

A study of Protein C level and Activated Partial Thromboplastin Time among neonates suffering of sepsis-Omdurman Maternity Hospital, Sudan.

ALBARA AHMED (✉ albaraa@fajr.edu.sd)

ALFAJR COLLEGE FOR SCIENCE AND TECHNOLOGY <https://orcid.org/0000-0003-3245-7585>

Babiker Mohammed

Karary University

Abdelhalim M Nasr

University of Bahri

Wafa I Elhag

Elneelain University

Awadelkareem Abass

University of Khartoum

Research note

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Abstract

Objectives: The study aimed to assess Activated Partial Thromboplastin Time (APTT) and Protein C (PC) among Sudanese neonates with sepsis (Cases) in Omdurman maternity hospital, Sudan compared with healthy neonates (controls) for recognizing haemostatic alteration in APTT and PC, considering the gender, gestational age, delivery mode among both groups, case group categorized additionally regarding; sepsis onset, outcome, and Gram stain typing of the causative agent.

Results: A total of 100 neonates divided equally into septic cases and healthy controls, died neonates were constituted 10 (20%) while 40 (80%) were recovered among case group.

APTT showed significant prolongation in septic neonates compared to controls (mean; 47.8 and 37.5 sec for cases and controls, respectively) P. value was 0.00. Among case group; dead neonates showed significant prolongation compared recovered (mean; 61.5 and 44.4sec) P.value 0.00. PC showed significant decrease in dead neonates compared to recovered (mean; 25.4 and 36.2% for dead and improved) p value 0.04. APTT&PC can be useful as marker of neonatal sepsis mortality.

Insignificant difference in PC reported between case and control group. None of the gender, gestational age, delivery mode, sepsis onset, and causative agent showed significant correlation with APTT and PC.

Introduction:

Neonatal sepsis is a global challenge causing high morbidity and mortality in neonates ^(1, 2, 3, 4). Although various microorganisms can cause neonatal sepsis, bacteria are the most common cause of neonatal sepsis in the world ^(2, 4). In all countries where data on hospitalization for sepsis are available, numbers increased steadily ⁽⁵⁾. In the U.S sepsis accounts for far more deaths than deaths from prostate cancer, breast cancer and Acquired Immune Deficiency Syndrome (AIDS) combined ⁽⁶⁾. In developing countries; sepsis accounts for 60–80% of lost lives annually, affecting about 6 million newborns and children ⁽⁷⁾. Pathophysiological mechanism of sepsis is not completely understood, but coagulation alterations are hallmark of the syndrome ⁽⁸⁾. Both inflammation and hemostasis are interrelated pathophysiologic processes that considerably affect each other, inflammation leads to activation of hemostatic system that in turn also considerably influences inflammatory activity ⁽⁹⁾.

The study aimed to assess activated partial thromboplastin time (APTT) and Protein C (PC) among Sudanese neonates with sepsis (case group) compared to healthy neonates (control) and to correlate gender, delivery mode, and gestational age with such coagulation parameter among both groups. Also to correlate APTT and PC with outcome, sepsis onset, and Gram stain typing of the causative agent among case group in order to study haemostatic alteration among neonates with sepsis.

Methods:

The study was prospective cross sectional hospital based study, conducted in Omdurman Maternity hospital, Sudan from June.2013 to April.2015 on 100 Sudanese neonates divided into; cases (neonates with proven sepsis by blood culture), and controls (healthy neonates).

Blood culture for identification of microorganism was done, positive culture included as case sample. Venous neonatal blood collected and plasma prepared for APTT and PC assessment.

APTT procedure

APTT assessed by clotting procedure; 50 microliter of plasma was obtained, then a metal ball added, then 50 microliter of cephaloplastin added, and incubated for 180 seconds, then 0.02 M calcium chloride was added (all reagent were pre-warmed at 37C for 15 minutes), then immediately clotting time was counted by semi-automated coagulometer (Stago Stat-4. France), calibrator (Uricalibrator, Stago. France) was used as control.

PC procedure:

50 microliter of 1/10 diluted (with Owren-Koller buffer) plasma was obtained, then a metal ball was added, then 50 microliter of PC deficient plasma (protein C deficient plasma, Stago. France) was added, 50 microliter of PC activator (highly purifies extract of Agkistrodon contortrix venom. Stago. France) was added and incubated for exactly 180 seconds, then 0.02 M of STA calcium chloride was added (all reagent were pre-warmed at 37C for 15 minutes), then clotting time was counted using semi-automated coagulometer (Stago Stat-4. France), calibrator (STA Uricalibrator. Stago. France) was used as control.

Data analysis:

Data was entered, tabulated, and analyzed by SPSS (statistical program for social and science) IBM SPSS Statistics 20.

