 279 consecutive infants irrespective of their birth weight in a study investigating capillary microcirculatory abnormalities in newborn infants of normotensive and hypertensive pregnant mothers. A total of 69 infants were excluded because of difficulty obtaining high-quality capillary images mainly because of motion artifact. In this study we included 44 LBW infants (ie, birth weight <2.5 kg) and 71 NBW infants born to mothers who were normotensive at the booking visit and throughout pregnancy. All of the infants were singletons. Gestational age was determined by ultrasound dating in the first trimester. Infants with neonatal asphyxia, congenital malformations, or chromosomal abnormalities were excluded from the study. Dietary habits and family history of diabetes mellitus, ischemic heart disease, stroke, hypercholesterolemia, and hypertension was obtained from both parents of the infants.

Orthogonal Polarized Spectroscopy
Infants were studied in their bed after they had been fed and while asleep on the postnatal ward or the neonatal unit. We used orthogonal polarized spectroscopy to examine skin capillary density at the plantar surface of the infant’s big toe. We cleansed the skin by several applications of a sticky tape (3M). We then applied a drop of high-quality mineral oil. Four microscopic fields, 0.62 mm² each, were recorded continuously for 30 seconds using the Cytoscan Device (Cytometrics, Philadelphia, PA), with ×10 objective, final magnification ×300. 12 Images were stored on a DVD recorder (Sony RDR-GX120) and were counted offline using the CapiScope computer software (KK-Technology). The number of all capillaries (ie, with stagnant and with continuously flowing red blood cells) was counted and double-checked by 2 investigators (P.N. and R.D.) independently. Basal capillary density, which represents functional capillary density, was calculated as the mean of these 4 microscopic fields. We used venous congestion to maximize the number of visualized perfused skin capillaries by applying a neonatal BP cuff around the calf muscles of the same leg. The cuff was then inflated and maintained at 30 mm Hg for 2 minutes, and further images were recorded to determine maximal capillary density, which represents structural (anatomic) capillary density. Skin and room temperatures were monitored during the study using YSI Tele-thermometers. In LBW infants who were born preterm, capillaroscopy was performed on the median age of 4 days and in NBW infants on median age of 1 day. The main reason for this difference was that some of the preterm infants were being looked after in the neonatal intensive care unit, and it was not always possible to perform capillaroscopy until they were discharged to the neonatal ward.

Statistical Analysis
We used unpaired Student t test to compare the groups. Statistical significance was declared when the P value was <0.05. All of the statistical analysis was carried out using the SPSS 19. Multiple regression analysis was carried out to examine whether there was a significant relationship between capillary density and skin temperature, gestational age, birth weight, and head circumference.

Results
Table 1 shows the baseline characteristics of the mothers and their newborn infants. There was no statistical difference in age, systolic and diastolic BPs, body mass index, smoking history, previous history of preeclampsia, or family history of cardiovascular disease between the mothers of LBW infants and mothers of NBW infants. A total of 115 neonates were included in the study. There were 44 LBW infants (26 boys and 18 girls), and 71 NBW infants (38 boys and 33 girls). Among the LBW infants, 18 (40.9%) were white, 7 (15.9%) were Afro-Caribbean, 12 (27.3%) were South Asian, and 7 (13.7%) were mixed (15.9%) were of mixed ethnicity. Among the NBW infants, 40 (56.3%) were white, 12 (16.9%) were Afro-Caribbean, 8 (11.3%) were South Asian, and 11 (15.5%) were of mixed ethnic origin. Among the LBW infants, 35 (75%) were born preterm (ie, before 37 completed weeks’ gestation; mean gestational age: 32.6±2.8 weeks; mean birth weight: 1823±446 g), and 11 were born at term (mean gestational age: 38.1±1.0 weeks; mean birth weight: 2339±177 g). All of the infants in the NBW group were born at term (37–42 completed weeks’ gestation).

As expected, there was significant difference in birth weight between the LBW and NBW groups (mean difference: −1381 g; 95% CI: −1569 to −1193 g; P<0.0001). LBW infants also had significantly smaller head circumference (mean difference: −4.0 cm; 95% CI: −4.9 to −3.2 cm; P<0.0001) and shorter body length (mean difference: −2.3 cm; 95% CI: −4.0 to −0.6 cm; P=0.01; Table 1).

