PEER REVIEW HISTORY

ARTICLE DETAILS

TITLE (PROVISIONAL)	Mean Platelet Volume in Preterm Infants as A Predictor of Late Onset Neonatal Sepsis: a retrospective comparative study.
AUTHORS	Leibovitch, Leah; Zohar, Hagar; Gavri-Beker, Ayelet; Goshen, Abigail; Strauss, Tzipora

VERSION 1 - REVIEW

REVIEWER NAME	***Gili Kenet***
REVIEWER AFFILIATION	Shiba Medical Center, Hematology
REVIEWER CONFLICT OF INTEREST	I work at the same institution as the authors
DATE REVIEW RETURNED	17-May-2024

GENERAL COMMENTS	In their retrospective study authors evaluated the role of platelet MPV as a potential marker and maybe predictor of sepsis in neonates, matched by gestational age and birth weight. This is an interesting study that found coerrelation between MPV, throbocytopenia and increased CRP among infants diagnosed with sepsis due to Gram positive/ negative bacteria or fungi. Authors also state there were more C sections among study group as compared to controls. Could authors speculate regarding the reasons? Were CS performed due to maternal reasons or fetal distress? Were mothers suffering from fever/ inflamation prior to CS?Was there a difference among the subgroups of sepsis with different causative agents? The manuscript could benefit from slight shortening of background data and native English editing

REVIEWER NAME	***Dror Mandel***
REVIEWER AFFILIATION	Tel Aviv Medical Center, Neonatology
REVIEWER CONFLICT OF INTEREST	None
DATE REVIEW RETURNED	26-May-2024

GENERAL COMMENTS	The study aims to examine an important morbidity on an important population of vulnerable very low birth preterm infants. Comments: 1. This is a nice study on a very important topic in Neonatology. However, there are many comorbidities that might influence PLT and MPV such as NEC, SIP, IVH, asphyxia. The existence of these should be carefully examined and in particular the time frame between them and the sepsis episode. For example, for NEC – there were 5 out of 68 (7.5%) infants in this group – what is the time frame between NEC occurrence and sepsis occurrence? Sepsis might be a consequence of NEC and vice versa. The same is true for IVH and PLT count and MPV value. Was sepsis documented

	during day 4-5-6 for example? And what was the impact of IVH that was present (if at all) on theses exact days on PLT and MPV? 2. The definition of late onset sepsis should be clarify. 72 hours cutoff for late onset sepsis is controversial. Why didn't you use the 7 days cutoff for defining LOS? 3. The range for all parameters should be mentioned. 4. A flow chart for inclusion of infants to the study is needed. How many deliveries, how many were excluded from the study and the reasons for exclusion, etc. 5. One of the reasons for neonatal thrombocytopenia is IUGR- did you exclude them from the study? Please comment on this topic as IUGR might influence PLT and MPV. 6. Results: Please use the same definition for the study group-it alternates between study and sepsis group. 7. The sepsis group had significant higher incidence of prematurity complication such as BPD,IVH.ROP and NEC-. A discussion on this difference is recommended. 8. Are there any differences in your results between bacteria and fungi infection? Between gram negative and positive? 9/

VERSION 1 – AUTHOR RESPONSE

Reviewer: 1

Gili Kenet, Shiba Medical Center

Authors also state there were more C sections among study group as compared to controls.

1. Could authors speculate regarding the reasons?

Thank you for that comment. During revision of our data we have found several typos in the data sheet. We have corrected them and there were no statistical differences between the study and control group regarding mode of delivery. We have changed it in the table and results. (Table 1 and page 7)

2. Were CS performed due to maternal reasons or fetal distress?

About a third of the CS were born due to maternal indication (36.5% in the sepsis group, 31.7% in the control group). Fetal distress as an indication for CS was 15.8% in the sepsis group compared to 7.94% in the control group. Our study focused on late onset sepsis and it seems that CS indication did not affect the incidence of LOS.

3. Were mothers suffering from fever/inflammation prior to CS?

Maternal fever as an indication for CS was noted in 4 mothers in the sepsis group and 2 mothers in the control group. This might affect early onset sepsis which is beyond the scope of our study.

4. Was there a difference among the subgroups of sepsis with different causative agents?

Thank you for the comment. Among the positive blood cultures :28 (44.4%) were gram negative bacteria, 32 (50.8%) were gram positive bacteria and 3 (4.8%) were fungi as mentioned in Page 7 first paragraph. The number of the subgroup were too small for statistical analysis

5. The manuscript could benefit from slight shortening of background data

Thank you for the comment, we have shortened it as suggested.

6. Native English editing

Thank you, we have revised as suggested.

Reviewer: 2

Dr. Dror Mandel, Tel Aviv Medical Center

Comments:

This is a nice study on a very important topic in Neonatology. However, there are many comorbidities that might influence PLT and MPV such as NEC, SIP, IVH, asphyxia.

1. The existence of these should be carefully examined and in particular the time frame between them and the sepsis episode. For example, for NEC – there were 5 out of 68 (7.5%) infants in this group –

Thank you for the comment. We have checked the time frame between IVH, NEC and the sepsis event. In the sepsis group there were 4 cases of IVH and in only 1 case the sepsis occured at the same day of the IVH. In the 3 others the intervals were 3,11, and 12 days apart from the IVH.

2. What is the time frame between NEC occurrence and sepsis occurrence?

We have checked the time frame between NEC and the sepsis events. In the sepsis group we had 5 cases of NEC, and in only 1 case the sepsis occurred at the same day of the NEC and in the 3 others the interval were 8,14,18 and 25 days apart

3. Sepsis might be a consequence of NEC and vice versa. The same is true for IVH and PLT count and MPV value. Was sepsis documented during day 4-5-6 for example?

