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Complete Venous Blood Counts In Healthy Term Nigerian Neonates.

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ABSTRACT

Serial complete blood counts were performed on venous blood of 104 healthy, normal term Nigerian neonates (55 male, 49 females). The mean weight of the neonates was 3465±398g in the first 10 days of life. There were significant depressions in the value of total leucocyte count (8.37±2.38x10^9/L) on day 7-10 compared to values on day 1-3 (10.57±4.10x10^9/L) ($p<0.0l$). Similarly, the value of neutrophils (3.09±1.59x10^9/L) on days 7-10 was significantly reduced compared to values obtained during days 1-3 (6.77±3.9x109/L) (<0.01). However, elevations of lymphocyte count (4.89±1.80 x109/L), eosinophils counts (0.22±0.20x10 IL) and platelets count (241±50x10/L) on days 7-10 were observed compared to values of lymphocytes count (3.58±1.40 x109/L), eosinophils count (0.12±0.17x109/L) and platelets count (208±48x109/L) on days 1-3 of life ($p<0.01$ in each case). The haematocrit, monocytes count and basophils count; were not significantly different within the study period ($p>0.1$). The present study concludes that Healthy normal term Nigerian neonates presents with varying proportions of total and differential white cell and platelets counts within the first 10 days of life. This may have implications for neonatal haematology in our environment.

Key words: haematological parameters, term neonates, Nigeria

INTRODUCTION

The first published data on neonatal haemogram dated back to 1924 when Lippman first reported the morphologic and quantitative characteristics of blood corpuscles in newborn period [1]. Since then other researchers [2-6] from different parts of the world have determined normal haematological values during neonatal period. Although it has been shown that some haematological parameters (haemoglobin level and total leukocyte and neutrophil counts) are significantly higher in neonates than in infants and adults [7, 8], great variations exist in blood parameters in the newborn infants. These variations depend on gender, race, and socio-economic status of parents and as well as on health status of the neonates, types of sample used (cord, venous, capillary blood), methods used for the analyses (automated or manual) and time of blood sampling [9-11]. Studies have shown that haematological parameters (total white blood cell, red blood cell, and differential white blood cell counts, haematocrit, haemoglobin e.t.c.) are indices for measuring neonatal well being and can as well be used to predict the presence of disease [5,6,12]. For example, perinatal brain damage, intrauterine growth restriction or maternal-foetal ABO blood group incompatibility are associated with an increase in nucleated red blood cells in cord blood [13, 14]. It has also been shown that viral and bacteria infections were associated with alterations in total leucocytes, lymphocyte population and platelets counts [15]. While haemoglobin levels in healthy and sick term babies have been found to be significantly higher than those of healthy and sick pre-terms babies, platelet counts in sick term newborns are significantly lower than those of healthy term newborns [8]. However, platelet counts in both healthy and sick pre-term babies are significantly lower than those of term babies. In addition to variation in absolute values of haematological parameters, newborn also show some morphological changes in blood picture. For instance, the red cells of the newborn have been reported to be markedly macrocytic at birth with mean cell volume and diameter falling after the first week of life and reaching adults' values by the ninth week [16]. Furthermore, blood films from new born have been found to show normochromic cells, polychromasia and 3-5% of the red cells may show fragmentation, target cell formation and distortion of shape with markedly elevated numbers nucleated in the presence of haemolysis or hypoxia [17].

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Studies of neonatal haematology have been very limited in Nigeria and most were done on capillary and/or cord blood [18-201. Although Ogala [2] in his study used venous blood, estimation of blood parameters was done electronically.

Electronic counters are fast and more convenient than manual methods in the estimation of blood parameters but the financial implications of running it has made it inaccessible to most laboratories in Nigeria. Assuredly, independent evaluation of manual and electronic counter methods has deposed that manual methods provide a satisfactory measurement of haematological parameters [21,22]. This study is therefore aimed to provide manually determined baseline data of venous blood parameters in the new born infants in their first ten days of life and to see how the results compare with previously determined values in capillary and cord blood.

MATERIALS AND METHODS

This study was conducted at Obafemi Awolowo University Teaching Hospital Complex, Ile-Ife, Osun State, Nigeria. Our subjects were neonates delivered to Nigerian parents of various ethnic groups and socio-economic status. Neonates were included if they were singleton, delivered at full- term, either by normal vaginal delivery or by lower segment Caeserean Section (LSCS), normal and healthy without perinatal aphyxia or any known disorder within the period of the study. Exclusion criteria include multiple deliveries, pre-terms and small for gestational age (SGA) babies. The Ethical Committee of Obafemi Awolowo University Teaching Hospital complex, Ile-Ife, approved the protocol for the study. Before enrolment, informed consents were obtained from the parents of the participating neonates. Mothers of neonates were interviewed to obtain sociodemographic data such as age, parity, level of education and occupation. Infants' birth weights and gestational ages at delivery were obtained from case notes of the mothers. In all, one hundred and four neonates were recruited into the study. Venous blood (1-2.5 ml) was taken from the neonates from the dorsal veins between day 1-3 and day 7-10 post delivery. The blood samples were collected in plastic EDTA bottles, gently but thoroughly mixed and the analyses done within two hours of collection.

