Comprehensive Psychoneuroendocrinology 21 (2025) 100280

Contents lists available at ScienceDirect

Comprehensive Psychoneuroendocrinology

journal homepage: www.sciencedirect.com/journal/comprehensive-psychoneuroendocrinology

Exploring stress and depressive symptoms in pregnancy and the IL-1β, IL-6, and C-reactive protein pathway: Looking for possible biomarker targets☆

Danielle Abukhalaf [a], Rebecca Koerner [a], Sapna Patel [a], Allyson Duffy [a], Stephanie Prescott [a,b,*]

[a] University of South Florida, College of Nursing, United States
[b] Inova Health Services, United States

ARTICLE INFO

Keywords:
Cortisol
Interleukins
Pregnancy
Perceived stress
Depressive symptoms

ABSTRACT

Background: Individuals undergo significant stress throughout pregnancy and are at high risk for depressive symptoms. Elevated stress and depressive symptoms are associated with inflammatory processes and adverse maternal-infant outcomes. However, the biological processes associated with psychosocial outcomes and the maternal immune system remain unclear. As such, we aimed to examine associations among perceived stress, depressive symptoms, salivary IL-1β, IL-6, and CRP levels, and hair and salivary cortisol levels during the second and third trimesters of pregnancy.

Methods: We conducted an ancillary study consisting of 37 pregnant individuals. Participants collected salivary samples and measures of perceived stress and depression at 17–19 weeks, 25–27 weeks, and 32–34 weeks gestation. We collected a one-time hair sample between 36 and 40 weeks. Provided salivary samples were used to detect changes in cortisol, IL-1β, IL-6, and CRP levels. Hair was used to detect changes in cortisol levels throughout pregnancy.

Results: Elevated levels of perceived stress and depressive symptoms are associated with increased salivary CRP levels, respectively ($p = 0.0142$, $p = 0.0008$). Salivary and hair cortisol increased significantly throughout the second and third trimesters of pregnancy ($p = 0.0004$ and $p < 0.0001$). We also observed variations in IL-6 during pregnancy ($p = 0.029$) and significant increases between 25 and 27 weeks ($p = 0.016$).

Conclusion: Salivary samples may provide a non-invasive measurement of alterations in cytokine and cortisol levels in pregnant individuals reporting elevated stress and depressive symptoms. These may be candidate biomarkers for mechanistic study possibly aiding providers in early detection of deleterious immunological processes which could result in adverse maternal-infant outcomes.

1. Introduction

The maternal immune system undergoes pro-inflammatory and anti-inflammatory changes throughout pregnancy that coincide with fetal development. An inflammatory environment in the first and early second trimester promotes the implementation of the blastocyst to the uterine wall [1]. Throughout the second trimester, the fetus undergoes rapid growth, and the mother produces mainly anti-inflammatory cytokines to promote tolerance of the fetal semi-allograft; in the last trimester, local and systemic inflammatory cytokine activation is required for active labor and delivery [2]. Maternal immune dysregulation duringpregnancy can have significant consequences on fetal and maternal health, including-pre-eclampsia, miscarriage, preterm labor, depressive symptoms, and low birth weight infants [3–6].

The Interleukin 1 (IL-1) proinflammatory family of cytokines is primarily associated with innate immunity and thus, inflammation making them invaluable in host defense against foreign antigens and infectious organisms and key players in autoimmune syndromes [7]. IL-1 is typically found dispersed in the cytoplasm but is released in response to microbial products or in sterile conditions in response to hypoxia or tissue injury [8]. IL1-β is produced by innate immune cells and functions as an inflammatory mediator by inducing the activation, differentiation, and proliferation of innate and adaptive immune cells and the expression of other interleukins, including IL-6, cytokines, chemokines, and adhesion molecules [7]; [9]. The IL-1 family of cytokines also plays an important role in implantation, placental development, communication

☆ Funded by the University of South Florida's Women's Health Collaborative.
* Corresponding author. University of South Florida, College of Nursing, United States.
E-mail address: stephanie.prescott@inova.org (S. Prescott).

https://doi.org/10.1016/j.cpnec.2024.100280
Received 31 July 2024; Received in revised form 26 November 2024; Accepted 24 December 2024
Available online 29 December 2024

between the maternal, placental, and fetal tissues, and labor induction during healthy pregnancy [10]. Imbalances between IL-1β and the IL-1 receptor antagonist (IL-1Ra) in maternal serum or at the maternal-fetal interface are implicated in adverse pregnancy outcomes, including preeclampsia, preterm birth, intrauterine growth restriction (IUGR), premature rupture of membranes (PROM), and recurrent miscarriage [10].

The proinflammatory IL-1β induces the production of IL-6 in response to signals from activated immune cells, however IL-6 has both pro- and anti-inflammatory properties and is produced by many cell types during infection, inflammation, or stress [11,12]. Once released, IL-6 stimulates the liver to produce C-reactive protein and other acute phase proteins, which promote the inflammatory response [13], but it has immune-modulating effects in T-cells where it both inhibits and promotes the forkhead box p3 (FoxP3) regulatory T cells responsible for maternal immune tolerance of the fetus [14]. IL-6 is thought to participate in embryonic implantation, and studies in mice indicate it has a role in placenta formation, though IL-6 activity appears to be tightly controlled through early and midgestation [15]. IL-6 activity resumes as labor approaches and is abundant in maternal serum near parturition [13]. Adverse pregnancy outcomes such as miscarriage, premature delivery, hypertension, and infertility are associated with both excess and insufficiency of IL-6 [16].

CRP is produced by the liver primarily in response to IL-6 and, to a lesser degree, IL-1β, but may also be produced in muscle, fat, and immune cells [17]; [18]. CRP is a component of the innate immune system that responds as a pattern recognition molecule that activates the complement system and participates in the activation of adaptive immunity [19]. CRP, an acute reactant, is generated in the liver in response to IL-6. CRP levels increase dramatically in response to inflammation and infection and decrease as the inflammation resolves. CRP binds to damaged tissue and to pathogens, thereby acting as an opsonin, activates complement, and triggers the release of inflammatory cytokines [19]. Elevations in CRP during pregnancy are associated with infection, autism, gestational diabetes, growth restriction, preterm delivery, and hypertension [20]; [21–23].

Elevated levels of IL-1β, IL-6, and CRP are associated with stress and depressive symptoms [24]; [25]. Approximately 11.6–34 % of pregnant individuals report stress, while depressive symptoms occur in 15–28 % of pregnant individuals [26]; [27]. Depression is a common mental health problem in pregnancy, with symptoms most prevalent in the first and third trimesters [28]. Perceived stress and depressive symptoms can increase pro-inflammatory cytokine levels, including CRP, IL-6, IL-1β, and cortisol levels [29,30]. In the setting of chronic stress or depression, imbalances between immune, endocrine, and central nervous systems result from disrupted cortisol feedback loops due to cortisol resistance [31]. Therefore, individuals with depressive symptoms and increased stress are at risk for excessive pro-inflammatory cytokines such as CRP, IL-6, and IL-1β and cortisol levels throughout pregnancy, possibly increasing their risk for adverse maternal and fetal outcomes.

Cortisol is released by the adrenal cortex in response to physiologic and psychological stressors and provides energy to the body by acting on the liver, muscle, adipose tissue, and pancreas to increase glucose availability [32]. It also supports cognitive function and suppresses inflammation [33]. Cortisol circulates both freely and is bound to proteins in the blood. Excess or insufficient cortisol is associated with many pathological conditions and psychological disturbances. During pregnancy, there is a gradual increase in maternal plasma cortisol levels, though cortisol at the fetomaternal interface differs from maternal plasma as the placenta can enzymatically neutralize maternal cortisol by converting it to cortisone [34]; [35]. Evidence suggests pregnancy-induced hypertension and maternal anxiety and depression may alter the enzymatic neutralization of maternal cortisol in the placenta, allowing increased fetal cortisol exposure and associated detrimental effects [36]; [35]; [74]).