Thank you. See our answer for questions 1 and 2.

4. And what was the impact of IVH that was present (if at all) on theses exact days on PLT and MPV?

Only 1 case of IVH on day 12 of life had sepsis on the same day. In this case we found a thrombocytopenia of 44,000 and MPV 10.5 in a CBC taken a few days before the IVH occurred. The patient had platelet transfusions due to thrombocytopenia and on the day of sepsis his platelet count was 117000 with an MPV of 12.6.

5. The definition of late onset sepsis should be clarify. 72 hours cutoff for late onset sepsis is controversial. Why didn't you use the 7 days cutoff for defining LOS?

Thank you for the comment. Neonatal sepsis is broadly categorized into early sepsis, caused by organisms acquired perinatally, and late onset sepsis (LOS), which appears after 3 calendar days from birth [Definied by "Identifying Healthcare Associated Infection, January 2024], generally due to post-natal hospital-acquired organisms. It was added it in page 3

6. The range for all parameters should be mentioned.

Thank you- It is mentioned in Page 4

7. A flow chart for inclusion of infants to the study is needed. How many deliveries, how many were excluded from the study and the reasons for exclusion, etc.

During the years 2016-2022 there were 68 cases of proven sepsis of which 5 were excluded due to a lack of data.

8. One of the reasons for neonatal thrombocytopenia is IUGR- did you exclude them from the study? Please comment on this topic as IUGR might influence PLT and MPV.

Thank you for the comment. IUGR were included if they didn't have persistent thrombocytopenia. In the sepsis group we had 18 (28.6%) infants defined as IUGR and 6 (9.5%) in the control group. Indeed, as expected the IUGR infants had a higher risk and incidence of LOS. We have also mentioned that in the Methods section page 5.

Our NICU is characterized by a relatively high proportion of IUGR babies.

9. Results: Please use the same definition for the study group-it alternates between study and sepsis group.

Thank you- done

10. The sepsis group had significant higher incidence of prematurity complication such as BPD,IVH.ROP and NEC-. A discussion on this difference is recommended.

Thank you for that comment. During revision of our data we have found several typos in the data sheet. We have corrected them and there were no statistical differences between the study and control group regarding prematurity complication. (Table 1)

11. Are there any differences in your results between bacteria and fungi infection? Between gram negative and positive?

Among the positive blood cultures :28 (44.4%) were gram negative bacteria, 32 (50.8%) were

gram positive bacteria and 3 (4.8%) were fungi as mentioned in Page 7 first paragraph. The

number of the subgroup were too small for statistical analysis.

VERSION 2 – REVIEW

REVIEWER NAME	***Dror Mandel***
REVIEWER AFFILIATION	Tel Aviv Medical Center, Neonatology
REVIEWER CONFLICT OF INTEREST	NA
DATE REVIEW RETURNED	21-Jul-2024

GENERAL COMMENTS	The authors answered my questions in the response letter, but I did not see any reference to any of my questions within the article itself. The messages that arise from the questions should be embedded within the article itself.

REVIEWER NAME	***Indrani Bhattacharjee***
REVIEWER AFFILIATION	Tufts University School of Medicine, Newborn Medicine
REVIEWER CONFLICT OF INTEREST	none
DATE REVIEW RETURNED	06-Aug-2024

GENERAL COMMENTS	Authors are recommended to clarify Time Intervals: Clearly define the time intervals for pre-infection, intra-infection, and post-infection to avoid ambiguity. 2. Could the authors attempt to clearly convey the reason for dividing the post infection as 1-7 days and 10-14 days? Is this with respect to platelet life span? Is there an expected change in setting of sepsis? What about other confounders like maternal complications which might effect platelet counts such as Preclampsia/ FGR etc

VERSION 2 – AUTHOR RESPONSE

Reviewer: 2

Dr. Dror Mandel, Tel Aviv Medical Center

Comments to the Author

The authors answered my questions in the response letter, but I did not see any reference to any of my questions within the article itself. The messages that arise from the questions should be embedded within the article itself.

Sorry for that and thank you for the remark. It was added and highlighted in pages 3,5,7 according to the comments in our previous respond.

Reviewer: 3

Dr. Indrani Bhattacharjee, Tufts University School of Medicine

Comments to the Author

1. Authors are recommended to clarify Time Intervals: Clearly define the time intervals for pre-infection, intra-infection, and post-infection to avoid ambiguity.

In our department routine CBC is taken every 7-10 days and extra blood test according to clinical condition.

Intra infection is the day of clinical symptoms and sepsis workup that was done including CBC CRP and blood culture.

Pre-infection – we were looking in retrospect for the CBC within 7 days before the sepsis.

Post infection –we were looking in retrospect on the CBC during the first week after the sepsis and on the CBC 10-14 days post sepsis.

2. Could the authors attempt to clearly convey the reason for dividing the post infection as 1-7 days and 10-14 days? Is this with respect to platelet life span? Is there an expected change in setting of sepsis?

Thank you for the remark - Indeed we have looked at the CBC within the week post-infection and on day 10-14 post sepsis since it was available and present platelets life span (we add a comment on page 6 and 7 and table 2).

What about other confounders like maternal complications which might effect platelet counts such as Preeclampsia / FGR etc

Thank you for the important remark - Maternal complication such as fever, eclampsia might affect the newborn immediately after birth with thrombocytopenia or IUGR. Our study focused on late onset sepsis where usually, the maternal conditions are not affecting the infant’s condition anymore. We added it in the result section page 7.