Capillary Density
LBW infants had significantly higher basal capillary density (mean difference: 10.5 capillaries per millimeter squared; 95% CI: 6.6–14.4 capillaries per millimeter squared; P<0.0001) and higher maximal capillary density (mean

<table>
<thead>
<tr>
<th>Variable</th>
<th>Low BW Infants (n = 44)</th>
<th>Normal BW Infants (n = 71)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Maternal data</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age, y</td>
<td>30.7±5.4</td>
<td>31.7±6.5</td>
<td>0.350</td>
</tr>
<tr>
<td>Blood pressure, mm Hg</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Systolic</td>
<td>115±9</td>
<td>115±11</td>
<td>0.984</td>
</tr>
<tr>
<td>Diastolic</td>
<td>73±8</td>
<td>71±8</td>
<td>0.212</td>
</tr>
<tr>
<td>Pulse rate, bpm</td>
<td>85±11</td>
<td>85±9</td>
<td>0.936</td>
</tr>
<tr>
<td>BMI, kg/m²</td>
<td>23.3±6.9</td>
<td>24.8±5.0</td>
<td>0.228</td>
</tr>
<tr>
<td>Primigravida, n (%)</td>
<td>25 (56.8)</td>
<td>38 (53.5)</td>
<td>0.753</td>
</tr>
<tr>
<td>Smokers, n (%)</td>
<td>6 (13.7)</td>
<td>12 (16.9)</td>
<td>0.829</td>
</tr>
<tr>
<td>History of preeclampsia, n (%)</td>
<td>2 (4.5)</td>
<td>5 (7.0)</td>
<td>0.278</td>
</tr>
<tr>
<td>Family history of IHD or HTN, n (%)</td>
<td>16 (36.4)</td>
<td>30 (42.3)</td>
<td>0.614</td>
</tr>
<tr>
<td>Neonatal data</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ethnicity, n (%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>White</td>
<td>18 (40.9)</td>
<td>40 (56.3)</td>
<td></td>
</tr>
<tr>
<td>Afro-Caribbean</td>
<td>7 (15.9)</td>
<td>12 (16.9)</td>
<td></td>
</tr>
<tr>
<td>South Asian</td>
<td>12 (27.3)</td>
<td>8 (11.3)</td>
<td></td>
</tr>
<tr>
<td>Other</td>
<td>7 (15.9)</td>
<td>11 (15.5)</td>
<td></td>
</tr>
<tr>
<td>Sex, girls, n (%)</td>
<td>18 (40.9)</td>
<td>33 (46.5)</td>
<td></td>
</tr>
<tr>
<td>Birth weight, g</td>
<td>1952±454</td>
<td>3333±519</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Gestational age, wk</td>
<td>34.0±3.5</td>
<td>39.7±1.2</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Preterm, n (%)</td>
<td>33 (75.0)</td>
<td>. . .</td>
<td></td>
</tr>
<tr>
<td>Length, cm</td>
<td>47.9±1.8</td>
<td>50.2±2.7</td>
<td>0.01</td>
</tr>
<tr>
<td>Head circumference, cm</td>
<td>30.6±2.3</td>
<td>34.6±1.8</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>

Data are mean±SD unless otherwise specified. IHD, ischemic heart disease; HTN, hypertension; BW, body weight; BMI, body mass index.

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Table 2. Capillaroscopy Data

<table>
<thead>
<tr>
<th>Variables</th>
<th>Low BW</th>
<th>Normal BW</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Skin temperature, °C</td>
<td>33.7±2.1</td>
<td>33.1±1.4</td>
<td>0.081</td>
</tr>
<tr>
<td>Room temperature, °C</td>
<td>27.3±4.4</td>
<td>27.5±1.7</td>
<td>0.743</td>
</tr>
<tr>
<td>Basal capillary density, per field (mm²)</td>
<td>36±6 (57±10)</td>
<td>29±6 (47±10)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Maximal capillary density, per field (mm²)</td>
<td>41±6 (65±10)</td>
<td>34±5 (54±8)</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>

Data are mean±SD. BW indicates birth weight.

Discussion

We report, for the first time, the existence of a strong inverse relationship between birth weight and both basal and maximal capillary densities, showing that LBW infants have significantly higher skin capillary density compared with their NBW counterparts. These results were unexpected, because based on the hypothesis of developmental programming and intrauterine origins of adult disease, the established presence of capillary rarefaction in essential hypertension, and our own reports of capillary rarefaction preceding the onset of hypertension, we had expected that LBW infants would show the capillary rarefaction seen in adults at high risk of developing essential hypertension,5,13 in whom LBW is a major risk factor. Our findings do not negate the hypothesis of the intrauterine origins of adult disease but suggest that the capillary rarefaction seen in adults with essential hypertension, although it might be programmed in utero, does not in fact occur during intrauterine life but does so remotely from birth, and, therefore, genetic factors rather than LBW, per se, may be involved, as has been shown recently.14

It is interesting to speculate on why LBW infants should have high capillary density. It is known that the microcirculation of the skin is subject to considerable changes in the first few days of extrauterine life.15 An extensive capillary network in the newborn’s skin forms in the first weeks after birth first in a disorderly manner, and then, between 14 and 17 weeks after birth, the length and variability of the diameter of the capillaries increase.16,17 This is consistent with the recent report by Top et al.,18 who examined the capillary microcirculation in the buccal mucosa in term neonates at 0 to 7 days, 8 to 28 days, 1 to 6 months, and at 3 years using orthogonal polarized spectroscopy imaging and found higher basal capillary density in infants <1 week old compared with older children, confirming that basal capillary density decreases progressively after the first week of life.18 This group did not study maximal capillary density given that they studied the buccal mucosa, and maximization would not have been possible. In LBW infants, the higher capillary density could be a compensatory response to improve tissue perfusion and nutrient delivery to counter the effects of the “insult” that caused the intrauterine growth restriction in the first place. After delivery, the abundant availability of nutrients presumably triggers a much more rapid and perhaps poorly controlled process of “capillary hyperpruning,” culminating in LBW infants having capillary rarefaction by some stage in later childhood. These postulations are supported by the limited research that has been undertaken in infants, young children, and young adults.