Monitoring biomarkers in saliva offers a non-invasive and convenient way to screen individuals for conditions that may lead to maternal and fetal complications. Salivary collection is simple, non-invasive, cost-effective, and available in resource-limited areas [37]; [38]. Cytokine and cortisol levels have successfully been measured in saliva for diagnostic purposes [39,40]. As free cortisol represents the majority of the bioactive components, sampling saliva, which contains only free cortisol, may be superior to blood sampling, which requires separation between free and bound components [41]. Hair cortisol measurements are stable representations of cortisol over a period (1 cm of hair = 1-month average cortisol level), whereas cortisol in the saliva varies throughout the day and represents the state of stress at the time of measurement [42]; [39]. Early detection of excess stress or depressive symptoms that contribute to acute or chronic overabundance of cortisol or cytokine levels may allow for mitigation strategies to be implemented early and reduce pregnancy-related complications.

There is a gap in the literature regarding the use of saliva and hair to assess the cytokines and cortisol levels of pregnant individuals with depressive symptoms and perceived stress. In this ancillary study, we aimed to assess salivary IL-1β, IL-6, and CRP levels, and hair and salivary cortisol levels in existing samples from a study conducted in 2018–2019 to assess the association between perceived stress and depressive symptoms during the second and third trimesters of pregnancy. In the parent study, participants provided salivary samples and completed the Perceived Stress Scale (PSS) and Edinburgh Postnatal Depression Scale (EPDS) at 17–19 weeks, 25–27 weeks, and 32–34 weeks. The three time points were selected to capture the second and third trimesters and align with routine prenatal visits. We hypothesized that there would be an increase in CRP, IL-6, and IL-1β levels, and in hair and salivary cortisol levels in the second and third trimesters in women with elevated perceived stress and/or depressive symptoms.

2. Methods

2.1. Participants

Fifty-nine participants from a parent study that examined stress perception throughout pregnancy and collected salivary and hair samples were screened for eligibility, and 37 participants permitted the use of their specimens. The University of South Florida Institutional Review Board approved the parent and ancillary study. The parent study recruited 59 pregnant individuals during their first trimester of pregnancy at an obstetrics and gynecology clinic in Tampa, Florida [43]. Inclusion criteria for the parent study were: (1) adults over 18 years of age; (2) in their first trimester of pregnancy; (3) hair length of at least 10 cm; (4) had access to a mobile phone. The exclusion criteria for the parent study were: (1) unable to read, write, or speak English. The inclusion criteria for this ancillary study were participants who provided written consent for the future use of their specimens in the parent study.

2.2. Salivary cytokines and cortisol

Participants were instructed to passively drool in a conical tube four times throughout the day between 17 and 19 weeks, 25–27 weeks, and 32–34 weeks and freeze the sample immediately after collecting. The four time points of salivary collection were upon awakening, 30 min after awakening, 3 h after awakening, and 12 h after awakening. These time points were selected to capture patterns of cortisol levels and the average total daily production of cortisol [44]. The weeks of gestation were chosen to coincide with routine prenatal visits and to capture the second and third trimesters. Participants were instructed to avoid saliva collection within 60 min of eating, drinking, brushing their teeth, and smoking. Participants were asked to bring frozen samples on ice to the three prenatal appointments. Upon receipt of the samples, they were aliquoted and then stored in a −80 °C freezer. Before the assay, salivary samples were thawed entirely, vortexed, and centrifuged at 1500×*g* for 15 min. Salivary cortisol levels were evaluated using Salimetrics cortisol

ELISA kits (#1–3002), and salivary levels of CRP, IL-6, and IL-1β were evaluated using Salimetrics ELISA kits (C-Reactive Protein ELISA Kit (Generation II) Item No. 1–2012; IL-6 ELISA KIT Item No. 1–3602; IL-1β ELISA Kit Item No. 1–3602). Standards and controls were completed with each assay. ELISAs were performed according to the manufacturer's instructions, and results were calculated using the standard curves for each assay. Values with a coefficient of variation >15 % were repeated, Undetectable results were reported as 0. Salimetrics reports the intra-assay precision as 0.06–2.07, coefficient of variation 3–7%; the inter-assay precision as 0.06–1.99, coefficient of variation 3–11 %; and the sensitivity as 0–0.007 μg/dL.

2.3. Hair cortisol

A 10 cm hair sample was collected in the parent study by cutting as close to the scalp as possible, either at the root from the nape of the neck or the posterior vortex between 36 and 40 weeks gestation. Hair grows approximately 1 cm a month, and the most proximal 1 cm segment to the scalp should approximate the last month's cortisol production [45]; therefore, the hair sample was further cut into three 3 cm segments to represent all three trimesters. Approximately 10 mg of each hair segment was weighed, then cut into smaller pieces, and placed into a grinding tube. Then, the hair was ground to powder, and the weight was reassessed. Instructions were followed to complete the assay Salimetrics cortisol ELISA kit.

2.4. Edinburgh postnatal depression scale

The Edinburgh Postnatal Depression Scale (EPDS) is a 10-item instrument used as a screening tool for perinatal depressive symptoms [46]. Scores can range from 0 to 30, with a clinical cut-off point indicating the mother may have depressive symptoms and need to be evaluated for clinical depression [46]. The parent study chose a clinical cut-off point of 10, which is recommended for the best sensitivity and specificity during pregnancy [47]. A score <10 was classified as low, and a score ≥10 was classified as high. This instrument has demonstrated evidence of reliability and validity for measuring depressive symptoms during pregnancy [47].

2.5. Perceived stress scale

The Perceived Stress Scale (PSS) is a psychological instrument that measures the degree to which situations in an individual's life are perceived as stressful [48]. The instrument has 14 items, and each item is self-rated on a 5-point Likert scale where 0 (never), 1 (almost never), 2 (sometimes), 3 (fairly often), and 4 (very often), and higher scores indicated higher levels of perceived stress [48]. A score <13 indicates low perceived stress, a score between 14 and 26 is medium, and a score >26 indicates a high amount of perceived stress. The PSS has demonstrated evidence of reliability and validity in pregnant women throughout pregnancy [49].

2.6. Statistical analyses

GraphPad Prism v. 10.2.3 for Mac OS 13.0 was used for analysis. Descriptive statistics are presented as percentages or means with standard deviations or as frequency distributions for categorical data (Table 1). Salivary samples were collected during regularly scheduled prenatal visits and grouped into time points 17–19 weeks, 25–27 weeks, and 32–34 weeks for analysis. Participants were divided for comparison into groups: PSS low (scores <13, = 13), medium (scores between 14 and 26, n = 13) and high (scores >26, n = 11), and EPDS low (scores <10, n = 28) and high (scores ≥10, n = 9). Robust nonlinear regression and false discovery rate (ROUT, Q = 1 %) was used to detect and remove significant outliers [50]. Data were then fitted to mixed effects models with Geisser-Greenhouse correction for non-sphericity and controlled for false discovery using the original Benjamini and Hochberg method (FDR<0.1) [51]. Fixed effects models were used because missing data ruled out a repeated measures ANOVA analysis approach, and the reasons for missing data were random. False discovery rates, expressed as q values, were used to quantify the rate of type 1 errors when conducting multiple comparisons.

Table 1
Participant characteristics (n = 37).

Variable	n (%) or Mean ± SD
Ethnicity	
Hispanic	5 (13.5)
Non-Hispanic	32 (86.5)
Race	
Asian	2 (5.4)
Black	2 (5.4)
Native Hawaiian/API	1 (2.7)
Other	3 (8.1)
White	29 (78.4)
Age	
Age range (years)	21–41
Gestational age at delivery	38.5 ± 2.20
Maternal age	31.86 ± 4.50
Body Mass Index	
Normal	18 (48.6)
Obese	10 (27)
Overweight	6 (16)
Underweight	2 (5.4)
Hypertensive during pregnancy	
Yes	8 (21.6)
No	34 (78.4)
Gestational Diabetes Mellitus	
Yes	3 (8.1)
No	34 (91.9)

3. Results

3.1. Participant characteristics

The individuals in this study were predominantly white and non-Hispanic, aged 21 to 41. Most of the participants did not have any comorbidities during this pregnancy and had a normal pregestational BMI. The infants of the women in this study delivered at a mean gestational age of 38.5 ± 2.20 weeks. The participant's characteristics are shown in Table 1.

