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# Acceptable concordance between Doppler indices throughout free-loop and placenta sites of umbilical cord

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## ABSTRACT

**Introduction:** There is a critical need to determine a standard site sampling along the umbilical cord that has acceptable reliability and validity to measure the umbilical artery indexes. This study aimed to assess the correlation of the placental insertion site of the cord with the free loop in terms of umbilical artery indices including pulsatility index (PI), resistance index (RI), and systolic/diastolic ratio (S/D).

**Methods:** This cross-sectional study was conducted on 150 singleton fetuses of 28 to 40 weeks of gestational age in the prenatal clinics of Imam Khomeini hospital from January 2019 to January 2021.

**Results:** Analyses of data have shown that there were not any significant correlations between the means of Doppler indices of UA with sex of fetus (P>0.05) or the number of mother's gravid (P>0.05).

There was a positive significant association between PI, RI, and S/D of the umbilical artery in the free loop of umbilical cord and placenta site after adjusting for gestational age group, fetus weight, and placenta position.

**Conclusion:** It seems the placenta site Doppler, considering its acceptable correlation with the free loop and its reliability, can also be a good choice as a standard sampling site for fetuses' circulation assessment especially in multiple pregnancies and fetuses with oligohydramnios.

**Keywords:** Resistance index, Pulsatility index, Systolic/diastolic ratio, Free-loop Doppler, Placenta insertion Doppler, Umbilical cord

## INTRODUCTION

Umbilical arterial (UA) Doppler is an ultrasound imaging test for assessing the fetal well- being during high-risk pregnancies in the second and third trimesters. This non-invasive diagnostic means is commonly utilized to examine the speed and direction of blood flow in the umbilical cord (UC) in order to find any abnormal velocity and consequently reduce perinatal mortality and morbidity (1-3.16). The UC carries nutrients and oxygen into the fetus then it is very important to evaluate and measure it at different sites but determining the best site for Doppler sampling along fetus cord is very challenging, especially in multiple pregnancies, in a fetus with a single umbilical artery or long cord (greater than mean range: 350-550 mm at 24 and 40 weeks) (4-9).

Free loop (FL), placental end of umbilical cord, perivesical arteries and abdominal insertion are the most usual sampling sites for calculation of Pulsatility (PI), resistance (RI) and Systolic/Diastolic ratio (S/D) indices. Among these four sites, Free loop as a standard site is easy for measurement but it has the least reproducible variation of parameters compared to other insonation sites. In cases where repeated fetal evaluation is required, a fixed site like placental insertion (PI) is also reliable and even better because of its anatomical landmark and reproductivity (10).

Doppler velocimetry parameters established at a different site of the umbilical artery show significant differences; for instance, there is a significant impedance declining from abdominal to placental end and it is usual for two operators to obtain completely different waveforms from the same subject during the same examination (11). Therefore, it seems there is a critical need to determine a standard site sampling along the umbilical cord that has acceptable reliability and validity to measure the umbilical artery indices. Considering all above-mentioned limitations, this study aims to examine both FL and PI sites in normal fetus to conduct a correlational study of statistical relationship among different selected indices: Pulsatility, resistance and systolic/diastolic ratio.

## MATERIALS AND METHODS

This cross-sectional research was conducted by a fellowship student of Perinatology and her colleagues in obstetric sonography department of Imam Khomeni teaching hospital in Tehran- Iran from May 2019 to January 2021. Ethics approval was obtained from the institutional review board of Tehran University of Medical Sciences according to Helsinki declaration and Registration number (IR.TUMS.MEDICINE.REC TUMS.IKHC.REC.1399.373). All gathered data were considered confidential and no extra cost was imposed on subjects. The researchers paid enough attention to ethical considerations of this approval. An appropriate consent paper was prepared and all potential subjects were informed about rights, minimal risks and benefits of this trial. The main researcher has explained the procedure of trial and make sure that the subject has understood the information clearly and given her voluntary approval for participating in this trial.

