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Original Article

Reference Intervals of Prothrombin Time and Activated Partial Thromboplastin Time for Third Trimester Pregnant Women

Valeurs de référence du temps de prothrombine et du temps de céphaline activée au troisième trimestre de grossesse

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Keywords: partial thromboplastin time; pregnant women; prothrombin time; reference values

ABSTRACT

Objective. Pregnancy is associated to hypercoagulable state with a decrease of activated partial thromboplastin time (aPTT) and prothrombin time (PT) during the third trimester. Reference intervals (RIs) are therefore needed for a better diagnosis of coagulation disorders during this period. We aimed to establish the RI of aPTT and PT for third trimester pregnant women **Population and Methods.** Plasma specimens were collected from adult consenting pregnant women during antenatal consultation. PT and aPTT assay were performed with a STA-R Max coagulometer (Stago, Asnières-sur-4 Seine, France). Data were analyzed according to the Clinical and Laboratory Standards Institute (CLSI) C28-A3 guideline. **Results:** A total of 120 women were recruited with a mean age of 29.9 ± 5.3 years and a mean gestational age of 37.3 ± 3.2 weeks. The RI of aPTT (sec), PT (sec), PT (%), and aPTT patient / control ratio were respectively 23.6 – 38.4, 11.1 – 14.4, 37 – 100 and 0.78 – 1.28. All these parameters didn't vary significantly across the age groups, nor the gestational age groups. **Conclusion:** This study present RIs for PT and aPTT of sub Saharan African pregnant women during the third trimester, which can be adopted by clinical laboratories in our context after appropriate validation.

RÉSUMÉ

Objectif. La grossesse est associée à une hypercoagulabilité qui se traduit par une diminution du temps de céphaline activée (TCA) et du temps de prothrombine (TP) surtout au troisième trimestre. Un meilleur diagnostic des coagulopathies pendant cette période passerait donc par l'établissement des valeurs de références. L'objectif de cette étude était de définir les valeurs de références du TCA et du TP au troisième trimestre de grossesse. **Population et Méthodes.** Nous avons inclus les femmes enceintes adultes consentantes reçues en consultation prénatale au troisième trimestre de grossesse chez qui un échantillon de plasma était prélevé. L'analyse du TCA et du TP a été réalisée grâce à un coagulomètre STA-R Max (Stago, Asnières-sur-4 Seine, France). Les données ont été analysées conformément à la directive C28-A3 du *Clinical and Laboratory Standards Institute*. **Résultats.** Au total, 120 femmes ont été recrutées avec un âge moyen de 29,9 ± 5,3 ans et un âge gestationnel moyen de 37,3 ± 3,2 semaines. Les valeurs de référence du TCA (sec), TP (sec), taux de prothrombine (%), et du rapport patient / contrôle pour le TCA étaient respectivement de 23,6 - 38,4; 11,1 - 14,4; 37 - 100 et 0,78 - 1,28. Aucun de ces paramètres ne variait significativement avec les tranches d'âge ni avec l'âge gestationnel. **Conclusion**. Cette étude présente les valeurs de référence du TCA et du TP au troisième trimestre de grossesse chez les femmes enceinte d'Afrique subsaharienne. Ces valeurs peuvent être adoptées par les laboratoires cliniques dans notre contexte après validation.

INTRODUCTION

Pregnancy is a state with numerous physiological changes including hematological modifications. Among these changes, there is an increased factor VII activity, leading to hypercoagulable state. Prothrombin time (PT) also shortens from the 20th week onwards compared to unpregnant women [1]. The level of D-dimers (the product of fibrin split) increases, suggesting the presence of fibrinolytic activity. Thus, they seems to be a balance between coagulation and fibrinolysis, explaining why thrombotic events are rare (about 1 event in 1000 pregnancies) during pregnancy [2]. Despite this low frequency, antenatal venous thromboembolism risk is 5-fold higher in pregnant women compare to nonpregnant women of the same age, and these events can occur in each of the three pregnancy trimesters [3]. Furthermore, coagulation disorders were the cause of 23% of deaths in a group of Cameroonian pregnant women admitted in

intensive care unit [4]. The third trimester present some particularities about coagulation profile: the activated partial thromboplastin time (aPTT), prothrombin time (PT), thrombin time (TT) decrease during this trimester [5]. All these variations of coagulation biomarkers suggest the need of reference intervals (RIs) for pregnant women, especially during the third trimester where high coagulation activities may prone thromboembolic diseases, and low coagulation activities may increase post-partum bleeding. We aim to establish the RIs of aPTT and PT for third trimester pregnant women. These references values will help to diagnose coagulation disorders in pregnancy.