Results:

A total of 100 venous neonatal blood samples (50 for both groups) were included. Gender distributions were; 26 females (52%), and 24 males (48%), 27 females (54%), and 23 (46%) for case and control group respectively. In gestational age, case and control groups were classified into term (37 week gestational age and above) and preterm (36 or less). 17 neonates were term (34%), and 33 were preterm (66%), 1 term neonate (2%) and 49 preterm (98%) for case and control.

Groups were categorized according to delivery mode; caesarean section and normal vaginal delivery neonates. 17 were delivered normally (34%), and 33 were delivered by cesarean Sect. 66%), 7 were delivered normally (14%), and 43 were delivered by cesarean sections (86%) for case and control.

Case group was classified considering their outcome; dead and recovered neonates. Dead were constituted 10 (20%), 40 were recovered (80%).

Case group was classified according to sepsis onset into; early onset (0–7 days) represented 17 (34%) and late onset (7–28 days) represented 33 (66%).

Gram stain typing of causative agent among case group were classified; Gram negative 41 (82%), Gram positive 9 (18%). Mortality among sepsis onset distributed among case group into; early 4 (40%) and late onset 6 (60%).

APTT prolonged in case group compared to control (47.9 and 37.5 second) (P.value 0.00).

PC decreased in case group compared to control (34.4 and 36.8%) (P.value 0.41). Table (1).

Table (1): APTT and PC mean and P.value among both groups.

Parameter	Mean	P.value
APTT (case)	47.8 sec	0.000
APTT (control)	37.5 sec	
PC (case)	34.3%	0.412
PC (control)	36.8%	

Outcome showed significant correlation of APTT and PC among case group. (P.value; 0.00 and 0.04 respectively) Among case group; APTT were significantly prolonged and protein C was significantly decreased in dead neonates compared to recovered. Table (2).

None of gender, gestational age, delivery mode, and Gram stain typing of causative bacterial gent showed significant correlation with APTT and PC.

Table (2): APTT and PC mean and P.value considering outcome among case group.

Parameter	Mean	P.value
APTT (Dead)	61.5 sec	0.00
APTT (Recovered)	44.4 sec	
PC (Dead)	25.4%	0.04
PC (Recovered)	36.2%	

Discussion:

Systemic inflammation is followed by coagulation system activation, and conversely, components of coagulation system significantly affect inflammatory response⁽¹⁰⁾. Significant prolonged APTT was in line with Krishna I et al⁽¹¹⁾ and Anggraini.D et al⁽¹²⁾ who found prolonged APTT in sepsis patients. APTT significantly decrease in dead neonates, this finding was in line with Christian Niederwanger et al⁽¹⁴⁾ and Benediktsson.S et al⁽¹³⁾ who observed APTT prolongation in patients with severe sepsis is associated with increased mortality. The outcome correlate with PC, dead septic neonates have significant lower PC

than recovered, this result was in line with Bhat.R et al ⁽¹⁵⁾ who concluded significant correlation between PC decrease and sepsis deaths. Prolongation of APTT and decreased PC indicates activation of intrinsic coagulation proteins and PC with continuation of activation of both, consume APTT (intrinsic factors)& PC, these goes in line with Disseminated Intravascular Coagulation (DIC), which is one of primary causes of sepsis deaths.

Conclusion:

APTT was significantly prolonged in neonatal sepsis (P.value 0.00). APTT also prolonged significantly in dead septic neonates compared to recovered one (P.value 0.00). PC decreased significantly in dead neonates compared to recovered in case group (P.value 0.04). (Prolonged APTT and decrease PC can be useful as a marker for neonatal sepsis mortality.

Limitation:

Firstly; limited resources locally, in term of Facility (financial) resources, and personnel (nurse, phlebotomist, and other health profession) in Neonatal Intensive Care Unit (NICU) and the equipment facility (the study is self-funded). Secondly; poor community health awareness, low educational level, and poor socio-economics of families of study population. Third; Limited local data of previous studies on similar parameter among Sudanese population.

Abbreviations:

APTT: Activated Partial Thromboplastin Time; PC: Protein C; AIDS: Acquired Immune Deficiency Syndrome; DIC: Disseminated Intravascular Coagulation. NICU: Neonatal Intensive Care Unit.

Declarations:

Ethics approval and consent to participate

Ethical clearance was obtained from the research ethical committee of Omdurman maternity hospital. Principal investigator obtained an informed consent form the neonates' mothers who included in the study before going on.

Availability of data and material

Individual data are available in digital repository of Alfajr College for Science and Technology. Khartoum, Sudan and can be obtained from the corresponding author on request. Datasets supporting the conclusions of this article are included within the article and it's available in digital repository of Alfajr College for Science and Technology (Soon will be available). [Repository.fajr.edu.sd](https://repository.fajr.edu.sd)

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Consent for publication

Not applicable

Competing interest

The authors declare that they have no competing interest

Author contributions:

¹ Corresponding author

² Supervision.

³ Clinical consultation, Management and diagnosis.

⁴ Reviewing, Analysis, Edition.

⁵ Co-supervisor.

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