Doppler waveforms were performed by a single operator (AK) to minimize interobserver variability by using an Advanced Technology Laboratory (ATL) and Philips Affinity 70 ultrasound machine. The power set at B / 92 MW / $cm^2$ spatial maximum temporal average severity. (7.5 MHz) was set for wall filter and the frequency of the pulse repetition was 5 MHz and the persistence was at middle.

Every subject was examined by Philips Affiniti 70 Ultrasound Machine (with a transabdominal convex probe 3.5-5.5 MHz) and following parameters such as Pustitlity index (PI), resistance index (RI) and Sytolic/diastolic ratio (S/D) were measured in UA doppler assessment. The implemented angle of insonation was less than $30^o$, the number of waves was 6-10 and the determination of indices was performed through fetal apnea. Obtained measurements were all recorded in a data collection form consisting of other demographic information like maternal age, fetal sex, BMI and placenta location. The population of 150 pregnant women with normal singleton fetus were invited to this study. The potential pregnant women had some other criteria including gestational age between 28 and 40 weeks and fetal weight between the $10^{th}$ and $90^{th}$ percentiles. Pregnant women with fetal anomaly, macrosomia, intra uterine growth restriction (IUGR), multiple pregnancy, complicated pregnancy and abnormality in UC structure were excluded from this research.

With respect to gestational age, participants were assigned in 4 groups <30, 30-34, 34-37 and > 37 weeks. Detailed ultrasound findings associated with umbilical arterial indices in two particular places of FL and PI were recorded. Finally, the correlation coefficients between the variables taken from these two sites were measured as the primary outcome.

***Sample size:*** Based on former investigations by Bhidea in 2019 (12) and using formula, the proposed sample size was calculated 147 subjects. With the proposed sample size, the study had a power of 95% and an alpha error of 0.05.

$$n = \left[ \frac{Z_\alpha + Z_\beta}{c} \right]^2$$

$$c = 0.5 * Ln\left[(1+r)/(1-r)\right]$$

R=0.264
N=147

**Data Analysis:** Detailed ultrasound findings associated with umbilical arterial indices were recorded in checklists. Analyses were performed by using software package SPSS version 24.0 (IBM, New York, USA). A P-value of less than 0.05 was determined as the level of statistical significance. We used Paired samples T-test and Non-parametric Wilcoxon signed ranks tests to assess differences in means. A Pearson correlation test was applied to evaluate concordance between umbilical parameters. Quantitative and qualitative parameters were reported by mean+SD and number (percent), respectively. Using Kolmogorov-Smirnov Test showed a normal distribution of quantitative data. Associations between variables were assessed by ANOVAs (General Linear Model, Post-Hoc-Bonferonni), and Chi square tests.

## RESULTS

One-hundred and fifty eligible fetuses with gestational age 28-40 weeks entered the study. The means of mothers' age, gestational age, and BMI were 30.75±6.09 years, 34.03±3.41 weeks, and 25.4±4.68, respectively. The mean of estimated fetal weight at the time of UA Doppler assessment was 2630±2473 gr. Detailed data related to participants' obstetrical and demographic characteristics as well as Doppler indices at FL and placenta insertion sites of umbilical artery are shown in Table 1.

Table1: Participants' demographic characteristic and the mean of umbilical artery Doppler indices at different sites

| variables | N=150 |
|---|---|
| Gender (n %) | |
| Male | 80 (53.3) |
| Female | 70 (46.7) |
| Placental site (n %) | |
| Anterior | 77 (51.3) |
| Posterior | 51 (34) |
| Fundal | 17 (11.3) |
| Lateral | 5 (3.3) |
| Gestational age (weeks; n %) | |
| <30 | 52 (34.7) |
| 30-34 | 23 (15.3) |
| 34-37 | 19 (12.7) |
| >37 | 56 (37.3) |
| Gravid (n %) | |
| Primi-para | 62 (41.3) |
| Multi-para | 88 (58.7) |
| PI (Mean±SD) | |
| Free loop | 0.94±0.17 |
| Placental insertion | 0.81±0.15 |
| RI (Mean±SD) | |
| Free loop | 0.60±0.07 |
| Placental insertion | 0.54±0.06 |
| S/D (Mean±SD) | |
| Free loop | 2.57±0.46 |
| Placental insertion | 2.23±0.37 |

PI: pulsatility index; RI: resistance index; S/D: systolic/diastolic ratio

Analyses of data have shown that there were not any significant correlations between the means of Doppler indices of UA with sex of fetus (P>0.05) or the number of mother's gravid (P>0.05). Increase of gestational age could significantly reduce the means of RI (p=0.001), PI (p=0.001), and S/D (p=0.001) indices at FL and placenta insertion sites of umbilical artery. Detailed data related to effects of gestational age on UA Doppler indices in these sites are demonstrated in Table 2.

