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Effect of antenatal maternal anxiety on the reactivity of fetal cerebral circulation to auditory stimulation, and early child development

Ефекат пренаталне анксиозности труднице на реактибилност феталне церебралне циркулације након аудитивне стимулације и рани развој детета

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Ефекат пренаталне анксиозности труднице на реактибилност феталне церебралне циркулације након аудитивне стимулације и рани развој детета

SUMMARY

Introduction/Objective Antenatal maternal anxiety (AMA) can have negative impact on child's pre- and postnatal development.

The aim was to examine the effect of AMA on changes in fetal blood flow throw middle cerebral artery (MCA) after acoustic stimulation, birth outcome, and early child development.

Methods Forty-three women in the third trimester of pregnancy and their children were enrolled in a longitudinal prospective study. The AMA (state and trait) was assessed using Spielberger's questionnaire. The MCA blood flow was assessed using Doppler ultrasound before and after defined acoustic stimulus. The data regarding neonatal body weight, Apgar score, first step and word appearance, estimated speech-language (ESLD), sensory-motor (ESMD), and social-emotional (ESED) development at the age of three were collected.

Results Fetuses from mothers with high state and trait anxiety had slower reaction time and higher relative pulsatility index after defined auditory stimulation. There are more preterm deliveries, lower scores on ESLD, ESMD, and ESED in mothers with high STAI-T compared to low. There is a negative correlation between reaction time, relative pulsatility index, STAI-T and ESLD.

Conclusion High levels of antenatal maternal trait anxiety could have a negative effect on both, fetal reactivity to sound stimulation and early child development.

Keywords: antenatal maternal anxiety; fetus; cerebral circulation; auditory stimulation; postnatal development

САЖЕТАК

Увод/Циљ Пренатална анксиозност мајке (ПАМ) може имати негативан утицај на пре- и постнатални развој детета.

Циљ овог истраживања био је да се испита ефекат ПАМ-а на промену протока крви кроз медијалну церебралну артерију (МЦА) фетуса након аудитивне стимулације, исход порођаја и рани развој детета.

Методе Четрдесет три труднице у трећем триместру и њихова деца су учествовали у лонгитудиналној проспективној студији. ПАМ је испитивана Спилбергеровим упитником. Проток крви кроз МЦА је испитан Доплер ултразвуком пре и после звучне стимулације. Прикупљени су подаци о телесној тежини на рођењу, Апгар скору, појави првих корака и речи, процењеном говорно-језичком, сензомоторном и социоемоционалном развоју на узрасту од 3 године.

Резултати Фетуси трудница са високим степеном анксиозности (тренутне и оне која је црта личности) су имали спорију реакцију и веће вредности пулсатилног индекса након аудитивне стимулације. Било је више претерминских порођаја, нижих постигнућа у процењеном говорно-језичком, сензо-моторном и социо-емоционалном развоју деце код мајки са високим степеном анксиозности као црте личности. Утврђена је негативна корелација између времена реакције, релативног пулсатилног индекса, анксиозности као црте личности и процењеног говорно-језичког развоја.

Закључак Висок нивоа пренаталне анксиозности труднице може имати негативан ефекат на феталну реактибилност на аудитивну стимулацију и рани развој детета.

Кључне речи: пренатална анксиозност мајке; фетус; церебрална циркулација; аудитивна стимулација; постнатални развој

INTRODUCTION

There is a strong body of evidence that antenatal maternal stress and anxiety can have a negative impact on child's pre- and postnatal development. Stress is caused by an existing, real factor or stressor. Anxiety is stress that continues to affect a person after stressor is gone. There are two ways how antenatal maternal anxiety can manifest itself: as state or trait. The major difference between state and trait anxiety is the duration of the condition. State anxiety