Bonamy et al19 used intravitral video-microscopy to study the skin basal capillary density on the dorsum of fingers in 60 children (aged 9.1–9.7 years), of which 39 were born very preterm (≤30 weeks’ gestation, and presumably with LBW) and 21 were born at term. They found that children who were born preterm had a significant 6.9% reduction in basal capillary density compared with children born at term. However, they included in their study preterm children born to mothers who developed preeclampsia during their pregnancy and who have been shown to have significant capillary rarefaction, which may influence capillary density in their offspring.20 Ijzerman et a21 studied nail fold capillaries in 21 healthy white prepubertal children (mean age: 8.6 years) who were born at term and found significant association between their birth weight and the increase in capillary recruitment during reactive hyperemia (ie, maximal capillary density − basal capillary density). Although they did not include any children with LBW in their study, they concluded that LBW is associated with impaired capillary recruitment independent.
of BP. It should be acknowledged, however, that not all studies have shown capillary rarefaction in individuals with a history of LBW. Irving et al,22 using intravital videomicroscopy, found no difference in basal or maximal capillary densities in young adults (mean age: 24 years) with LBW compared with control subjects. Goh et al23 examined skin capillary density in 17 LBW subjects and 21 high BW subjects at 3 months of age and found no difference in skin capillary density between the 2 groups. However they found that cutaneous microvascular vasodilatory function (the maximum hyperemic response) was lower in LBW infants compared with their high BW counterparts.

Overall the evidence points to an association between LBW and deviations from normality in the microcirculation in young subjects. Thus, Leeson et al24 found a correlation between flow-mediated dilatation and LBW in healthy school children, whereas Goodfellow et al25 found that young adults of LBW have an impairment in endothelium-dependent vasodilatation in the conduit arteries compared with NBW individuals. Martin et al26 documented impaired skin microvascular endothelium-dependent vasodilatation in 3-day–old LBW infants, as well as in prepubertal children (mean age: 9±1.3 years) with a history of LBW.7 Preterm birth was also associated with abnormal retinal vascularity in several studies, including narrower bifurcation angles,27 narrower retinal arterioles,28,29 and increased retinal arteriolar tortuosity.30 Retinal arteriolar narrowing has been demonstrated in several population-based cohorts to predict the subsequent development of essential hypertension.31–33

Could our findings simply be an artifact? One may postulate that LBW infants may have relatively less surface area and taut skin, therefore showing apparently higher capillary density. However, this is an unlikely explanation, because the plantar surface of the big toe is not thought to be affected by changes in body weight, because it is not a site for fat storage.

Several studies have suggested that microcirculatory abnormalities and, in particular, capillary rarefaction, may be considered as one of the putative mechanisms linking early life exposures and subsequent cardiovascular disease.4,5,13,34 LBW has been suggested as an independent risk factor for cardiovascular disease, because both clinical and epidemiological studies have shown that it is associated with an increased prevalence of insulin resistance, essential hypertension, and dyslipidemia, which predisposes to coronary heart disease.35–37 Barker has suggested that delayed fetal growth could lead to a reduced number of glomeruli and vascular cells, and the subsequent catch-up accelerated postnatal growth in the presence of plentiful nutrition, and salt could then result in the metabolic syndrome, diabetes mellitus, and essential hypertension.38,39

Longitudinal studies of capillary density in NBW and LBW infants commencing from birth to various stages in early childhood would define background physiological processes. In utero events are somewhat more challenging to elucidate. Although we have not addressed the issue in this article, it will also be necessary to tease out the impact of gestation versus birth weight. In conclusion, we found that LBW, whether at term or preterm, is not associated with capillary rarefaction but with high capillary density. These results suggest that early life events may have more influential effects on the microcirculatory abnormalities observed in hypertension.

Perspectives
Our study shows that LBW infants are born with higher capillary density than their NBW counterparts. The LBW infants are at higher risk of developing essential hypertension in adult life, and the capillary rarefaction seen in association with the essential hypertension clearly does not occur in utero but remotely from birth. Our findings suggest that early life events may have more influential effects on the microcirculatory abnormalities observed in hypertension. This has major implications for our understanding of the concept of the intrauterine origins of adult disease and clearly warrants further investigation.

Acknowledgments
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Disclosures
None.

References


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