POPULATION AND METHODS

Ethics approval

The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. Before the start of the study, an ethical clearance was obtained from the institutional ethical committee of the Yaoundé Catholic University. Furthermore, this study was conducted in accordance with the ethical principles found in the 1964 Declaration of Helsinki and its later amendments. Inform consent was obtained from each participant before inclusion and data were handled anonymously.

Study population and setting

We included a total of 120 Cameroonian adult pregnant women above 28 weeks of gestational age (GA). Patients were recruited between December 2019 and February 2020 in Essos Hospital Center in Yaoundé-Cameroon, during the third trimester antenatal consultation at the obstetrical unit. Exclusion criteria were: (i) past history of diabetes or hypertension; (ii) ongoing infection or anticoagulant therapy; (iii) personal history of abnormal bleeding or thrombotic disorders and (iv) any other known comorbidity which may affect the coagulation process. Before inclusion, an informed consent was obtained from the participants, and the study was approved by the institutional ethical committee of the Yaoundé central Africa catholic university and by the hospital's administration.

Sample collection and analysis

The plasma specimens were obtained by venipuncture from the antecubital vein and collected into a 1:9 volume of 0.109 mol/l trisodium citrate containing vacuum tubes. Samples were immediately centrifugated at 3 500 x g for 08 minutes at room temperature, and analyzed within the 06 hours of collection. PT and aPTT assay were performed with a STA-R Max coagulometer (Stago, Asnières-sur-4 Seine, France), according to the specifications and laboratory standard operating procedure recommended by the manufacturer, with the dedicated reagents.

Statistical analysis

Categorical data were described as number (percentage) while quantitative variables were described as median (interquartile range). The normality of the distribution was assessed both by graphical and statistical methods, using a histogram and the Shapiro-wilk test. Continuous data were compared across the different groups with the Kruskal Wallis test. The RIs were established with the non-parametric method based on the Clinical and Laboratory Standards Institute (CLSI) C28-A3 guideline, and the limits were calculated with 90% confidence intervals [6]. Data were entered in EXCEL (Armonk, New York, United States) and analyzed using MedCalc Statistical Software version 19.2.6 (MedCalc Software Ltd, Ostend, Belgium) [7]. P-value less than 0.05 was considered statistically significant.

RESULTS

A total of 120 reference pregnant women were included in the study, with a mean age of 29.9 ± 5.3 years (Range: 19 – 45 years). There were 19 (15.8%) women aged below 25 years, 38 (31.7%) between 25 – 29 years, 41 (34.2%) between 30 – 34 years, 18 (15%) between 35 – 39 years and 4 (3.3%) above 39 years. The mean gestational age (GA) was 37.3 ± 3.2 weeks (Range: 29 – 43 weeks), with respectively 18 (15%), 23 (19.2%) and 79 (65.8%) women below 34 weeks, between 34 – 37 weeks and above 37 weeks of GA. The results of the four parameters were not normally distributed as shown by the histograms **(figure 1)** and the statistical tests.

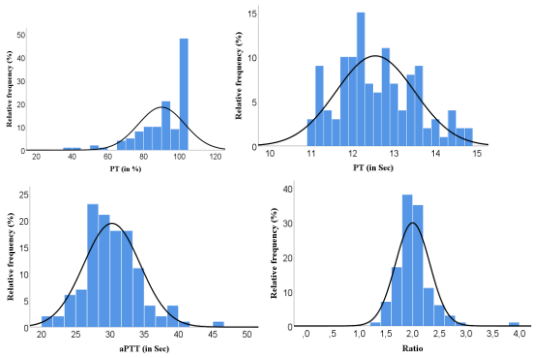

Figure 1: Histogram depicting the distribution of the different coagulation parameters.
*PT: Prothrombin time; aPTT : Activated partial thromboplastin time; Ratio: aPTT patient / control ratio.

There was not statistically significant difference of PT, aPTT and aPTT patient / control ratio between the different age (Figure 2) and gestational age groups (Table 1).