3.2. Stress, depressive symptoms, and associated biomarkers across pregnancy

Perceived Stress scores (PSS) and The Edinburgh Postnatal Depression Scale (EPDS) were not significantly different throughout pregnancy (p = 0.08 and p = 0.77, respectively), though the PSS scores were higher at 25–27 weeks than at the 17–19 weeks or 32–34 weeks (p = 0.03, q = 0.08 and p = 0.06, q = 0.08, respectively). Stress and depression scores across pregnancy are presented in Fig. 1.

Salivary and hair cortisol increased significantly throughout pregnancy (p = 0.0004 and p < 0.0001, respectively). However, salivary cortisol levels did not change by PSS (p = 0.47) or EPDS (p = 0.82). Hair cortisol did not vary by stress level over pregnancy (p = 0.42), but hair cortisol trends were higher in women with EPDS scores ≥10 throughout pregnancy (p = 0.11). See cytokine and cortisol levels across pregnancy in Fig. 2.

IL-1β did not vary significantly across pregnancy (p = 0.24) but trended lowest at 17–19 weeks (q = 0.14), and in women with high PSS scores when compared to women with low PSS scores (p = 0.17), and in women with high EPDS scores (p = 0.24). Fig. 3. IL-6 varied significantly

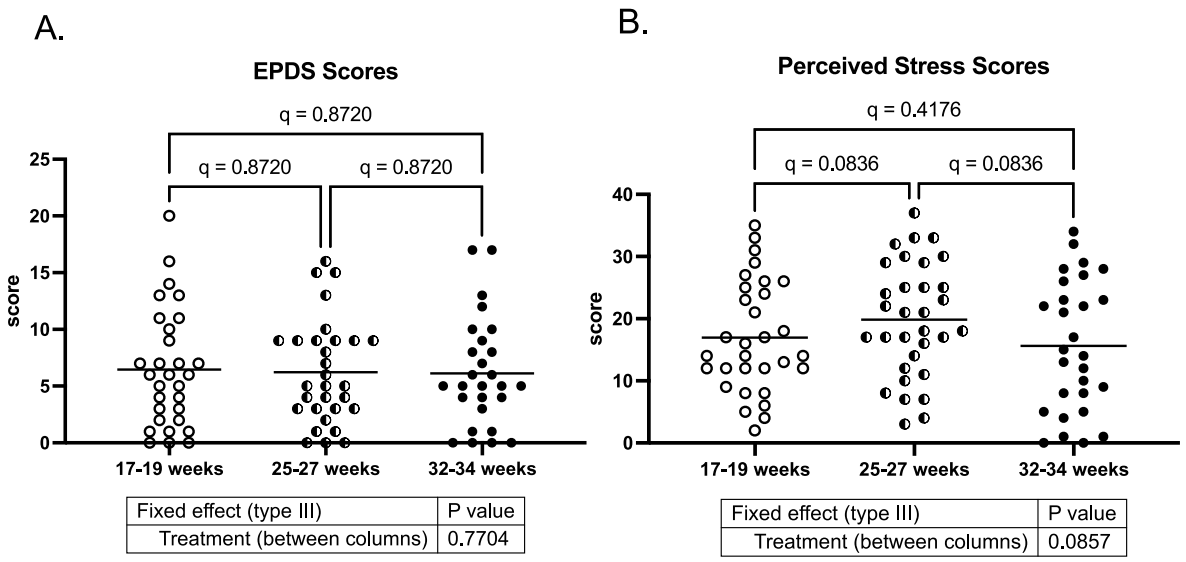


Fig. 1. Edinburgh Postnatal Depression Scale (EPDS) and Perceived Stress Scale (PSS)
Edinburgh Postnatal Depression and Perceived Stress scores did not differ significantly across pregnancy. Scores at 3 timepoints across pregnancy were fitted to mixed effects models to accommodate missing data and were corrected for multiple comparisons using the original Benjamani and Hochber method. False Discovery Rates of $q < 0.1$ are considered significant.

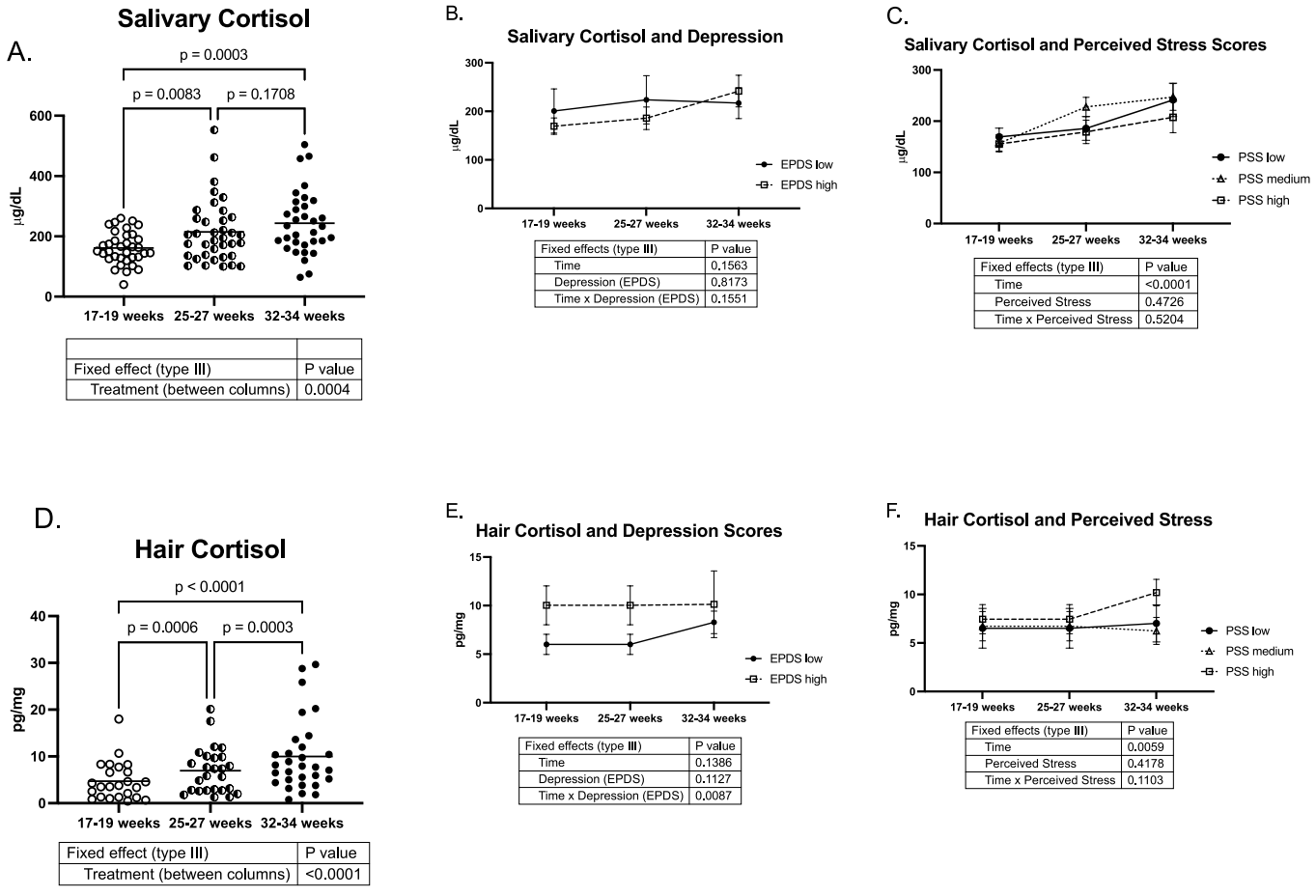
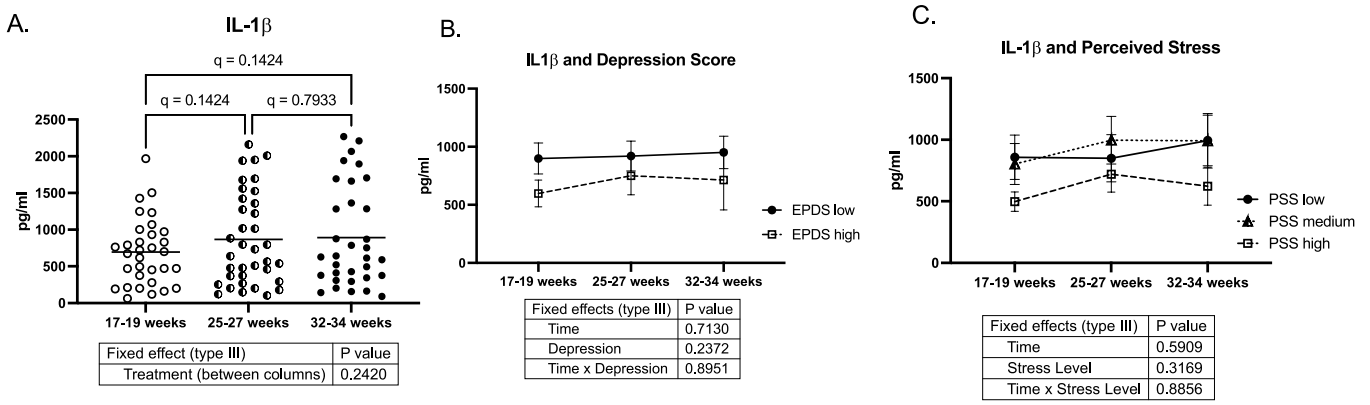