Table 2: Comparison the mean of umbilical artery Doppler indices in different gestational age

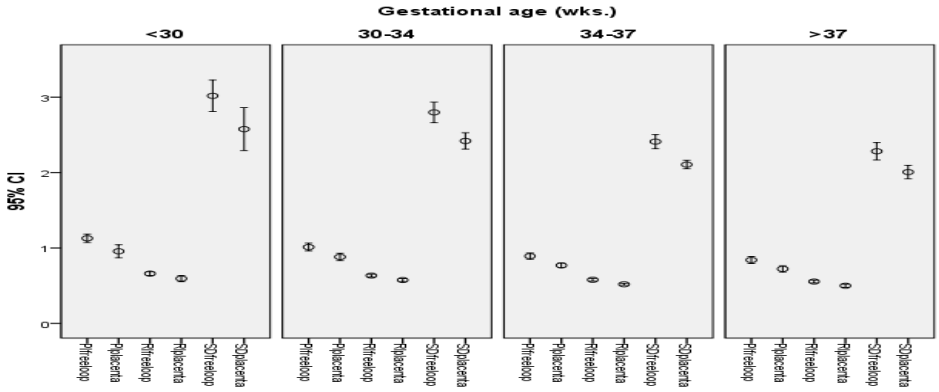
| Indices / Gestational age | <30 Weeks (Mean±SD) | 30-34 Weeks (Mean±SD) | 34-37 Weeks (Mean±SD) | >37 Weeks (Mean±SD) | P value |
|---|---|---|---|---|---|
| **PI** | | | | | |
| Free loop | 1.06±0.17 | 0.95±0.13 | 0.89±0.15 | 0.85±0.14 | 0.0001 |
| Placental | 0.92±0.17 | 0.83±0.12 | 0.76±0.11 | 0.73±0.10 | 0.0001 |
| **RI** | | | | | |
| Free loop | 0.64±0.05 | 0.61±0.06 | 0.57±0.06 | 0.56±0.05 | 0.0001 |
| Placental | 0.58±0.07 | 0.54±0.05 | 0.51±0.04 | 0.50±0.05 | 0.0001 |
| **S/D** | | | | | |
| Free loop | 2.89±0.46 | 2.63±0.37 | 2.36±0.34 | 2.32±0.32 | 0.0001 |
| Placental | 2.50±0.43 | 2.25±0.26 | 2.08±0.20 | 2.03±0.22 | 0.0001 |

PI: pulsatility index; RI: resistance index; S/D: systolic/diastolic ratio

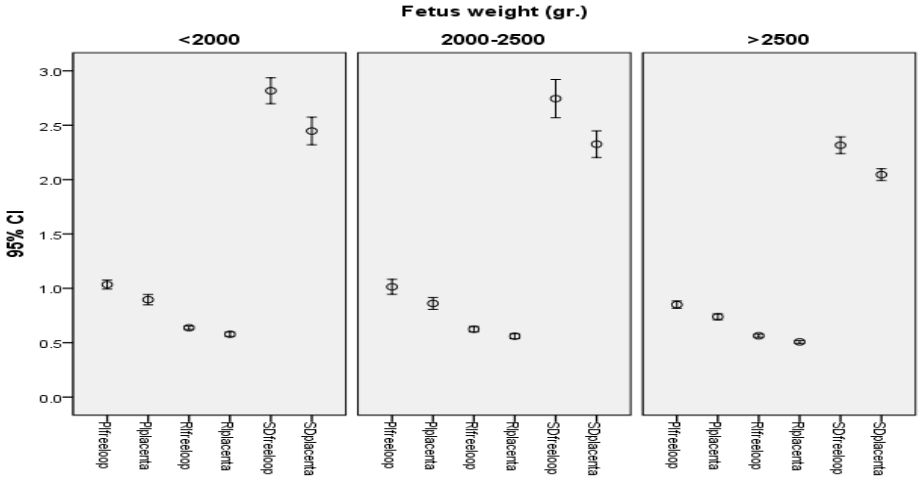
The average of PI, RI, and S/D were significantly lower in the placenta site compare to the free loop of the umbilical cord. These significant differences were also detected in different gestational age categories (group 1 (lower than 30 weeks), group 2 (30-34 weeks), group 3 (34-37 weeks), group 4 (greater than 37 weeks)), and different fetus weight classifications (<2000 grams, 2000-2500 grams, more than 2500 grams). (Figure 1 A-B)