Haematocrit was determined by micro-method using capillary tube as described by Dacie and Lewis [23].

Total white blood cell count was done as in a standard text [23]. Briefly whole blood was diluted 1:20 in Turcks' solution (2% glacial acetic acid tinted with methyl violet). With a capillary tube, this was applied on charged haemocytometer and counted under light microscope at 40x objective. For differential white blood cell counts, dried thin films made from thoroughly mixed whole blood samples were stained by standard Leishman staining method. Platelets counts were done by Brecher and Cronkite method [21], using 1% ammonium oxalate as diluent and a settling time of 40 minutes. The level of education and/occupation of the parents assessed socio-economic status of infants.

STATISTICAL ANALYSIS

Data were analysed for means and standard deviations and significant test was done by students' t-test. Level of significance was considered as $p<0.05$, and $p<0.01$.

RESULTS

A total of one hundred and four neonates comprising fifty-five (52.9%) males and forty-nine (47.1%) females participated in the study. By ethnic origin, sixty (57.7%) of the neonates were Yoruba, twenty-nine (27.9%) Igbo, eight (7.7%) Hausa, and seven (6.7%) were other tribes. At birth, the mean weight of infants was 3465 $\pm$ 398g (range 3112-3886g) and mean gestational age 39.8$\pm$1.1wks (range 38.2-40.9wks). Fifty-two (50%) of the neonates were from lower socio-economic background; thirty-four (32.7%) and eighteen (17.3%) were from middle and upper socioeconomic class respectively. Of the 104 neonates, thirteen (12.5%) were delivered through lower segment Caesarean section while 91 (87.5%) were through normal vaginal delivery. Records on parity showed that 29 (27.9%) of the mothers were primigravidae, 42 (40.4%) have upto three children while the remaining 33 (31.7%) have more than three children. Also, sixty-eight (65.4%) of the mothers completed normal antenatal visits while 36 (3 4.6%) did not (data not shown). From table 1, total white blood cell count significantly decreased from 10.57 $\pm$ 4.10x10^9/L on day 1-3 to 8.37$\pm$ 2.38x10^9/L on day 7-10 ($p < 0.001$). While haematocrit remained fairly constant within the study period platelets counts increased from 208 $\pm$ 48x10^9/L on day 1-3 to 241 $\pm$ 50x10^9/L by day 7-10 ($p < 0.001$). Table 2 shows that while lymphocytes and eosinophils were significantly raised on day 7-10 (4.89$\pm$ 1.80(58.9%) and 0.22 $\pm$ 0.20(2.7%) from their initial values of 3.58 $\pm$ 1.40(37.4%) and 0.12 $\pm$ 0.17(1.2%) on

day 1-3 ($p< 0.001$), neutrophils were significantly depressed [(6.77±3.9 (60.3%) vs. 3.09 ± 1.59(37.2%), $p<0.001$]. However, monocytes and basophils did not show significant difference on day 1-3 and day 7- 1(($p>0.001$)]. The predominant blood pictures were macrocytosis, normocytosis and spherocytosis (data no shown).

Table 1: Haematocrit, Total white blood cell and Platelets counts in the first 10 days of post-natal life.

Ages/days	Hct ± SD(%)	Twbc ±SD x 10^9/L	Platelets ±SDx10^9/L
1-3 (n=104)	46.0±5.4	10.57± 4.10	208±48
7-10 (n=104)	45± 5.4	8.37± 2.38*	241±50*

Twbc = Total white blood cell; Hct= Haematocrit; *p <0.01.

Table 2: Differential white blood cell counts in the first 10 days of post-natal life.

Ages/day	Neut. (%)	Lymph. (%)	Mono. (%)	Baso. (%)	Eosin. (%)
1-3(n=104)	6.77±±3.9	3.58±±1.40	0.11±±0.12	0.01±0.04	0.12±0.17
Mean±SDx10^9/L	(60.3)	(37.4)	(1.0)	(0.1)	(1.2)
7-10 (n= 104)	3.09±± 1.59	4.89±± 1.80	0.08±0.11	0.02±0.06	0.22±0.20
Mean±± SDx10/L	(37.2)*	(58.9)*	(1.0)	(0.2)	(2.7)*

Neut. = Nuetrophils; Lymph.= Lymphocytes; Mono.= Monocytes; Baso. = Basophils.
Eosin.= Eosinophils; *P < 0.01.