is short-term while trait anxiety is long-term, more stable tendency to experience negative emotions across many situations. Hence, trait anxiety is treated as a personality characteristic. That may be the reason why trait anxiety has a stronger impact on fetal development compared to state anxiety. Maternal stress and anxiety can lead to preterm birth and lower birth weight [1]. Moderate to high levels of antenatal maternal stress might have a negative impact on intellectual and language development in the early postnatal period [2]. In addition, studies are reporting an association between antenatal maternal anxiety and postnatal development of hyperactivity in children [3]. However, underlying mechanisms of antenatal maternal anxiety impact on fetal and early child development are not fully explored. It is known that placenta controls fetal exposure to the maternal environment, so its role could be of great importance. Cortisol appears to cross the placenta and thus may affect the fetus and disturb ongoing developmental processes. There are studies showing that high levels of anxiety could alter fetal umbilical and cerebral blood flow [4, 5]. Antenatal maternal anxiety may have a negative effect on the distribution of a blood flow to the fetus. This might disturb the transportation of the important nutrients to the fetal developing organs. There is a possibility to examine fetal reactivity to external defined auditory stimulus by detecting changes in fetal middle cerebral artery (MCA) blood flow change. The fetal auditory system is fully functional by the 25th week of gestation and is considered to be the major fetal information funnel. Changes in fetal cerebral circulation due to auditory stimulation are the basis of the procedure known as prenatal auditory screening (PAS) [6]. It was demonstrated previously in the group of gravid women that gestational hypertension was associated with significantly altered pulsatility index (Pi) values [7, 8, 9]. It was concluded that PAS measurements of Pi indices may be used as an indicator of psychophysiological development in the early postnatal period, while the absence of Pi changes might indicate a slow-down in fetal neurodevelopment, and particularly the maturation of the fetal auditory system.

Knowing that more expressed MCA blood flow in PAS procedure is associated with high risk pregnancies, the aim of this preliminary study was to examine if fetal MCA blood flow changes after auditory stimulation are in correlation with antenatal maternal anxiety. We also wanted to explore the potential effect of antenatal maternal anxiety on birth outcome and early speech-language, sensory-motor, and social-emotional development. To the best of our knowledge, this is the first study that conducted a longitudinal follow-up design to examine an association between fetal reactivity to defined sound stimulation and early speech-language development due to antenatal maternal anxiety.

METHODS

Participants

Participants were recruited from the larger sample used for the longitudinal investigation of the effect of different antenatal factors on the early child development. From that sample, the sample of 72 pregnant women was selected. The general anamnestic data were collected from the participants' medical records. Participants with chronic hypertension, preeclampsia, eclampsia, diabetes, hypo- or hyperthyroidism or any other medical condition were not included in the study. Subjects used no alcoholic beverages, tobacco or any other type of psychoactive substances. None of the participants were taking any hormones during pregnancy nor any medication. None of the participants had diagnosed an anxiety disorder or any psychiatric condition. From the initial sample of 72 women, the study finally included 43 gravid women with singleton pregnancies, in gestational age between 28 and 41 weeks of the pregnancy and their babies. Table 1 presents the sample characteristics.

Procedure

The study comprised four steps. Complete study protocol has been approved by local Ethical Boards, which operate in accordance with the Ethical principles for medical research involving human subjects, established by Declaration of Helsinki 2013. Participants had a detailed written explanation of each of the four steps. All the participants signed their written consent prior the study. The study was conducted in the Narodni Front Clinic for Gynecology and Obstetrics, Belgrade, Serbia, and the Institute for Experimental Phonetics and Speech Pathology, Belgrade, Serbia.

Assessment of the antenatal maternal anxiety

The first step was aimed at evaluating anxiety level in healthy pregnant women in the third trimester using Spielberger's questionnaire form Y [10]. A total of 73 women participated in this step. The questionnaire is designed to assess anxiety as both emotional

state (STAI-S) and personality trait (STAI-T). Before the study, a cutoff point of scores > 40 were selected for both state anxiety and trait anxiety [11]. The internal consistency for the state (Cronbach's $\alpha = 0.81$) and trait anxiety (Cronbach's $\alpha = 0.89$) in the sample were high. The questionnaire was given to pregnant women before the procedure of testing fetal response to defined auditory stimulation.

Testing fetal response to defined auditory stimulation

The second step was to test fetal reactivity to sound stimulation using color Doppler sonography. The first and second steps were conducted on the same day. PAS was performed following the Protocol of Prenatal auditory screening as previously described in [7, 8, 9].