Fig. 2. Salivary and Hair Cortisol Levels and EPDS and PSS scores
Salivary and hair cortisol levels increase significantly across pregnancy, and hair cortisol trends higher in women with EPDS >10. Levels were fitted to mixed effects models to accommodate missing data and were corrected for multiple comparisons using the original Benjamani and Hochber method. False Discovery Rates of $q < 0.1$ are considered significant. Hair cortisol levels were normalized by the weight of the hair tested. Means with SEM.



Fixed effect (type III)	P value
Treatment (between columns)	0.2420

Fixed effects (type III)	P value
Time	0.7130
Depression	0.2372
Time x Depression	0.8951

Fixed effects (type III)	P value
Time	0.5909
Stress Level	0.3169
Time x Stress Level	0.8856

Fig. 3. Salivary IL-1b levels and EPDS and PSS scores
Salivary IL-1β levels did not vary significantly across pregnancy, but trends lower in women with EPDS >10 and in women with high PSS scores. Levels were fitted to mixed effects models to accommodate missing data and were corrected for multiple comparisons using the original Benjamani and Hochber method. False Discovery Rates of $q < 0.1$ are considered significant. Means with SEM.

throughout pregnancy ($p = 0.029$) and is elevated at 25–27 weeks ($p = 0.016$, $q = 0.049$). Women who reported low or moderate perceived stress levels had higher IL-6 levels beginning at 25 weeks and at 34 weeks, while women in the high EPDS group trend lower than those in the low EPDS group but are not significantly different ($p = 0.37$). Fig. 4.

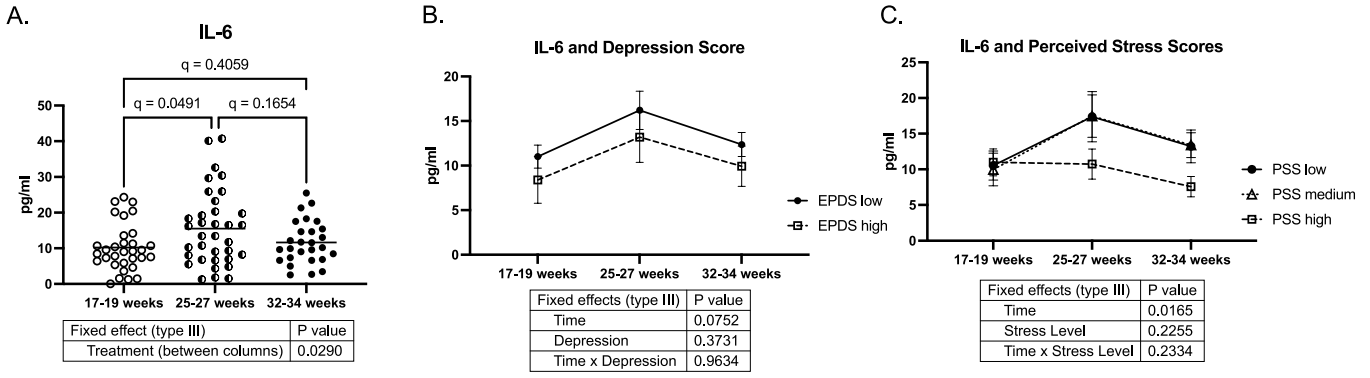
Salivary CRP does not vary significantly across pregnancy ($p = 0.09$), although it trends higher at 32–34 weeks than at 17–19 weeks ($p = 0.023$, $q = 0.068$), or at 25–27 weeks ($p = 0.09$, $q = 0.14$). Salivary CRP levels are, however, significantly increased in women with elevated PSS scores ($p = 0.0142$) and in women with high EPDS scores (EPDS ≥10, $p = 0.0008$). Fig. 5.

4. Discussion

We investigated the association between perceived stress and depressive symptoms, salivary cortisol, and hair cortisol levels, and IL-1β, IL-6, and C-reactive protein (CRP) at three time points during the second and third trimesters of pregnancy. We expected that pregnant women with elevated perceived stress and/or depressive symptoms would have increased hair and salivary cortisol levels and hypothesized that the inflammatory pathway of IL-1β, IL6, and CRP would be elevated with higher perceived stress or depressive symptoms. Existing research in pregnant women has revealed a positive correlation between plasma cytokines, hair and plasma cortisol levels, and perceived stress or depressive symptoms, but research into these biomarkers in saliva is limited [52–57]. As elevated levels of these cytokines and inflammatory markers have been associated with adverse maternal and neonatal outcomes, we projected that the ease and accessibility of salivary testing may lead to early identification of women who might benefit from early intervention to reduce stress and depressive symptoms.

In our cohort, 11 of 37 women reported high stress (PSS score >26), and 9 of 37 women reported increased (EPDS ≥10) depressive symptoms. The proportion of women experiencing these symptoms did not change throughout the three time points. These percentages in our population are consistent with previous studies documenting the prevalence of increased stress and depression in pregnancy [58]; [59]. There were no significant differences in EPDS scores over the three time points and only a slight increase in reported stress scores at the 25–27-week time point, which returned to baseline by 32–34 weeks. Twenty-seven weeks marks the end of the second trimester, a time when pregnant women are beginning to feel symptoms of pregnancy, including aches and pains, mood swings, fatigue, bloating, and constipation, which may account for the slight increase in stress in the cohort at this time point (Fig. 1).

Salivary and hair cortisol increased significantly throughout the second and third trimesters ($p = 0.0004$ and $p < 0.0001$), which is consistent with prior studies [60]; [61,62]. The increase in cortisol over pregnancy is seen despite no increase in reported stress or depression in this cohort, which reinforces that cortisol increases are a physiologic change during pregnancy independent of disease or psychological



Fixed effect (type III)	P value
Treatment (between columns)	0.0290

Fixed effects (type III)	P value
Time	0.0752
Depression	0.3731
Time x Depression	0.9634

Fixed effects (type III)	P value
Time	0.0165
Stress Level	0.2255
Time x Stress Level	0.2334

Fig. 4. Salivary IL-6 levels and EPDS and PSS scores
Salivary IL-6 levels varied significantly across pregnancy, but trends lower in women with EPDS >10 and in women with high PSS scores. Levels were fitted to mixed effects models to accommodate missing data and were corrected for multiple comparisons using the original Benjamani and Hochber method. False Discovery Rates of $q < 0.1$ are considered significant.