Table 1: Median values of coagulation parameters during the different week ranges of the third trimester

Variables	Gestational age (weeks)			P value
	< 34 (n = 18)	[34 – 37[(n = 23)	≥ 37 (n = 79)	
PT (Sec), *Median (IQR)*	12.6 (12.1 – 13.4)	12.4 (11.7 – 13.3)	12.3 (11.9 – 13.3)	0.687
PT (%), *Median (IQR)*	90.5 (76.9 – 100)	96 (83.2 – 100)	93.3 (87 – 100)	0.873
aPTT (Sec), *Median (IQR)*	28.9 (27.3 – 31.1)	31.8 (28.2– 33.8)	30 (27.3 – 32.1)	0.190
Ratio, *Median (IQR)*	0.96 (0.89 – 105)	1.02 (0.91 – 1.15)	0.99 (0.91 – 10.5)	0.429

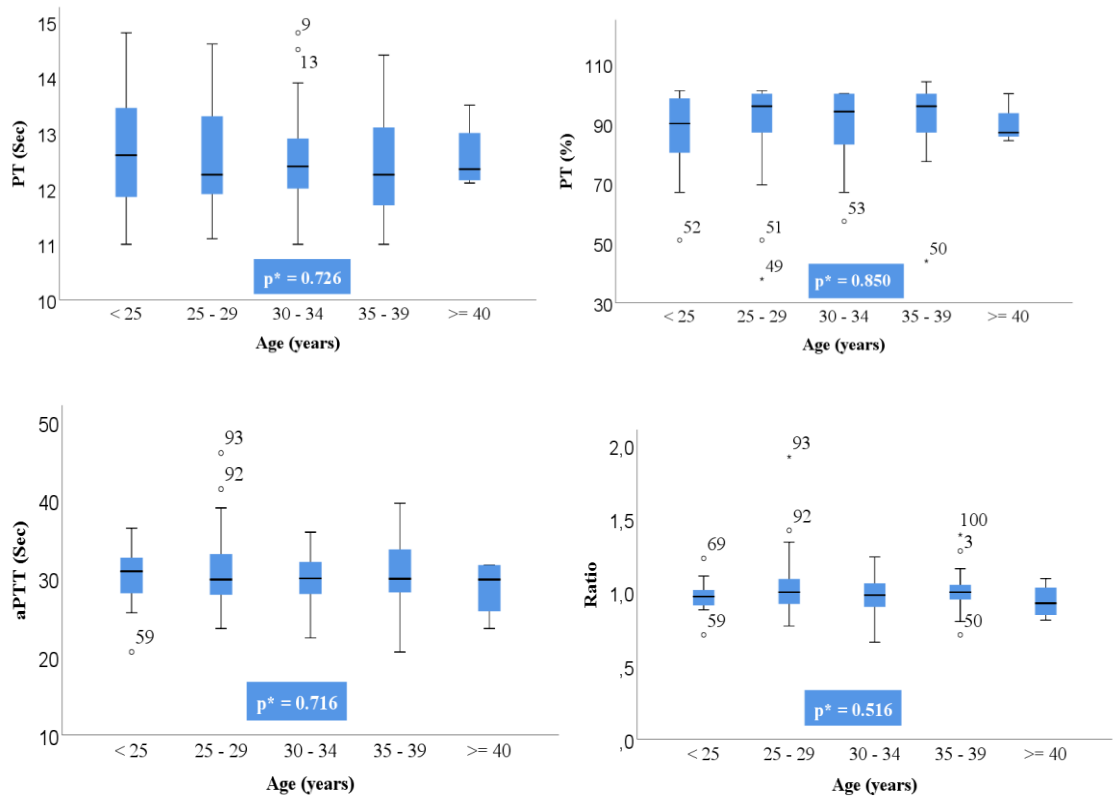

Figure 2: Box plots describing the distribution of coagulation parameters according to age groups.

**p: p values compare the median values of corresponding parameter between the different age groups with the Kruskal Wallis test.*
PT: Prothrombin time; aPTT : Activated partial thromboplastin time; Ratio: aPTT patient / control ratio.

The RIs for PT in seconds, PT in percentage, aPTT in seconds and aPTT patient / control ratio were respectively (11.1 – 14.4), (67 – 100), (23.6 – 38.4), and (0.78 – 1.28) (Table 2).

Table 2: Reference intervals for PT, aPTT and Ratio in the study population

Variables	Lower limit	90% CI	Upper Limit	90% CI
PT, Sec	11.1	11 – 11.2	14.4	13.9 – 14.8
PT, %	67	65.3 - 72.1	100	100 - 101
aPTT, Sec	23.6	20.6 – 25.1	38.4	35.9 – 41.4
Ratio	0.78	0.71 - 0.81	1.28	1.2 – 1.42

DISCUSSION

Reference intervals are useful for clinical decision, including diagnosis or patient's management. Routine coagulation parameters such as PT and aPTT have been shown to change during pregnancy, due to physiological need of pregnant women and fetuses. RIs of these parameters have been widely studied in Caucasian pregnant women, but few have been done in sub Saharan Africa women. This study provides RIs for PT and aPTT in third trimester pregnant Cameroonian women. We included 120 adult participants without hemostasis disorders symptoms, nor any other known comorbidity.