Collecting data regarding the neonatal status

The third step was collecting the data regarding neonatal status of the new-born babies (a total of 47 babies participated in this step). The data about gestational age at birth (GAB: 1—term: >37th week of gestation, and 2—preterm: < 37th week of gestation), body weight (BW) and Apgar score (AS) in the fifth minute were collected.

Assessment of the early child development at the age of three

The final fourth step was testing speech-language, sensory-motor, and social-emotional development of children at the age of three and collecting the data regarding early motor (appearance of the first step) and language development (appearance of the first word) at the age of one year using parental questionnaire (a total of 43 children participated in this step). To assess speech and language development (SLD), sensory-motor development (SMD) and social-emotional development (SED) of three-year-old children included in this study, we used The Scale for Evaluation of Psychophysiological Abilities of Children (SEPAC) [12, 13, 14] which comprises of three subscale tests (test for assessing SLD, test for assessing the SMD and the test for the assessment of SED) and is used in children aged 0-7 years. All data are collected by the method of individual testing. Depending on the achieved test scores (TS)

on each subscale, expressed in percentages of the correctly done tasks, we calculated the estimated development (ED). The ED is expressed in months corrected for the chronological age (CA) using the formula: ED = CAxTS. Estimated speech-language (ESLD), sensory-motor (ESMD), and social-emotional development (ESED) areused for further analyses. The anamnestic data obtained from the children's parents in face-to-face interviews enabled collection of the data regarding the appearance of the first steps and the first words (calculated in months).

Statistical analyses

The scale variables (PiB, PiR, RT, RePi, GA, MA, STAI-S, STAI-T scores,and BW) were normally distributed so parametric statistics were used. Analysis of covariance (ANCOVA) was used for the comparison of group means. We used covariates in the ANCOVA model to remove their potential contribution to the outcome variable, leaving just the adjusted group means. We employed partial correlation to measure the association between RT, RePi, STAIS, and STAI-T (as continuous variables), after controlling for the GA, MA, and fetal sex. Further, we used partial correlation to measure the association between STAI-T, RT, RePi, ESLD, ESMD, and ESED, after controlling for GA, GAB, and sex. Neonatal BW was compared between low and high anxiety groups using ANCOVA controlling for MA, fetal sex, and GAB. Apgar score was not normally distributed, so we used nonparametric Kruskal Wallis test for the comparison of the group mean. The χ^2 test was employed to explore differences in the distribution of GAB (in term, preterm) between low and high anxiety strata. The effect size was estimated using partial Eta square (η^2) and Cohen's *d*. For each comparison, a 95% confidence interval (CI) was used. Statistical analyses were performed using Statistical Package for the Social Sciences (SPSS) for Windows (v. 22.0).

RESULTS

Effect of antenatal maternal anxiety on fetal reactivity to auditorystimulation

We found no significant effect of STAI-S and STAI-T level on PiB and PiR. Figure 1 presents the mean values of RT and RePi due to low and high STAI-s and STAI-T strata.

There is a statistically significant effect of STAI-S level on RT: $F(1,38) = 15.537$, p< .01), CI [3.746-4.894], $\eta^2 = .285$, Cohen's d = 1.18. We also found a statistically significant effect of STAI-S level on RePi: $F(1,38) = 15.537$, p< .001), CI [13.297-22.597], $\eta^2 = .124$, Cohen's d = 0.73. The effect of STAI-T on RT was also notable: $F(1,38) = 9.678$, p = .003), CI [3.644-4.858], $\eta^2 = .199$, Cohen's d = 0.90, as well as on RePi: $F(1,38) = 5.654$, p = .02), CI [13.307-22.582], $\eta^2 = .127$, Cohen's d = 0.70. From the Figure 1, it can be seen that both RT and RePi values are higher in the high STAI-S and STAI-T strata compared to low anxiety strata.