DISCUSSION

This study has shown the mean haematocrit, total leucocytes count and Platelets count of healthy normal full-term Nigerian neonates. The values obtained for these parameters in the present study were lower than those quoted for Caucasians (Hct 47.0 ± 6.0%, total leucocyte count 12.3 ± 4.8x10^9/L, platelet count 269.9± 57.7x10^9/L) [4,5]. However, the values were in agreement with that reported by Alur and his co-researchers [9] in Ohio, U.S.A. and Scott-Emuakpor et. al [24] in African neonates. These results were also higher than those reported by Abdurrahaman and Adekoje [20] in northern Nigeria except for platelets (Hct= 42 %, total leucocytes 9.25 x10^9/L, platelet 173 x10^9/L). It then implies that the haematological values quoted in most of the western textbooks may not apply to Nigerian neonates. For instance the packed cell volume in the present study (45.5 ± 5.4%) in the first 10 days of life is lower than 60-62% quoted elsewhere [16]. Haematocrit is a measure of haemoglobin concentration and has been shown to vary with nutritional status; state of hydration and the type of sample used for its estimation [25]. The lower haematocrit observed in the present study may be partly attributed to intrauterine shift of fluid in infants as non of the neonate showed evidence of dehydration during the period of this study. It has been reported that intrauterine fluid shift may be triggered by the last stage of labour in infants delivered vaginally [26, 27]. However, the presence of suboptimal nutrition cannot be ruled out as it has been shown that in Africa, maternal nutritional status is a predictor of intrauterine growth and well being [28]. Our data showed that half of the neonates were delivered of mothers from lower socio-economic class who by implication may have sub-optimal nutrition especially the micronutrients. Additionally, antenatal non-compliance and multiparity were found in higher proportion of the mothers. Hence, the lower Hct values observed in the present study may also be partly attributed to some mothers not receiving the routine haematinics given during ANC visits or as a consequence of maternal depletion syndrome which was consequently transferred to their foetuses. Cord blood haemoglobin has been found to be lower in the

presence of low maternal haemoglobin and in newborn infants delivered by Cesarean section [29].

However, we could not establish a relationship between neonatal Hct and maternal nutritional status or parity. Nevertheless, the comparable values of Hct for both day 1-3 and day 7-10 showed that Hct was relatively stable during neonatal period and may not be affected by immediate extrauterine factors. It has also been shown that capillary blood samples have higher Hb, Hct, red blood cell (RBC), white blood cell (WBC), and lymphocyte counts than venous blood [25] thus suggesting that the lower Hct recorded in the present study may be related to the sample used (venous blood) and this underline the importance of considering the sample source when using haematological reference ranges. The significantly raised leucocytes in the first 3-day of life in recent study is consistent with previous findings [25]. This may be partly attributed to labour induced stress. In this study, 12.5% of the neonates were delivered by caesarean section while majority were through normal vaginal deliveries. Even though we did not have data on labour events, prolonged labour has been associated with increased leucocyte counts [30]. It has also been shown that mode of delivery may affect neutrophlil function or lymphocyte subpopulation [31]. Significantly high WBC counts in cord blood has been reported after vaginal delivery or vacuum extraction than after elective caesarean delivery [32]. Additionally, subclinical infections cannot be ruled out as the possible cause of leucocytosis in the present study even though our subjects were apparently healthy, as raised WBC has also been associated with neonatal infections [33]. It has been earlier reported [3,34] that neutrophils and lymphocytes account for about 85% of the white blood cell population by day 7, which is in corroboration with the present findings. Although haemoconcentration has been given as a probable cause of the elevated values of neutrophils especially in the first 3 days of life, this has been countered in favour of increased population of this leucocyte subset [35]. Accordingly, increase in lymphocyte subset may be attributed to displacement of neutrophils from marginal layer vessels, which has been established to occur in adults after violent exercise [36]. In the newborn, violent exercise can be likeed to personal labour during childbirth. The rise in lymphocyte count on day 7-10 may also be due to increased immunogenic stimulation in the extrauterine environment; probably in response to BCG vaccines as this period coincide with the time the first dose of the vaccine is given. Eosinophilia in day 7-10 in this study is in accord with earlier report [35] and may be associated with recovery from hyperadrenalism or shock state of birth and subsequent neonatal hypoadrenalism [37]. The reported platelet count in both day 1-3 and day 7- 10 in the present study is lower than that reported by earlier researchers for the same neonatal age but was not to the level of thrombocytopaenia [10,25]. The blood film pictures of the neonates in this study were consistent with reports of previous study [38]. However, reporting of manual blood picture is subjective and objections have been raised against its usefulness as values vary greatly according to the expertise of individuals performing the test. The present findings show that:

- Healthy normal tern Nigerian neonates have lower haematologieal parameters than their Caucasian counterparts in the first 10 days of life with relatively stable Hct.
- Total leucocyte count (dominated by neutrophils) was higher in the first three days of life and decreased subsequently with lymphocytes predominating.
- Eosinophils and platelets increased with neonatal age being higher in day 7-10 than in day 1-3.
- Macrocytic, normocytic and spherocytic red blood cells were common blood picture in healthy normal term Nigerian neonates. In conclusion, the haematological values recorded in this study were lower than those observed for Caucasiati neonates of the same age but the patterns were the same for both races. These findings may be of relevance in interpreting complete blood counts of neonates in this environment.

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