Figure 1. The mean (S.D) PI, RI, and S/D measurement comparison in the free loop and the placenta site according gestational age (A), fetus weight (B).

A.




B.



As detailed data are shown in Table 3, there were also significant differences in UA Doppler indices between groups with different gestational ages. PI indices in free loop and placental insertion sites among subjects with gestational age <30 weeks were significantly higher than these indices in those with gestational age>30 weeks (p<0.05). There were significant differences between groups with gestational age <30 weeks and gestational age>34 weeks regarding to UA RI indices in all free loop and placental sites (p<0.05). S/D indices of free loop, placental insertion sites were significantly higher in the group with gestational age <30 compared to those with gestational age>30 weeks (p<0.05).

Table 3: The mean differences scores for UA indices in each gestational age (Post Hoc Analysis)

| Indices | UA Sites | Gestational age group | | Mean Difference (I-J) | Std. Error | 95% Confidence Interval | 95% Confidence Interval | |
|---|---|---|---|---|---|---|---|---|
| | | | | | | | Lower Bound | Lower Bound |
| **PI** | Free loop | <30w | 30-34 | 0.106 | 0.038 | 0.041 | 0.002 | 0.209 |
| | | | 34-37 | 0.171 | 0.041 | 0.0001 | 0.061 | 0.282 |
| | | | >37 | 0.214 | 0.029 | 0.0001 | 0.135 | 0.294 |
| | Placental | <30w | 30-34 | 0.094 | 0.033 | 0.037 | 0.003 | 0.184 |
| | | | 34-37 | .157 | 0.036 | 0.0001 | 0.060 | 0.254 |
| | | | >37 | .193 | 0.026 | 0.0001 | 0.124 | 0.263 |
| | | 30-34 | >37 | 0.099 | 0.033 | 0.020 | 0.010 | 0.189 |
| **RI** | Free loop | <30w | 34-37 | 0.0736 | 0.016 | 0.0001 | 0.030 | .116 |
| | | | >37 | 0.0832 | 0.011 | 0.0001 | 0.052 | 0.114 |
| | | 30-34 | >37 | 0.0472 | 0.014 | 0.011 | 0.007 | 0.087 |
| | Placental | <30w | 34-37 | 0.0734 | 0.016 | 0.0001 | 0.030 | 0.116 |
| | | | >37 | 0.0837 | 0.011 | 0.0001 | 0.052 | 0.114 |
| | | 30-34 | >37 | 0.0437 | 0.014 | 0.023 | 0.004 | 0.083 |
| **S/D** | Free loop | <30w | 30-34 | 0.261 | 0.097 | 0.049 | 0.000 | 0.522 |
| | | | 34-37 | 0.527 | 0.104 | 0.0001 | 0.248 | 0.806 |
| | | | >37 | 0.571 | 0.074 | 0.0001 | 0.370 | 0.771 |
| | | 30-34 | >37 | 0.310 | 0.096 | 0.010 | 0.052 | 0.567 |
| | Placental | <30w | 30-34 | 0.255 | 0.079 | 0.010 | 0.042 | 0.4689 |
| | | | 34-37 | 0.426 | 0.085 | 0.0001 | 0.198 | 0.6540 |
| | | | >37 | 0.4724 | 0.061 | 0.0001 | 0.308 | 0.6361 |
| | | 30-34 | >37 | 0.216 | 0.078 | 0.040 | 0.005 | 0.427 |