Effect of antenatal maternal anxiety on GAB, BW, and AS

It can be seen from Table 2 that BW was lower in babies from mother who had high levels of both STAI-S and STAI-T anxiety. However, we found no significant effect of STAI-S and STAI-T anxiety level on BW. In addition, we found no effect of STAI-S and STAI-T anxiety level on AS. We found statistically significant difference in distribution of GAB between low and high STAI-S: $\chi^2 (1) = 10.759$, p < .01, as well as STAI-T: $\chi^2 (1) = 17.319$, p < .01. There were significantly more preterm deliveries in high compared to low STAI-S and STAI-T strata.

Effect of antenatal maternal anxiety on early speech-language, sensory-motor, and social-emotional development

We found no statistically significant effect of STAI-S on AFW, AFS, ESLD, ESMD, and ESED. In addition, we found no statistically significant effect of STAI-S on AFW and AFS. But, there was a statistically significant effect of STAI-T on ESLD: $F(1,39) = 9.526$, p = .004, $\eta^2 = .196$, Cohen's d = 0.96, ESMD: $F(1,39) = 8.197$, p = .007, $\eta^2 = .174$, Cohen's d =

0.80, and ESED: $F_{(1,39)} = 6.004$, $p = .019$, $\eta^2 = .133$, Cohen's $d = 0.69$. Children from mothers who were in the high STAI-T strata achieved lower scores on each subscale of the SEPAC compared to low strata (see Table 3).

Is there an association between STAI-S, STAI-T, fetal reactivity to auditory stimulation and ESLD, ESMD, and ESED?

We found positive linear correlations between RT and STAI-S ($r(38) = .337$, $p = .033$), RT and STAI-T ($r(38) = .392$, $p = .012$), RePi and STAI-S ($r(38) = .367$, $p = .02$), and RePi and STAI-T ($r(38) = .356$, $p = .024$) (see Figure 2).

There were statistically significant negative linear correlations between RT and ESLD ($r(38) = -.597$, $p = .001$), RePi and ESLD($r(38) = -.380$, $p = .016$), and STAI-T and ESLD ($r(38) = .394$, $p = .012$). There are also weak negative correlations between RT and ESMD ($r(38) = -.297$, $p = 0.62$), STAI-T and ESMD ($r(38) = -.309$, $p = 0.57$), and RePi and ESMD ($r(38) = -.277$, $p = 0.84$), but they did not reached the level of statistical significance (see Figure 3).

DISCUSSION

There is a growing body of research aimed at examining how the maternal psychological state might induce long-lasting impact on fetal as well as early child development [9,15]. Lower pulsatility index values in the MCA and higher in the umbilical artery of fetuses of mothers with high levels of trait anxiety was reported [5]. The authors concluded that there might be a change in blood distribution in favor of the brain in fetuses of highly anxious mothers. Our results are in line with this finding. We found a higher cerebral blood flow through MCA after defined auditory stimulation in fetuses of mothers with high anxiety level (both state and trait) compared to low anxiety. Also, positive correlation between mean fetal artery resistance values and maternal anxiety levels are reported [4]. A recent study found an increase in fetal MCA pulsatility index in fetuses of mothers with both anxiety and gestational hypertension [9].

Studies are showing that timing of testing the effect of different threatening factors on the fetal behavior is an important variable that needs to be taken into consideration. No effect of antenatal maternal anxiety on fetal spontaneous motor behavior in the first half of pregnancy was reported [16]. On the contrary, an increased fetal wakefulness and increased fetal heart rate variability in fetuses of high anxious mothers in the last trimester of pregnancy have been reported [17]. We examined fetuses from the last trimester of the pregnancy. The main reason for this is the time when auditory system is mature enough so fetal reactions to defined auditory stimulation can be measured. The second reason is that the overall fetal reactivity is more expressed in second half of the pregnancy as fetus gets more mature. An increased percentage of fetal body movement and fetal heart rate variability in fetuses from highly anxious mothers has been reported [18]. This might be in line with our finding that fetal MCA RePi was higher in fetuses of mothers with high anxiety level. This might imply that reactivity is increased in fetuses of high anxious mothers. However, still remains unclear why in fetuses of high anxious mothers an increased MCA blood flow as a reaction to the external auditory stimulus was found alongside longer reaction time. In addition, we found an effect of both high state and trait anxiety on fetal MCA blood flow increase after auditory stimulation compared to low anxiety. This might be because both maternal state and trait anxiety were measured just before the experimental procedure of testing fetal reaction to defined auditory stimulation. Hence, the association between state anxiety and fetal reactivity to auditory stimulation could be biased. However, the state anxiety assessed by STAI is a measure of the current anxiety level that might have an effect on fetal cerebral circulation.