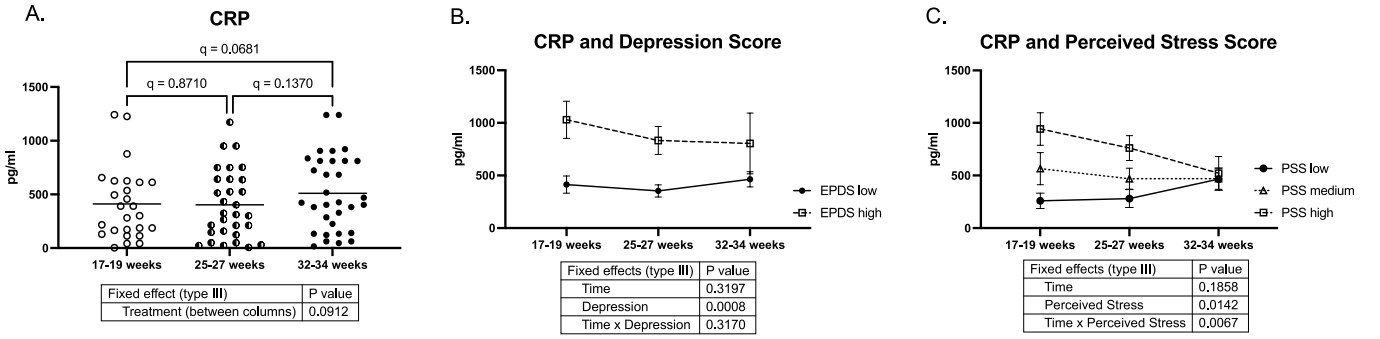


Fig. 5. Salivary C-Reactive Protein (CRP) Levels, EPDS and PSS scores

Salivary CRP levels at three timepoints across pregnancy were fitted to mixed ef fects models to accomadate missing data and were corrected for multiple comparisons using the original Benjamini and Hochber method. FDR <0.1 is considered significant. Means with SEM. Salivary CRP levels did not vary significantly across pregnancy but are significantly higher in women with EPDS >10 and in women with high PSS scores. Levels were fitted to mixed effects models to accommodate missing data and were corrected for multiple comparisons using the original Benjamani and Hochber method. False Discovery Rates of q < 0.1 are considered significant.

burden. Bleker et al. [34] studied serum cortisol in 3039 women in 2017 and found that changes in cortisol were explained by biological factors, including maternal age, infant sex, and CRP levels, and lifestyle variables, such as smoking and sleep duration, rather than depression, anxiety, self-reported stress, or fatigue. There were, however, no significant differences in salivary or hair cortisol levels by either stress or depressive symptoms in this study (Fig. 2). There was a trend, though, toward higher hair cortisol levels throughout pregnancy ($p = 0.12$) in women with depressive symptoms and at 32–34 weeks in women perceiving high stress ($p = 0.03$, without FDR correction). Some previous studies provide evidence of hair cortisol, a more stable measure of sustained cortisol elevation, increases in pregnant individuals with perceived stress and depressive symptoms [52]. Cortisol is important for fetal organ maturation, but excess levels can lead to neurodevelopmental, autoimmune, metabolic, and cardiovascular diseases [63]. Placental enzymes inactivate much of the maternal cortisol to prevent fetal excess, but severe maternal depression and anxiety have been linked to reduced activity of these protective measures, potentiating increased fetal exposure to cortisol [34]; [35]. Future studies should be powered and designed to examine the mechanisms by which cytokines influence maternal mood and should include placental enzymes to further examine the relationships between those enzymes and maternal depression and anxiety.

In our cohort, IL-1β did not vary significantly across pregnancy ($p = 0.24$) but trended lowest in women with high PSS scores ($p = 0.21$) (Fig. 3), and in women with high EPDS scores ($p = 0.26$). IL-1β is a cytokine that augments the body's response to antigens and activates lymphocyte function, thus protecting against pathogens and sterile damaged tissue. It is critical to host defense, but also contributes to autoimmune diseases [7]. Previous research has revealed that in pregnancy, inflammatory pathways driven by IL-1β were localized to reproductive tissues prior to labor but mainly in women entering labor [64]. IL-1 alpha, IL-1β, and IL-1 receptor antagonists have been used with 86 % sensitivity and 92 % specificity to predict term labor in cervicovaginal fluid [65]. In plasma, IL-1β normally increases shortly before delivery before returning to normal, but elevations have been associated with chorioamnionitis, preterm rupture of membranes, and adverse neonatal outcomes such as cardiovascular dysfunction and growth B IL-1β [10]. Perhaps the increased demand from both psychoneurological symptoms and compensation during pregnancy causes a reduction in available IL-1β or the sequestering of IL-1β in distal tissues. A longitudinal study with larger sample size is required to replicate and understand these findings.

IL-6 has many functions during pregnancy. It not only directs T-cell differentiation to modulate the maternal immune response to the fetal semi-allograft, but also participates in the regulation of implantation and placenta formation [16]. IL-6 levels are significantly increased during healthy pregnancy [13], though there is conflicting evidence surrounding when IL-6 is highest during pregnancy; some prior studies have documented IL-6 peaking in the third trimester while others document the peak during the second trimester [13]. In this study, we observed peak IL-6 levels in all tested women at 25–27 weeks ($p = 0.016$), the time point just at the end of the second trimester. Interestingly, as with IL-1β, women in our cohort who reported high perceived stress or depressive symptoms trended lower compared to women with lower stress and depressive symptoms. This difference increased through pregnancy but never attained statistical significance. Although this is consistent with our previous results, as IL-1β is a potent inducer of IL-6, these results are contrary to prior studies in which salivary or plasma IL-6 levels increase in response to acute stress and severity of depressive symptoms throughout pregnancy [66]; [67]. The variation in results in many cytokine studies results from small sample sizes, different sampling methods, different sampling times, and comorbid conditions. Also, selective serotonin reuptake inhibitors (SSRIs) are commonly used antidepressants during pregnancy and can reduce systemic IL-6 levels [68].; though most of the women with high EPDS and PSS scores reported not taking them in this study. This may be important and merit further research as SSRI use, especially later in pregnancy. SSRI exposure has been linked to respiratory, motor, central nervous system, and gastrointestinal symptoms in 10–30 % of newborns [69]. In acute stress situations, cortisol can inhibit the inflammatory response, therefore decreasing cytokine levels, while abnormal elevation of IL-6 during pregnancy has been linked to preeclampsia in the mother and cognitive impairment in the child [70]; [71,72]. Well designed and controlled studies are needed to elucidate IL-6 as a possible biomarker of acute stress and severe depressive symptoms in pregnancy.

Serum elevations of CRP in pregnancy are seen in the setting of maternal or intrauterine infection and have been documented in the first trimester of fetuses delivered small for gestational age or prematurely [23]. In our cohort, salivary CRP levels increased between the first and third time periods ($q = 0.06$ and $q = 0.14$) and were significantly elevated in women with high PSS scores ($p = 0.0142$), and in those with EPDS scores ≥ 10 ($p = 0.0008$). Differences were greatest at the earliest time point and resolved by the last time point. These findings are consistent with prior studies that observed an increase in plasma CRP levels in women with high PSS scores [54] and depressive symptoms [56], but this is somewhat inconsistent with our own findings of suppressed IL-1β and IL-6 in these groups. IL-1β is known to induce IL-6, which induces CRP, so elevations in CRP in these groups must be in response to other signaling molecules, which were not examined in this

study. Both IL-6 and CRP have two mechanisms of release, and both molecules participate in tissue repair. Thus, it is possible that changes in response to pregnancy complicate the analysis of these biomarkers [73]. Notably, although CRP is elevated in both high stress and depressive symptom groups in our study, it does decrease throughout pregnancy in contrast to the low stress and depressive symptom groups, which increase throughout pregnancy. CRP may be a useful biomarker in pregnant women with stress and depressive symptoms, but requires well designed longitudinal studies to understand the various mechanisms interacting to change the level of this biomarker throughout pregnancy.

5. Limitations

As an ancillary study using samples from a previous study, our study was not powered or designed to test the causal mechanisms of the associations discovered. Lack of diversity within the sample also limited the ability to generalize our results as most of our participants were White and non-Hispanic. Importantly, there was no information on the participants' oral health, which may have influenced our biomarkers, as poor oral health is associated with increased inflammation and salivary cytokines. If used for future biomarker discovery, oral health exam would be needed as part of prenatal care as this may be a notable confounder of salivary cytokine results. Poor oral health has been linked to adverse health outcomes. For this study, we eliminated extreme outliers in all data sets using statistical methods to understand the average response to stress throughout pregnancy, but the outliers may yield insights into individual sources of variation of clinical significance. Surely, if a patient presented with extremely high inflammatory markers or acute phase reactants, it would trigger further clinical investigation, but the inclusion of outliers impeded analysis of the group, and our sample had low incidence of adverse pregnancy outcomes.