PI: pulsatility index; RI: resistance index; S/D: systolic/diastolic ratio; PV: Perivesical

Table 4: Associations between UA indices and locations of placenta

| Placental location | Free loop | Placental insertion |
|---|---|---|
| **PI** | | |
| Anterior | 0.93±0.15 | 0.80±0.14 |
| Posterior | .98±0.19 | .84±0.17 |
| Foundal | .91±0.21 | .77±0.14 |
| Lateral | .95±0.21 | .82±0.17 |
| P value | 0.385 | 0.299 |
| **RI** | | |
| Anterior | 0.59±0.06 | 0.5383±0.06 |
| Posterior | .61±0.06 | .5524±0.07 |
| Foundal | .58±0.09 | .5253±0.07 |
| Lateral | .60±0.09 | .5500±0.08 |
| P value | 0.402 | 0.504 |
| **S/D** | | |
| Anterior | 2.54±0.40 | 2.21±0.32 |
| Posterior | 2.64±0.50 | 2.30±0.45 |
| Fundal | 2.50±0.53 | 2.15±0.35 |
| Lateral | 2.58±0.60 | 2.26±0.41 |
| P value | 0.644 | 0.418 |

PI: pulsatility index; RI: resistance index; S/D: systolic/diastolic ratio

There were no significant associations between UA indices and locations of placenta (p>0.05); PI, RI and S/D indices in different sites of umbilical artery were not significantly different between subjects with anterior, posterior, fundal or lateral placenta (Table 4).

Determining correlation between indices, further analyses by Pearson Correlation test showed that there were significant positive associations between PI with RI (p= 0.0001; Pearson Correlation=0.921) and S/D (p= 0.0001; Pearson Correlation=0.946) indices of free loop. Moreover, significant positive associations were observed between PI of free loop with PI of umbilical artery in placental insertion site (p= 0.0001; Pearson Correlation=0.813). This significant positive relationship was also found between free loop and placental site regarding to RI (p=0.0001; Pearson Correlation=0.744) or to S/D (p=0.0001; Pearson Correlation =0.709) indices. These positive significant associations existed after adjusting for gestational age group, fetus weight, fetus gender and placenta location (table5)

Table 5. Correlation of umbilical artery parameter measurements at the two sites.

| **Variables** | **Pulsatility index** | | **Resistance index** | | **Systolic/diastolic ratio** | |
|---|---|---|---|---|---|---|
| | **Pearson Correlation** | **P-value** | **Pearson Correlation** | **P-value** | **Pearson Correlation** | **P-value** |
| **Overall** | 0.813 | <0.001 | 0.744 | <0.001 | 0.709 | <0.001 |
| **Gestational age (wks.)** | | | | | | |
| <30 (N=17) | 0.717 | 0.001 | 0.287 | 0.265 | 0.257 | 0.319 |
| 30-34 (N=47) | 0.788 | <0.001 | 0.760 | <0.001 | 0.684 | <0.001 |
| 34-37 (N=57) | 0.758 | <0.001 | 0.730 | <0.001 | 0.708 | <0.001 |
| >37 (N=29) | 0.770 | <0.001 | 0.664 | <0.001 | 0.761 | <0.001 |
| **Fetus weight (gr.)** | | | | | | |
| <2000 (N=47) | 0.770 | <0.001 | 0.538 | <0.001 | 0.460 | 0.001 |
| 2000-2500 (N=36) | 0.772 | <0.001 | 0.798 | <0.001 | 0.780 | <0.001 |
| >2500 (N=67) | 0.778 | <0.001 | 0.706 | <0.001 | 0.729 | <0.001 |
| **Placenta position** | | | | | | |
| Anterior (N=77) | 0.737 | <0.001 | 0.695 | <0.001 | 0.623 | <0.001 |
| Posterior (N=51) | 0.875 | <0.001 | 0.742 | <0.001 | 0.729 | <0.001 |
| Fundal (N=17) | 0.831 | <0.001 | 0.838 | <0.001 | 0.870 | <0.001 |
| Lateral (N=5) | 0.931 | 0.021 | 0.928 | 0.023 | 0.855 | 0.065 |