Studies have shown that negative maternal emotions during pregnancy are associated with an adverse pregnancy outcome. The effect of high antenatal anxiety on both low birth weight for gestational age and preterm delivery are the most reported results [1,3,9,19]. On the contrary, no significant correlation between antenatal maternal anxiety and gestational age at birth was reported [20]. We found no statistically significant effect of both state and trait antenatal maternal anxiety on neonatal body weight. However, there were significantly more preterm deliveries in mothers with a high level of both state and trait anxiety. We found no effect of anxiety on Apgar score. There is a question why we found a significant effect of both state and trait anxiety on gestational age at birth. The answer might lie in the timing of the anxiety assessment. We assessed antenatal maternal state and trait anxiety in the third trimester. However, a recent study found that the effect of antenatal maternal anxiety on the

incidence of preterm delivery remained significant regardless of the gestational period when anxiety was assessed [21].

Antenatal maternal anxiety has been associated with postnatal behavioral, emotional, and cognitive functioning [22]. Our study elicited similar results. We found a negative effect of antenatal maternal trait anxiety on speech-language, sensory-motor, and social-emotional development assessed at the age of three. It needs to be noted here that the effect size was highest for the effect of antenatal maternal anxiety on speech-language development. Antenatal maternal psychological state had an adverse effect on infant's speech perception and speech-language milestones and emotional regulation [23]. Not all studies found an association between prenatal exposure to maternal distress and postnatal speech-language development [24]. We found no significant effect of antenatal maternal state anxiety level on early child development at the age of three. Opposite results are reported in a recent study [25]. The authors reported a significant association between antenatal maternal state anxiety and poorer children's cognitive development at two and three years.

Limitations

This study has several limitations. First, the sample is very narrow. Hence, caution is needed in the interpretation and generalization of the results. The main idea of this study was to probe the potential effect of solely antenatal maternal anxiety on fetal reactivity to auditory stimulus, birth outcome, and postnatal development, so many factors had to be kept under control. Knowing that many factors have an effect on child development, we had to eliminate as much potentially confounding or to keep them under control (either methodologically during sample selection or using appropriate statistical approach). There is a recent study exploring the effect of antenatal maternal anxiety and stress on postnatal development with similar strict sample criteria [26]. The second limitation is that maternal anxiety was assessed only once during pregnancy. Although there are studies that found no association between timing of anxiety assessment, this could be misleading especially regarding state anxiety effect on the birth outcome we found. The third limitation is that we did not measure postpartum maternal anxiety. And finally, there is always a risk that some adverse factors occurred in children during postnatal development that was not reported by parents.

CONCLUSION

This longitudinal prospective study showed a potential negative effect of high levels of antenatal maternal trait anxiety on both, fetal reactivity to sound stimulation (slower reaction time and increased relative pulsatility index) and early child speech-language and social-emotional development. In addition, there is an association between slower fetal reaction time to auditory stimulation and slower early speech-language development.

Conflict of interest: None declared

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Table 1. Sample characteristics

n	43
Maternal age (years)	31.26 (4.18)
Education	
high (BS, MS, PhD)	36 (83.7%)
medium (high school diploma)	7 (16.3%)
low	0 (0%)
Marital status	
married/living with partner	43 (100%)
single	0 (0%)
Maternal monthly salary	
above average	34 (79.1%)
average	9 (20.9%)
low	0 (0%)
STAI-S score	38.21 (9.63)
low	29 (67.44%)
high	14 (32.56%)
STAI-T score	36.47 (6.96)
low	29 (67.44%)
high	14 (32.56%)
Gestational age (weeks) at testing fetal reaction to sound stimulation	35.09 (4.31)
Male fetuses (N)	17 (39.53%)
Children's chronological age at testing (in months)	39.70 (1.92)