6. Conclusion

In this study, we observed that elevated levels of perceived stress and depressive symptoms are associated with increased salivary cytokines and salivary and hair cortisol levels in pregnant individuals in the second and third trimesters. Further study of these biomarkers is warranted as saliva is readily available for collection at routine visits and requires no special training to collect. If these biomarkers are confirmed as important indicators of increased stress and depression, it would be feasible to collect longitudinally at routine pregnancy visits, thereby alerting providers early to possible problems that may be addressed before adverse outcomes develop. Oral health exams would need to be included to eliminate inflammatory confounders. Large studies powered to distinguish effects due to common conditions of pregnancy that include diverse populations and those with pregnancy complications would aid in understanding the natural variations that occur during pregnancy and differentiating the mechanisms and expected results of changes in cytokine levels in this pathway when problems develop.

CRediT authorship contribution statement

Danielle Abukhalaf: Writing – original draft, Methodology, Formal analysis, Data curation. **Rebecca Koerner:** Writing – review & editing, Project administration, Methodology, Investigation, Data curation, Conceptualization. **Sapna Patel:** Investigation, Data curation. **Allyson Duffy:** Writing – review & editing, Project administration, Methodology, Investigation, Funding acquisition, Data curation, Conceptualization. **Stephanie Prescott:** Writing – review & editing, Visualization, Supervision, Software, Methodology, Investigation, Funding acquisition, Formal analysis, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

References

[1] A.H. Jarmund, G.F. Giskeodegard, M. Ryssdal, B. Steinkjer, L.M.T. Stokkeland, T. S. Madssen, S.N. Stafne, S. Stridsklev, T. Moholdt, R. Heimstad, E. Vanky, A. C. Iversen, Cytokine patterns in maternal serum from first trimester to term and beyond, Front. Immunol. 12 (2021) 752660, https://doi.org/10.3389/fimmu.2021.752660.
[2] L.J. Yockey, A. Iwasaki, Interferons and proinflammatory cytokines in pregnancy and fetal development, Immunity 49 (3) (2018) 397–412, https://doi.org/10.1016/j.immuni.2018.07.017.
[3] D. Kong, H. Wang, Y. Liu, H. Li, H. Wang, P. Zhu, Correlation between the expression of inflammatory cytokines IL-6, TNF-α and hs-CRP and unfavorable fetal outcomes in patients with pregnancy-induced hypertension, Exp. Ther. Med. 16 (3) (2018) 1982–1986, https://doi.org/10.3892/etm.2018.6393.
[4] X. Lan, L. Guo, S. Zhu, Y. Cao, Y. Niu, S. Han, J. Yan, First-trimester serum cytokine profile in pregnancies conceived after assisted reproductive technology (ART) with subsequent pregnancy-induced hypertension, Front. Immunol. 13 (2022) 930582, https://doi.org/10.3389/fimmu.2022.930582.
[5] S.K. Munro, B. Balakrishnan, A.C. Lissaman, P. Gujral, A.P. Ponnampalam, Cytokines and pregnancy: potential regulation by histone deacetylases, Mol. Reprod. Dev. 88 (5) (2021) 321–337, https://doi.org/10.1002/mrd.23430.
[6] P. Shriyan, P. Sudhir, O.C.P. van Schayck, G.R. Babu, Association of high cortisol levels in pregnancy and altered fetal growth. Results from the MAASTHI, a prospective cohort study, Bengaluru, Lancet Regional Health Southeast Asia 14 (2023) 100196, https://doi.org/10.1016/j.lansea.2023.100196.
[7] C.A. Dinarello, Overview of the IL-1 family in innate inflammation and acquired immunity, Immunol. Rev. 281 (1) (2018) 8–27, https://doi.org/10.1111/imr.12621.
[8] P. Rider, Y. Carmi, O. Guttman, A. Braiman, I. Cohen, E. Voronov, R.N. Apte, IL-1α and IL-1β recruit different myeloid cells and promote different stages of sterile inflammation, J. Immunol. 187 (9) (2011) 4835–4843, https://doi.org/10.4049/jimmunol.1102048.
[9] B. Van Den Eeckhout, J. Tavernier, S. Gerlo, Interleukin-1 as innate mediator of T cell immunity, Front. Immunol. 11 (2020) 621931, https://doi.org/10.3389/fimmu.2020.621931.
[10] O. Equils, C. Kellogg, J. McGregor, M. Gravett, G. Neal-Perry, C. Gabay, The role of the IL-1 system in pregnancy and the use of IL-1 system markers to identify women at risk for pregnancy complications, Biol. Reprod. 103 (4) (2020) 684–694.
[11] E. Choy, S. Rose-John, Interleukin-6 as a multifunctional regulator: inflammation, immune response, and fibrosis, Journal of Scleroderma and Related Disorders 2 (2_suppl) (2017) 1–5, https://doi.org/10.5301/jsrd.5000265.
[12] P.M. Ridker, From C-reactive protein to interleukin-6 to interleukin-1: moving upstream to identify novel targets for atheroprotection, Circ. Res. 118 (1) (2016) 145–156, https://doi.org/10.1161/CIRCRESAHA.115.306656.
[13] T. Spence, P.J. Allsopp, A.J. Yeates, M.S. Mulhern, J.J. Strain, E.M. McSorley, Maternal serum cytokine concentrations in healthy pregnancy and preeclampsia, Journal of Pregnancy 2021 (2021) 6649608, https://doi.org/10.1155/2021/6649608.
[14] X. Gao, Y. Zhong, Y. Liu, R. Ding, J. Chen, The role and function of regulatory T cells in toxoplasma gondii-induced adverse pregnancy outcomes, The Journal of Immunology Research 2021 (2021) 8782672, https://doi.org/10.1155/2021/8782672.
[15] S. Deb, C. Tessier, A. Prigent-Tessier, U. Barkai, S. Ferguson-Gottschall, R. K. Srivastava, J. Faliszek, G. Gibori, The expression of interleukin-6 (IL-6), IL-6 receptor, and gp130-kilodalton glycoprotein in the rat decidua and a decidual cell line: regulation by 17beta-estradiol and prolactin, Endocrinology 140 (10) (1999) 4442–4450, https://doi.org/10.1210/endo.140.10.7063.
[16] J.R. Prins, N. Gomez-Lopez, S.A. Robertson, Interleukin-6 in pregnancy and gestational disorders, J. Reprod. Immunol. 95 (1–2) (2012) 1–14, https://doi.org/10.1016/j.jri.2012.05.004.
[17] N.R. Sproston, J.J. Ashworth, Role of C-reactive protein at sites of inflammation and infection, Front. Immunol. 9 (2018) 754, https://doi.org/10.3389/fimmu.2018.00754.
[18] L. Wirestam, S. Pihl, M. Saleh, J. Wetterö, C. Sjöwall, Plasma C-reactive protein and pentraxin-3 reference intervals during normal pregnancy, Front. Immunol. 12 (2021) 722118, https://doi.org/10.3389/fimmu.2021.722118.
[19] T.W. Du Clos, Function of C-reactive protein, Ann. Med. 32 (4) (2000) 274–278, https://doi.org/10.3109/07853890009011772.
[20] A. Amirian, F.A. Rahnemaei, F. Abdi, Role of C-reactive Protein(CRP) or high-sensitivity CRP in predicting gestational diabetes Mellitus: systematic review, Diabetes Metabol. Syndr. 14 (3) (2020) 229–236, https://doi.org/10.1016/j.dsx.2020.02.004.
[21] R. Hamadeh, A. Mohsen, F. Kobeissy, A. Karouni, H. Akoum, C-reactive protein for prediction or early detection of pre-eclampsia: a systematic review, Gynecol. Obstet. Invest. 86 (1–2) (2021) 13–26, https://doi.org/10.1159/000515530.
[22] R. Nadeem, T. Hussain, H. Sajid, C reactive protein elevation among children or among mothers' of children with autism during pregnancy, a review and meta-analysis, BMC Psychiatr. 20 (1) (2020) 251, https://doi.org/10.1186/s12888-020-02619-8.
[23] R. Nikbakht, E.K. Moghadam, Z. Nasirkhani, Maternal serum levels of C-reactive protein at early pregnancy to predict fetal growth restriction and preterm delivery:

a prospective cohort study, International Journal of Reproductive Biomedicine 18 (3) (2020) 157–164, https://doi.org/10.18502/ijrm.v18i3.6710.
[24] S. Harsanyi, I. Kupcova, L. Danisovic, M. Klein, Selected biomarkers of depression: what are the effects of cytokines and inflammation? Int. J. Mol. Sci. 24 (1) (2022) https://doi.org/10.3390/ijms24010578.
[25] L. Orsolini, S. Pompili, S. Tempia Valenta, V. Salvi, U. Volpe, C-reactive protein as a biomarker for major depressive disorder? Int. J. Mol. Sci. 23 (3) (2022) https://doi.org/10.3390/ijms23031616.
[26] E.V. Moraes, R.N. Campos, M.M. Avelino, Depressive symptoms in pregnancy: the influence of social, psychological and obstetric aspects, Rev. Bras. Ginecol. Obstet. 38 (6) (2016) 293–300, https://doi.org/10.1055/s-0036-1585072.
[27] K. Tanpradit, K. Kaewkiattikun, The effect of perceived stress during pregnancy on preterm birth, Int. J. Wom. Health 12 (2020) 287–293, https://doi.org/10.2147/IJWH.S239138.
[28] H. Lee, K.E. Kim, M.Y. Kim, C.G. Park, J.Y. Han, E.J. Choi, Trajectories of depressive symptoms and anxiety during pregnancy and associations with pregnancy stress, Int. J. Environ. Res. Publ. Health 18 (5) (2021), https://doi.org/10.3390/ijerph18052733.
[29] L.M. Christian, A. Franco, R. Glaser, J.D. Iams, Depressive symptoms are associated with elevated serum proinflammatory cytokines among pregnant women, Brain Behav. Immun. 23 (6) (2009) 750–754, https://doi.org/10.1016/j.bbi.2009.02.012.
[30] M.E. Coussons-Read, M.L. Okun, C.D. Nettles, Psychosocial stress increases inflammatory markers and alters cytokine production across pregnancy, Brain Behav. Immun. 21 (3) (2007) 343–350, https://doi.org/10.1016/j.bbi.2006.08.006.
[31] W. Yin, J.P. Godbout, J.F. Sheridan, Interleukin-1 beta in psychosocial stress, in: Stress: Immunology and Inflammation, Elsevier, 2024, pp. 53–63, https://doi.org/10.1016/B978-0-12-817558-3.00007-X.
[32] T. Kuo, A. McQueen, T.-C. Chen, J.-C. Wang, Regulation of glucose homeostasis by glucocorticoids, Adv. Exp. Med. Biol. 872 (2015) 99–126, https://doi.org/10.1007/978-1-4939-2895-8_5.
[33] A. Levine, O. Zagoory-Sharon, R. Feldman, J.G. Lewis, A. Weller, Measuring cortisol in human psychobiological studies, Physiol. Behav. 90 (1) (2007) 43–53, https://doi.org/10.1016/j.physbeh.2006.08.025.
[34] L.S. Bleker, T.J. Roseboom, T.G. Vrijkotte, R.M. Reynolds, S.R. de Rooij, Determinants of cortisol during pregnancy - the ABCD cohort, Psychoneuroendocrinology 83 (2017) 172–181, https://doi.org/10.1016/j.psyneuen.2017.05.026.
[35] B.R. Dahlerup, E.L. Egsmose, V. Siersma, E.L. Mortensen, M. Hedegaard, L. E. Knudsen, L. Mathiesen, Maternal stress and placental function, a study using questionnaires and biomarkers at birth, PLoS One 13 (11) (2018) e0207184, https://doi.org/10.1371/journal.pone.0207184.
[36] M. Aufdenblatten, M. Baumann, L. Raio, B. Dick, B.M. Frey, H. Schneider, M. G. Mohaupt, Prematurity is related to high placental cortisol in preeclampsia, Pediatr. Res. 65 (2) (2009) 198–202, https://doi.org/10.1203/PDR.0b013e31818d6c24.
[37] M. Babaei, S. Rezaei, S. Saghafi Khadem, I. Shirinbak, S. Basir Shabestari, The role of salivary C-reactive protein in systemic and oral disorders: a systematic review, Med. J. Islam. Repub. Iran 36 (2022) 138, https://doi.org/10.47176/mjiri.36.138.
[38] Y.Z. Szabo, D.C. Slavish, Measuring salivary markers of inflammation in health research: a review of methodological considerations and best practices, Psychoneuroendocrinology 124 (2021) 105069, https://doi.org/10.1016/j.psyneuen.2020.105069.
[39] S. Chojnowska, I. Ptaszynska-Sarosiek, A. Kepka, M. Knas, N. Waszkiewicz, Salivary biomarkers of stress, anxiety and depression, J. Clin. Med. 10 (3) (2021), https://doi.org/10.3390/jcm10030517.
[40] T. Diesch, C. Filippi, N. Fritschi, A. Filippi, N. Ritz, Cytokines in saliva as biomarkers of oral and systemic oncological or infectious diseases: a systematic review, Cytokine 143 (2021) 155506, https://doi.org/10.1016/j.cyto.2021.155506.
[41] M.H. Choi, Clinical and technical aspects in free cortisol measurement, Endocrinology and Metabolism (Seoul, Korea) 37 (4) (2022) 599–607, https://doi.org/10.3803/EnM.2022.1549.
[42] R.A. Caparros-Gonzalez, B. Romero-Gonzalez, H. Strivens-Vilchez, R. Gonzalez-Perez, O. Martinez-Augustin, M.I. Peralta-Ramirez, Hair cortisol levels, psychological stress and psychopathological symptoms as predictors of postpartum depression, PLoS One 12 (8) (2017) e0182817, https://doi.org/10.1371/journal.pone.0182817.
[43] R. Koerner, S. Prescott, D. McSkimming, A. Alman, A. Duffy, M. Groer, The salivary microbiome during pregnancy: associations with clinical and sociodemographic characteristics, J. Perinat. Neonatal Nurs. 37 (4) (2023) 287–294, https://doi.org/10.1097/JPN.0000000000000715.
[44] J. Khil, S. Picardo, C.H. Seow, Y. Leung, A. Metcalfe, E.E. Afshar, N. Sharifi, T. Campbell, N. Letourneau, D. Dewey, G.F. Giesbrecht, A.P.S. Team, Physiological and psychological stress in pregnant women with quiescent inflammatory bowel disease: a pilot study using salivary biomarkers, JGH Open 4 (4) (2020) 692–697, https://doi.org/10.1002/jgh3.12317.
[45] E. Russell, G. Koren, M. Rieder, S. Van Uum, Hair cortisol as a biological marker of chronic stress: current status, future directions and unanswered questions, Psychoneuroendocrinology 37 (5) (2012) 589–601, https://doi.org/10.1016/j.psyneuen.2011.09.009.
[46] J.L. Cox, J.M. Holden, R. Sagovsky, Detection of postnatal depression. Development of the 10-item Edinburgh postnatal depression scale, Br. J. Psychiatry 150 (1987) 782–786, https://doi.org/10.1192/bjp.150.6.782.
[47] B. Levis, Z. Negeri, Y. Sun, A. Benedetti, B.D. Thombs, Accuracy of the Edinburgh Postnatal Depression Scale (EPDS) for screening to detect major depression among pregnant and postpartum women: systematic review and meta-analysis of individual participant data, BMJ 371 (2020) m4022, https://doi.org/10.1136/bmj.m4022.
[48] S. Cohen, T. Kamarck, R. Mermelstein, A global measure of perceived stress, J. Health Soc. Behav. 24 (4) (1983) 385–396.
[49] C.M. Bann, C.B. Parker, W.A. Grobman, M. Willinger, H.N. Simhan, D.A. Wing, D. M. Haas, R.M. Silver, S. Parry, G.R. Saade, R.J. Wapner, M.A. Elovitz, E.S. Miller, U.M. Reddy, Psychometric properties of stress and anxiety measures among nulliparous women, J. Psychosom. Obstet. Gynaecol. 38 (1) (2017) 53–62, https://doi.org/10.1080/0167482x.2016.1252910.
[50] H.J. Motulsky, R.E. Brown, Detecting outliers when fitting data with nonlinear regression - a new method based on robust nonlinear regression and the false discovery rate, BMC Bioinf. 7 (2006) 123, https://doi.org/10.1186/1471-2105-7-123.
[51] A.C. Bathke, O. Schabenberger, R.D. Tobias, L.V. Madden, Greenhouse–geisser adjustment and the ANOVA-type statistic: cousins or twins? Am. Statistician 63 (3) (2009) 239–246, https://doi.org/10.1198/tast.2009.08187.
[52] K. Bowers, L. Ding, S. Gregory, K. Yolton, H. Ji, J. Meyer, R.T. Ammerman, J. Van Ginkel, A. Folger, Maternal distress and hair cortisol in pregnancy among women with elevated adverse childhood experiences, Psychoneuroendocrinology 95 (2018) 145–148, https://doi.org/10.1016/j.psyneuen.2018.05.024.
[53] L.M. Christian, Effects of stress and depression on inflammatory immune parameters in pregnancy, Am. J. Obstet. Gynecol. 211 (3) (2014) 275–277, https://doi.org/10.1016/j.ajog.2014.06.042.
[54] J.M. Keane, A.S. Khashan, F.P. McCarthy, L.C. Kenny, J.M. Collins, S. O'Donovan, J. Brown, J.F. Cryan, T.G. Dinan, G. Clarke, S.M. O'Mahony, Identifying a biological signature of prenatal maternal stress, JCI Insight 6 (2) (2021), https://doi.org/10.1172/jci.insight.143007.
[55] M.Y. Kim, G.U. Kim, H.K. Son, Hair cortisol concentrations as a biological marker of maternal prenatal stress: a systematic review, Int. J. Environ. Res. Publ. Health 17 (11) (2020), https://doi.org/10.3390/ijerph17114002.
[56] M. Lahti-Pulkkinen, P. Girchenko, R. Robinson, S.M. Lehto, E. Toffol, K. Heinonen, R.M. Reynolds, E. Kajantie, H. Laivuori, P.M. Villa, E. Hamalainen, J. Lahti, K. Raikkonen, Maternal depression and inflammation during pregnancy, Psychol. Med. 50 (11) (2020) 1839–1851, https://doi.org/10.1017/S0033291719001909.
[57] G.R. Rinne, J. Hartstein, C.M. Guardino, C. Dunkel Schetter, Stress before conception and during pregnancy and maternal cortisol during pregnancy: a scoping review, Psychoneuroendocrinology 153 (2023) 106115, https://doi.org/10.1016/j.psyneuen.2023.106115.
[58] S. Ajinkya, P.R. Jadhav, N.N. Srivastava, Depression during pregnancy: prevalence and obstetric risk factors among pregnant women attending a tertiary care hospital in Navi Mumbai, Ind. Psychiatr. J. 22 (1) (2013) 37–40, https://doi.org/10.4103/0972-6748.123615.
[59] C.-L. Dennis, K. Falah-Hassani, R. Shiri, Prevalence of antenatal and postnatal anxiety: systematic review and meta-analysis, Br. J. Psychiatr. 210 (5) (2017) 315–323, https://doi.org/10.1192/bjp.bp.116.187179.
[60] K.L. D'Anna-Hernandez, R.G. Ross, C.L. Natvig, M.L. Laudenslager, Hair cortisol levels as a retrospective marker of hypothalamic-pituitary axis activity throughout pregnancy: comparison to salivary cortisol, Physiol. Behav. 104 (2) (2011) 348–353, https://doi.org/10.1016/j.physbeh.2011.02.041.
[61] G. Mastorakos, I. Ilias, Maternal and fetal hypothalamic-pituitary-adrenal axes during pregnancy and postpartum, Ann. N. Y. Acad. Sci. 997 (2003) 136–149, https://doi.org/10.1196/annals.1290.016.
[62] C.A. Sandman, L. Glynn, C.D. Schetter, P. Wadhwa, T. Garite, A. Chicz-DeMet, C. Hobel, Elevated maternal cortisol early in pregnancy predicts third trimester levels of placental corticotropin releasing hormone (CRH): priming the placental clock, Peptides 27 (6) (2006) 1457–1463, https://doi.org/10.1016/j.peptides.2005.10.002.
[63] J.L. Irwin, A.L. Meyering, G. Peterson, L.M. Glynn, C.A. Sandman, L.M. Hicks, E. P. Davis, Maternal prenatal cortisol programs the infant hypothalamic-pituitary-adrenal axis, Psychoneuroendocrinology 125 (2021) 105106, https://doi.org/10.1016/j.psyneuen.2020.105106.
[64] J.C. Owen, S.P. Garrick, B.M. Peterson, P.J. Berger, M.F. Nold, A. Sehgal, C. A. Nold-Petry, The role of interleukin-1 in perinatal inflammation and its impact on transitional circulation, Frontiers in Pediatrics 11 (2023) 1130013, https://doi.org/10.3389/fped.2023.1130013.
[65] Y.J. Heng, S. Liong, M. Permezel, G.E. Rice, M.K.W. Di Quinzio, H.M. Georgiou, The interplay of the interleukin 1 system in pregnancy and labor, Reprod. Sci. 21 (1) (2014) 122–130, https://doi.org/10.1177/1933719113492204.
[66] Q. Sha, Z. Madaj, S. Keaton, M.L. Escobar Galvis, L. Smart, S. Krzyzanowski, A. T. Fazleabas, R. Leach, T.T. Postolache, E.D. Achtyes, L. Brundin, Cytokines and tryptophan metabolites can predict depressive symptoms in pregnancy, Transl. Psychiatry 12 (1) (2022) 35, https://doi.org/10.1038/s41398-022-01801-8.
[67] D.C. Slavish, J.E. Graham-Engeland, J.M. Smyth, C.G. Engeland, Salivary markers of inflammation in response to acute stress, Brain Behav. Immun. 44 (2015) 253–269, https://doi.org/10.1016/j.bbi.2014.08.008.
[68] R. Hou, G. Ye, Y. Liu, X. Chen, M. Pan, F. Zhu, J. Fu, T. Fu, Q. Liu, Z. Gao, D. S. Baldwin, Z. Tang, Effects of SSRIs on peripheral inflammatory cytokines in patients with Generalized Anxiety Disorder, Brain Behav. Immun. 81 (2019) 105–110, https://doi.org/10.1016/j.bbi.2019.06.001.
[69] A.L. Jefferies, Selective serotonin reuptake inhibitors in pregnancy and infant outcomes, Paediatr. Child Health 16 (9) (2011).
[70] C. Omere, L. Richardson, G.R. Saade, E.A. Bonney, T. Kechichian, R. Menon, Interleukin (IL)-6: a friend or foe of pregnancy and parturition? Evidence from