We found that PT and aPTT was not different across the gestational age groups. A similar result was found by Cui et al. in Chinese cohort of pregnant women, were aPTT and PT didn't vary significantly between the second and the third trimester of pregnancy [8]. Therefore, they may not be necessary to have specific RIs for the different GA groups in third trimester pregnant women. PT RI in our population (11.1 – 14.4) was higher than the RI found by Cui et al. (8.6 – 12.4) during the third trimester of pregnancy, while the mean age was similar in both populations. This difference suggests that coagulation parameters vary by race and or ethnicity, as it have been shown earlier[9]. Thus, there is a need of RIs for each specific population. To the best of our knowledge, our study is the first to report RIs of PT and aPTT in a group of third trimester African pregnant women.

This study has some limitations including the hospital-based sample which may not represent the whole population, considering that in our context many pregnant women don't attend antenatal consultation. Furthermore, the healthy state of the participants was based on participant past history of known comorbidities. Therefore, they may have some undiagnosed diseases.

CONCLUSION

PT and aPTT don't vary across the different age and gestational age groups in third trimester pregnant women. We have presented RIs of these parameters in a sub-Sahara African population, which can be adopted by clinical laboratories in our context, after appropriate validation. Further studies are needed for a better description of coagulation parameters RIs in the different ethnicities.

Authorship contributions

(I) Conception and design: P. Angandji; J. Ambomo; M.P.N. Balogog; C. Tayou; (II) Administrative support: M.P.N. Balogog; C. Tayou; (III) Provision of study materials and patients: M.P.N. Balogog; (IV) Collection and assembly of data: J. Ambomo; M.P.N. Balogog; (V) Data analysis and interpretation: P. Angandji; J. Ambomo; M.P.N. Balogog; G.S. Wafeu; C. Tayou; (VI) Manuscript writing: All authors; (VII) Final approval of manuscript: All authors

Conflicts of interest

The authors have no conflicts of interest to declare

REFERENCES

1. Cerneca F, Ricci G, Simeone R, Malisano M, Alberico S, Guaschino S. Coagulation and fibrinolysis changes in normal pregnancy. Increased levels of procoagulants and reduced levels of inhibitors during pregnancy induce a hypercoagulable state, combined with a reactive fibrinolysis. Eur J Obstet Gynecol Reprod Biol. 1997 May;73(1):31–6.
2. Bremme K, Ostlund E, Almqvist I, Heinonen K, Blombäck M. Enhanced thrombin generation and fibrinolytic activity in normal pregnancy and the puerperium. Obstet Gynecol. 1992 Jul;80(1):132–7.
3. Andersen BS, Steffensen FH, Sorensen HT, Nielsen GL, Olsen J. The cumulative incidence of venous thromboembolism during pregnancy and puerperium. Acta Obstet Gynecol Scand. Wiley-Blackwell; 1998 Feb;77(2):170–3.
4. Owono EP, Metogo MAJ, Tchokam L, Danwang C, Kago TL, Afane EA, et al. Complications obstétricales admises en réanimation : épidémiologie, diagnostic et pronostic. Health Sci Dis. 2017;18(1):48–52.
5. Han L, Liu X, Li H, Zou J, Yang Z, Han J. Blood Coagulation Parameters and Platelet Indices : Changes in Normal and Preeclamptic Pregnancies and Predictive Values for Preeclampsia. 2014;(Dd):1–14.
6. Gary L. Horowitz, Sousan Altaie JCB et al. EP28-A3c: Defining, Establishing, and Verifying Reference Intervals in the Clinical Laboratory; Approved Guideline—Third Edition. Clin Lab Stand Inst. 2010;28(October):12.
7. MedCalc software download page. [Internet]. [cited 2020 Jul 8]. Available from: https://www.medcalc.org/download.php
8. Cui C, Yang S, Zhang J, Wang G, Huang S, Li A, et al. Trimester-specific coagulation and anticoagulation reference intervals for healthy pregnancy. Thromb Res. Elsevier Ltd; 2017;156:82–6.
9. Lutsey PL, Cushman M, Steffen LM, Green D, Barr RG, Herrington D, et al. Plasma hemostatic factors and endothelial markers in four racial/ethnic groups: the MESA study. J Thromb Haemost. 2006 Dec;4(12):2629–35.