Means (standard deviations) are present for the variables maternal age; STAI-S and STAI-T score, gestational age at testing fetal reaction to sound stimulation, and children's chronological age at testing; maternal monthly salary is stratified according to the national average for 2014

Table 2. Birth outcome due to STAI-S and STAI-T level

STAI	LEVEL	BW	AS	GAB-% of ptD
STAI-S	low	3456.9 (480.09)	8.90 (0.72)	3.45% (1/29)
	high	3103.57 (786.04)	8.64 (1.08)	13.95% (6/14)***
STAI-T	low	3543.10 (444.96)	8.93 (0.75)	0% (0/29)
	high	2925.00 (707.85)	8.57 (1.02)	50% (7/14)***
	total	3341.86 (610.71)	8.81 (0.85)	16.28% (7/43)

STAI-S – state anxiety; STAI-T – trait anxiety; BW – neonatal body weight in grams; AS – Apgar score in the 5th minute after delivery; GAB – gestational age at birth; ptD – preterm delivery ($< 37^{th}$ week of gestation);

means (standard deviations) are present for BW and AS

***$p < 0.01$

Table 3. Estimated speech-language, senso-motor, and socio-emotional development in 3 years old children due to antenatal maternal STAI-S and STAI-T level

STAI	LEVEL	AFS	AFW	ESLD	ESMD	ESED
STAI-S	low	12.17 (2.74)	12.21 (1.26)	34.20 (2.04)	34.93 (6.35)	37.53 (4.63)
	high	13.71 (3.89)	13.21 (1.85)	32.08 (3.07)	30.05 (7.58)	35.35 (4.67)
STAI-T	low	12.17 (2.83)	12.40 (1.28)	34.31 (1.89)	35.13 (6.25)	37.87 (4.14)
	high	13.79 (3.76)	13.29 (1.77)	31.86 (3.10)***	29.63 (7.44)***	34.64 (5.18)*
	total	12.70 (3.20)	12.53 (1.53)	33.51 (2.59)	33.34 (7.07)	36.82 (4.70)

STAI-S – state anxiety; STAI-T – trait anxiety; AFS – appearance of the first steps (in months); AFW – appearance of the first words (in months); ESLD – estimated speech-language development (in months); ESMD – estimated sensomotor development (in months); ESED – estimated socio-emotional development (in months);