functional studies in fetal membrane cells, Front. Physiol. 11 (2020) 891, https://doi.org/10.3389/fphys.2020.00891.
[71] J.M. Rasmussen, A.M. Graham, S. Entringer, J.H. Gilmore, M. Styner, D.A. Fair, C. Buss, Maternal Interleukin-6 concentration during pregnancy is associated with variation in frontolimbic white matter and cognitive development in early life, Neuroimage 185 (2019) 825–835, https://doi.org/10.1016/j.neuroimage.2018.04.020.
[72] M. Ravi, B. Bernabe, V. Michopoulos, Stress-related mental health disorders and inflammation in pregnancy: the current landscape and the need for further investigation, Front. Psychiatr. 13 (2022) 868936, https://doi.org/10.3389/fpsyt.2022.868936.
[73] M. Del Giudice, S.W. Gangestad, Rethinking IL-6 and CRP: why they are more than inflammatory biomarkers, and why it matters, Brain Behav. Immun. 70 (2018) 61–75, https://doi.org/10.1016/j.bbi.2018.02.013.
[74] K.J. O'Donnell, V. Glover, E.D. Barker, T.G. O'Connor, The persisting effect of maternal mood in pregnancy on childhood psychopathology, Dev. Psychopathol. 26 (2) (2014) 393–403, https://doi.org/10.1017/S0954579414000029.