## DISCUSSION

Umbilical artery Doppler is consumed broadly in prenatal surveillance monitoring of fetal well-being, but the determination of standard sampling sites on umbilical cord on human fetus has still remained challenging (3). Some studies demonstrated this fact that the free loop of the umbilical cord as a standard sampling site is easily identifiable and has a better sensitivity and moderate reliability compared to placenta site; although, it has the least reproductible result because of the influence of the effect of the site of insonation. Contrary to these findings, there were also some researchers who advised placenta end as a reliable insonation site in particular in multiple pregnancy, and /or when comparing repeated measurements longitudinally because it had the most reproducibility of waveform patterns and indices (3, 10, 13.17.18).

This study showed strong significant (p-value<0.001) correlations between the free loop of the umbilical cord and the placenta site. There was, in fact, a positive significant association between PI, RI, and S/D of the umbilical artery in free loop of umbilical cord and placenta site after adjusting for gestational age group, fetus weight, fetal sex and placenta site. The researcher found that the PI of umbilical cord at the fetal end was higher and it was gradually falling down to the placental end. The fall demonstrated a decreasing gradient across the length of cord as previously reported by other authors (10).

This study also showed that the umbilical artery parameters from free loop to placenta end with developing pregnancy decreases considerably. For instance, The S/D ratio diminishes with advancing gestational age while diastolic flow increases: At <30 weeks, the 50$^{th}$ percentile for the S/D ratio in free loop and placenta end are 2.89 and 2.5 respectively; these changes for the pregnancy of 30 -34

weeks are 2.63 and 2.25; for the pregnancy of 34-37 weeks , these parameters are 2.36 and 2.08 ; and at >37 weeks, the 50th percentile of free loop and placenta end are 2.32 and 2.03 in the same order.

Sex related differences in Umbilical cord Doppler has been examined in some cross-sectional studies. A few studies found some sex differences in Doppler indices like pustility index. In the case of Christian Widnes et al., they monitored 294 pregnancies (152 female and 142 male) and found that sex-specific reference ranged for UA Doppler indices and female fetus had 2-8% higher values for UA Doppler indices than male fetuses (14) . Although, in our study there was not any significant correlation between umbilical cord Doppler indices and fetus sex.

In this research there was also no significant difference in umbilical cord doppler parameters between primigravid and multigravid women. Ayoola and et al. examined a total of 400 pregnant women (82 primigravid subjects and 312 mulitigravid subjects with 2 to 5 children) that their ages ranged from 18 to 44 years and they similarly found no statistically significant differences across gravidity groups in mean Doppler parameters for both primigravid and multigravid subjects (15).

There are two major limitations in the study that could be addressed in future research. First the study focused on only two sites of free-loop and placenta sites; and the researcher did not measure PI, RI and S/D ratio indices at other remaining sites (previsical and abdominal insertion sites) of umbilical cord. Second, the study did not include any follow-up visit to evaluate any probable pregnancy complications. The future longitudinal research is recommended to include documentation of the final outcome of examined pregnancies and study umbilical indices in other remaining sites.

## CONCLUSION

Measurement of PI , RI and S/D ratio indices in umbilical artery Doppler of fetus is considered to be very important for monitoring high-risk pregnancies. Because of wide variations of Doppler indices, finding a feasible and standard site for obtaining the best ultrasound waveforms during the examination have always been a challenging problem in clinical assessments. In this study, the researcher analyzes the variability in both free-loop and placenta sites respectively: PI of free-loop has 80% correlation to PI of placenta site; RI and S/D ratio of free-loop have nearly 70% correlation to RI and S/D ratio of placenta site. Considering such acceptable correlation between measured parameters in both sites, the placental site Doppler can also be a good choice as a standard sampling site for fetuses' circulation assessment especially in multiple pregnancies and fetuses with oligohydramnios

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