means (standard deviations) are present;
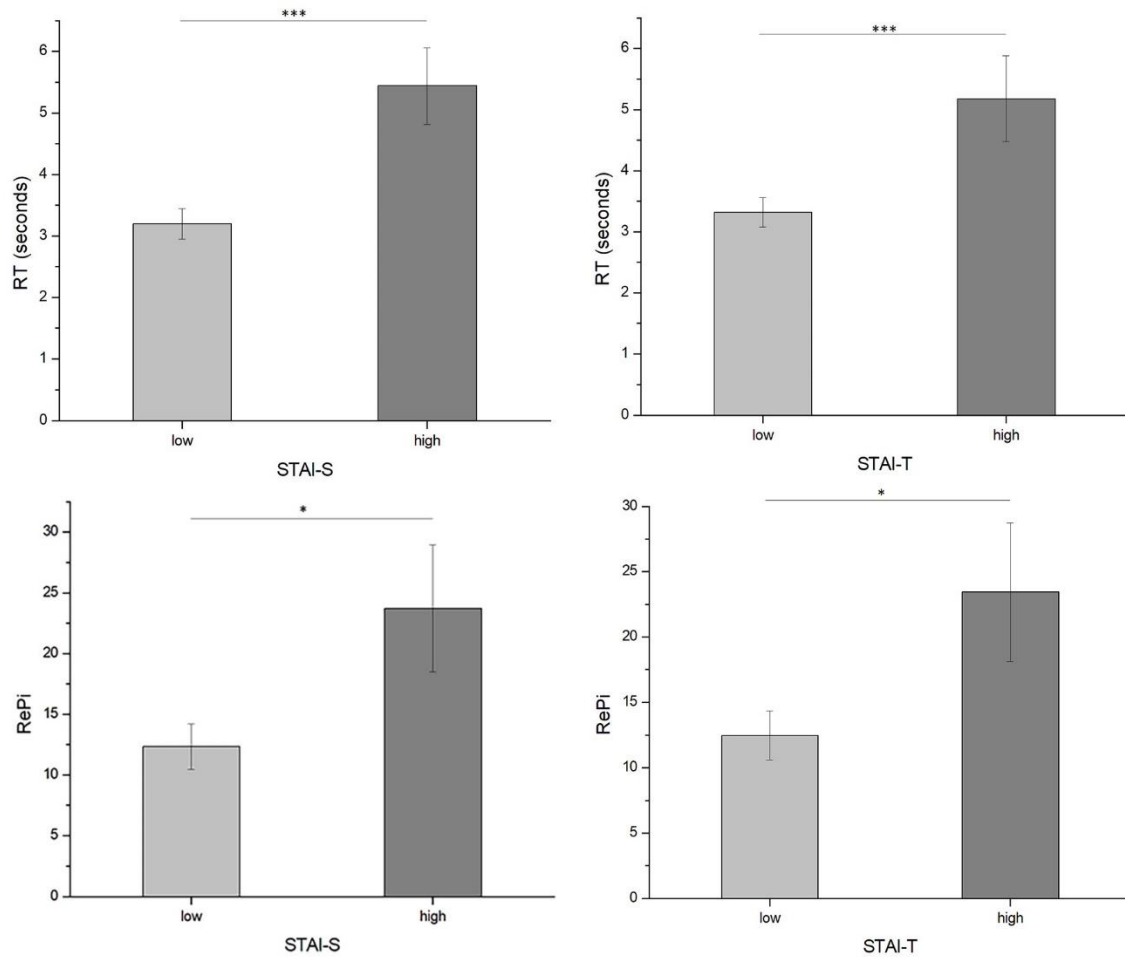
*p < 0.05;

***p < 0.01

Figure 1. Fetal MCA blood flow reaction time (RT) and relative pulsatility index (RePi) after auditory stimulation; the mean (SE) are present for low and high state (STAI-S) and trait (STAI-T) anxiety strata;
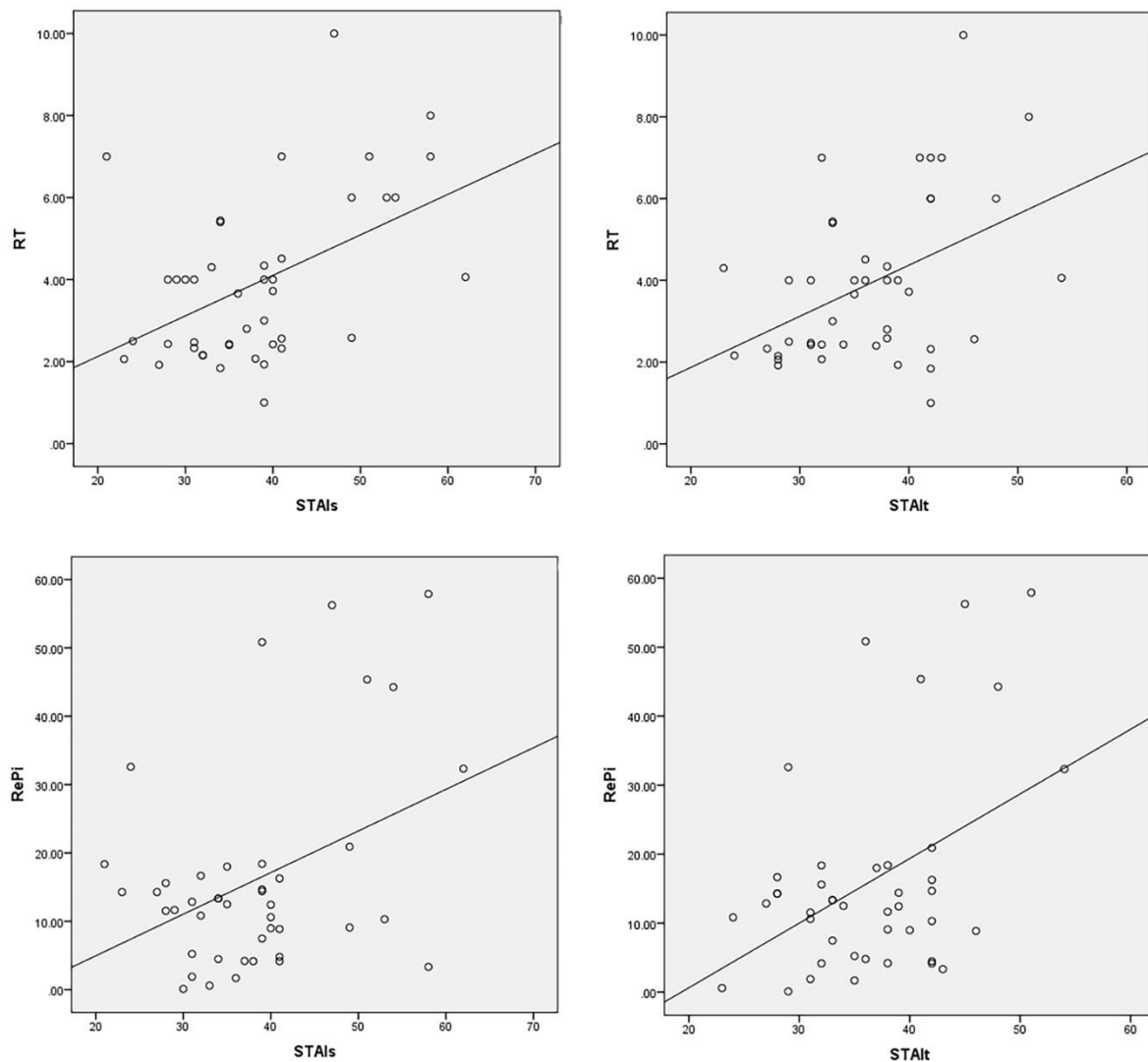
* p < 0.05;

