CLINICAL PRACTICE ARTICLE

Effect of Phototherapy on Neutrophil VCS Parameters and White Blood Cells

Nilgün Altuntas[1], Özlem Ceylan Dogan[2] and Fatih Mehmet Kislal[3]

ABSTRACT

Objective: To investigate the effect of phototherapy (PT) on WBC parameters and neutrophil volume, conductivity and scatter (VCS) parameters.

Study Design: Comparative cross-sectional study.

Place and Duration of Study: Keçiören Training and Research Hospital, Turkey, from October 2016 and January 2017.

Methodology: Term newborns who had received PT for indirect hyperbilirubinemia were inducted. Total serum bilirubin, neutrophil, eosinophil, basophil, monocyte, lymphocyte counts, and neutrophil VCS parameters before and after PT were compared.

Results: The mean age of the neonates at admission was 6.05 ±3.7 days. The mean gestational age at the time of birth was 37.44 ±2.09 weeks. The mean duration of PT was 46.37 ±17.00 hours. PT was associated with a significant increase in eosinophil (p=0.039) and basophil counts (p=0.034), a significant decrease in leucocyte (p=0.036) and neutrophil counts (p=0.031). There was no significant change in monocyte (p=0.79) and lymphocyte counts (p=0.93). There was a significant decrease in neutrophil volume values and a significant increase in neutrophil scatter values after PT. There was no effect of PT on neutrophil conductivity values.

Conclusion: PT affects some WBC components and neutrophil volume and scatter parameters. There is a need for further prospective clinical researches on this topic before starting to use neutrophil VCS parameters in the diagnosis of sepsis.

Key Words: *Phototherapy, Neutrophil, VCS parameters.*

INTRODUCTION

Indirect hyperbilirubinemia is the most common cause of neonatal jaundice. Bilirubin is one of the end products of heme catabolism. Even though bilirubin has antioxidant properties and protects the lungs and even the intestines,[1] it is also known to have some side effects. For this purpose, bilirubin reaching pathological values should be lowered. Phototherapy (PT) is the most commonly used treatment in newborns with indirect hyperbilirubinemia. PT provides the urine and bile excretion of bilirubin by converting bilirubin into its constitutive and structural isomers and colorless oxidation products.

Besides having clinically important benefit, many side effects of PT such as loose stools, hyperthermia, dehydration, skin burn, retinitis, low platelet count, increased red cell osmotic fragility, bronze baby syndrome, riboflavin deficiency, and DNA damage,[2] have been described till now. To date, the effects of PT on eosinophils, platelets, T and B lymphocytes have been investigated in some studies.[3,4] To the best of the authors' knowledge, there is no study in the literature that focuses on relationship between neutrophil volume, conductivity and scatter (VCS) parameters and PT in jaundiced newborns.

Some authors proposed that the morphologic changes seen in neutrophils in sepsis could be analysed by using the Coulter LH 750 with VCS technology.[5-8] The VCS technology of the Coulter LH 750 hematology analyser uses direct current impedance to measure cell volume of neutrophils, radio frequency opacity to measure conductivity for internal composition of neutrophils, and a laser beam to measure light scatter for cytoplasmic granularity and nuclear structure.[9,10] In real life setting, PT sometimes may be used in newborns with sepsis. Finally, if PT affects VCS parameters, it may cause misinterpretation of the results. Therefore, the aim of the study was to investigate the effect of PT on WBC parameters and neutrophil VCS parameters.

METHODOLOGY

The present study was a comparative cross-sectional study performed between October 2016 and January 2017 at Keçiören Training and Research Hospital, Turkey. This study was approved by the Local Medical Ethics Committee and was in accordance with the

[1] *Department of Pediatrics, Division of Neonatology, Ankara Yildirim Beyazit University, Ankara, Turkey*
[2] *Department of Biochemistry, Ankara University, Ankara, Turkey*
[3] *Department of Neonatology, Keçiören Training and Research Hospital, Turkey*

Correspondence: Dr. Nilgün Altuntas, Department of Pediatrics, Division of Neonatology, Ankara Yildirim Beyazit University, Ankara, Turkey
E-mail: nilgunaltuntas@hotmail.com

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Helsinki Declaration. Medical records of the last three years were evaluated and term babies, who admitted to our center's neonatal intensive care unit with the reason of indirect hyperbilirubinemia and underwent PT for at least 24 hours based on American Academy of Pediatrics (AAP) guidelines, were included in the study.

Babies in a pathological condition that could affect whole blood parameters (mothers of infants with a history of bleeding, preeclampsia or eclampsia, chorioamnionitis, chronic disease, PROM (premature rupture of membranes), diabetes hemolytic disease, thyroid disease, smoking, taking drugs in pregnancy such as anticonvulsant, antidepressant, insulin, chemotherapy or cortisone and neonates with birth asphyxia) were excluded from the study. Demographic characteristics of infants, WBC counts, total bilirubin values, and neutrophil VCS parameters pre- and post-procedure were determined by reviewing the files.

Statistical analyses were performed using the SPSS version 16.0 programme. The variables were investigated using Kolmogorov-Simirnov or Shapiro-Wilk's test to determine whether or not they are normally distributed. Presentation of parametric numerical data was given as mean ± standard deviation (SD), and frequency and percentage were computed for categorical data. Non-parametric numerical data was expressed as medians and interquartile range (IQR). Data before and after PT was compared with Wilcoxon test based on distribution of data. P-value smaller than 0.05 was considered statistically significant.