*** p < 0.01

Figure 2. Correlations between fetal MCA blood flow reaction time (RT) and relative pulsatility index (RePi) after auditory stimulation and antenatal maternal anxiety; partial correlations, controlled for maternal age, gestational age when fetal reactivity to auditory stimulation was assessed, and sex (covariates), are present in scatter plots
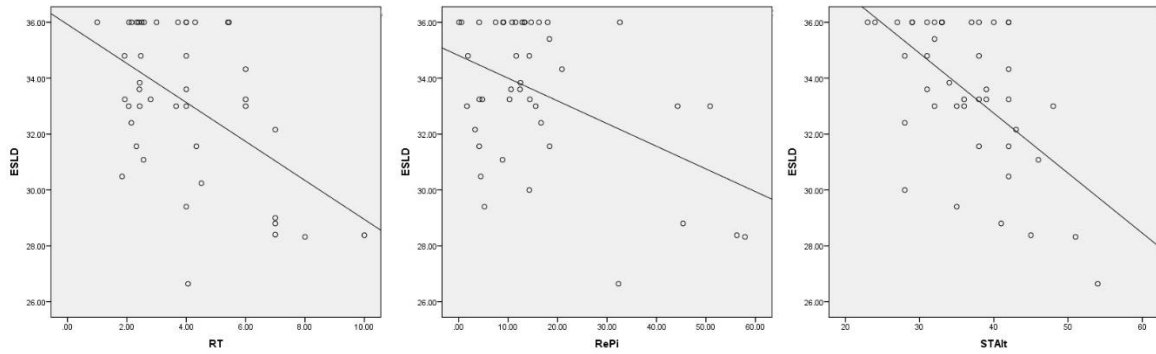
Figure 3. Correlations between estimated speech-language development and fetal MCA blood flow reaction time (RT), relative pulsatility index (RePi), and antenatal maternal trait anxiety (STAI-T); partial correlations, controlled for gestation age at birth, gestational age when fetal reactivity to auditory stimulation was assessed, and sex (covariates), are present in scatter plots