RESULTS

A comparative cross-sectional study was performed on 74 term newborns who received PT for indirect hyper-bilirubinemia. Demographic features of the patients are given in Table I. When whole blood parameters before and after PT were compared, PT was associated with a significant increase in eosinophil (p=0.039) and basophil counts (p=0.034), a significant decrease in leucocyte (p= 0.036) and neutrophil values (p=0.031). There was not a significant change in monocyte (p=0.79) and lymphocyte counts (p=0.93).

When neutrophil VCS parameters were evaluated, there was a significant decrease in mean neutrophil volume (MNV, p=0.008), a significant increase in mean neutrophil scatter (MNS, p=0.013). There was no effect of PT on mean neutrophil conductivity (MNC) values (p=0.981). Comparison of VCS parameters and WBC components before and after PT is given in Table II.

Table I: Demographic features of the patients.

Parameters	
Gender	
Male n (%)	45 (61.81%)
Female n (%)	29 (39.18%)
Mean birth weight (grams)*	3084 ±539
Mean gestational age (weeks)*	37.44 ±2.09
Mean age at admission (days)*	6.05 ±3.7
Mean duration of phototherapy (hours)*	46.37 ±17.00

**Mean ±SD*

Table II: Comparison of VCS parameters and whole blood components before and after phototherapy.

Parameters	Before PT	After PT	p-value
Leucocyte (/mm3) *	12300 (9900-14750)	11000 (9250-13400)	0.036
Neutrophil (/mm3) *	4400 (2875-7600)	3950 (2375-5250)	0.031
Lymphocytes(/mm3) *	5000 (3775-6600)	5150 (4200-6250)	0.935
Eosinophils (/mm3) *	500 (400-600)	500 (400-900)	0.039
Monocytes (/mm3) *	1500 (1100-1900)	1400 (1100-1925)	0.79
Basophil (/mm3) *	100 (0-200)	100 (0-300)	0.034
Neutrophil VCS parameters			
MNV*	142.25 (138.38-145.60)	140.70 (135.82-144.55)	0.008
MNC*	137.15 (134.78-141.72)	137.20 (133.98-142.62)	0.981
MNS*	128.70 (123.78-135.05)	131.90 (126.55-137.27)	0.013

*PT: Phototherapy; MNV: Mean neutrophil volume; MNC: Mean neutrophil conductivity; MNS: Mean neutrophil scatter. * Median (interquartile range (IQR)).*

DISCUSSION

Neutrophil VCS parameters have been used as an indicator of bacterial sepsis in recent years.[5-8] Chaves *et al*. studied peripheral blood samples from 69 patients with positive blood cultures for bacteria and 35 control subjects.[5] They observed a significant increase in MNV and a significant decrease in MNS from septic patients compared with control subjects. An elevation of MNV was associated with a higher WBC count and percentage of neutrophils; and was present even in patients who did not have leukocytosis or neutrophilia. They believed that MNV has a potential to be an additional indicator for acute bacterial infection.[5] In another study, Suresh *et al*. found a significant increase in MNV, a significant decrease in MNS from patients with both systemic and localised infections as compared with control subjects.[6] They did not find any significant change in MNC values.[6] Shen *et al*. studied MNV and MNS parameters in patients with drug-induced neutro-penia and found significant increase in MNV, as well as a significant decrease in MNS in neutropenic patients approximately one week prior to development of neutropenia compared to healthy controls as well as to case controls.[11] Neutrophil VCS parameters were also studied in newborns. Çelik *et al*. observed significant increases in MNV and significant decreases in MNC and MNS in septic newborns.[8] There were significant decreases in MNV, whereas MNC and MNS increased at the end of the treatment. In this study, gram-negative sepsis caused higher MNV than gram-positive sepsis.[8]

In this study, PT affected neutrophil VCS parameters. There was a significant decrease in MNV, a significant increase in MNS. There was not any effect of PT on MNC. Though PT is frequently used in neonatal intensive care units, and if it really affects VCS parameters, it may cause misinterpretation of the results

in diagnosing septic patients who underwent PT. The present results demonstrated that PT may lead to conflicting effects in MNV and MNS, so diagnosing sepsis may be overlooked. As MNC is not affected by PT in this syudy, it can be used in differentiation. The authors propose that further prospective clinical researches should be done on this topic before starting to use neutrophil VCS parameters in diagnosing sepsis.

The effect of PT on WBC components was also studied. It demonstrated a significant increase in eosinophil counts of patients who underwent PT. To the authors' knowledge, there is only one study that studied the effect of PT on eosinophil counts.[3] They observed a significant increase in eosinophil counts with PT. They interpreted that high levels of bilirubin may induce a decrease in eosinophil levels by suppressing VCAM-1. Thus, any treatment that aims to decrease total serum bilirubin levels may be expected to increase the levels of eosinophil. They also commented that bilirubin is protective against allergic disease and that PT might trigger allergic disease. There are some studies to support this claim. These studies showed that jaundice and/or PT increase risk of asthma later in life.[12-14] The present results with regard to the effect of PT on eosinophil counts were compatible with results of this study. Additionaly, a significant decrease was revealed in leucocyte counts. So decreased leucocyte counts might be important as assessing peripheral white blood cell count in septic patients.

The results of this study are subjected to some limitations. First, the authors did not get a control group consisting of healthy term newborns who did not need PT. Had there been a control group, one could also make comments about the effects of bilirubin. Second, it is a single centre study with a relatively small sample size, which might underestimate or overestimate the relationship between PT and neutrophil VCS parameters. More specifically designed prospective studies are needed to externally cross-validate our findings in a larger cohort of these patients.

CONCLUSION

PT affects some WBC components and neutrophil volume and scatter parameters. There is a need for further prospective clinical researches on this topic before starting to use neutrophil VCS parameters in the diagnosis of sepsis.

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