

*Full Length Research Paper*

# **Salivary cortisol, postpartum psychological status and bonding attachment**

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**While several studies have investigated how stress-related situations affect cortisol levels, few studies have examined the relationship between cortisol levels and psychological measures related to postpartum psychological status. This study investigated the relationship between salivary cortisol and postpartum psychological status and bonding attachment. This study is a cross-sectional study. Participants were recruited at the 1-month postpartum checkup and included 70 consenting mothers at 1 month postpartum. Salivary cortisol levels, profile of mood states (POMS), general health questionnaire (GHQ), maternal anxiety screening scale (MASS), and mother-infant bonding scale (MIBS) were measured for association. Salivary cortisol levels were significantly associated with scores on the GHQ; depression dimension of POMS; childbirth satisfaction, child-rearing ease, confident, and lack of confidence subscales of MASS; and MIBS ( $r=.369$   $P=.002$ ). There were particularly strong associations with GHQ ( $r=.427$   $P=.000$ ), childbirth satisfaction subscale of MASS ( $r=-.412$   $P=.000$ ), and MIBS ( $r=.369$   $P=.002$ ) scores. The measurement of maternal salivary cortisol at one month postpartum could be a method to screen mothers for postpartum depression. However, it should be taken into account that salivary cortisol levels may reflect factors other than psychological state, and further investigation of related factors is necessary.**

**Key words:** Postpartum depression, stress, anxiety, cortisol.

## **INTRODUCTION**

The declining birthrate, aging population, and nuclear family trend in Japan have created serious social problems, such as postpartum mothers' childcare anxiety, increasing postpartum depression (Igura et al., 2018), and mothers' abuse of their children. As a countermeasure, several attempts have been made to provide support, such as postpartum hospitalization at hospitals and midwifery clinics, home visits at local health centers, and outpatient childcare counseling at hospitals, but they have yet to spread sufficiently to solve the

problem. Another necessity is to identify mothers who need special support or urgent help and take action as early as possible. Although there are methods such as questionnaires for ascertaining the psychological state, most of the tests are conducted at postpartum examinations and infant health examinations. Questionnaire-based surveys require substantial time for the mother to answer the questions, and the data obtained from the questionnaire survey may be greatly influenced by the mother's subjectivity. Therefore, it is

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necessary to incorporate objective methods of testing. Previous studies have suggested an association between salivary cortisol levels in perinatal women and social stress (Deligiannidis et al., 2016), post-traumatic stress (Van den Heuvel et al., 2019), and isolated environment (Meng et al., 2020). In general, stress is perceived by the cerebral cortex first, and then transmitted to the hypothalamus. It is known that the two pathways are hypothalamus-pituitary-adrenal cortex-cortisol secretion and cerebral cortex-hypothalamus-sympathetic nervous system-cortisol secretion. It is known that catecholamine and cortisol are secreted by stress load, and studies have been conducted to measure hormones such as cortisol and catecholamine in blood as stress indicators. However, in order to use these hormones as stress indicators, it is necessary to collect samples in a noninvasive way. For this reason, research has recently been conducted on salivary substances as stress indicators. Salivary cortisol concentrations are highly correlated with blood cortisol concentrations, and diurnal variations show a similar pattern (Yamanaka, 2005). It has also been reported that the time lag between blood and cortisol concentrations is only a few min and is not affected by salivary exudation rate (Oda et al., 2000), and many studies have been conducted using salivary cortisol concentration as a stress indicator. Thus, basic research has been conducted to measure cortisol in saliva samples. Van (1996) and Wolff et al (2006) found that cortisol level is a valid assessment of mental stress and physical stress, as reflected in Trait Anxiety on the State-Trait Anxiety Inventory (STAI). In addition, background factors have also been found to affect salivary cortisol levels, smoking (Jack et al., 2002), education (Maureen, 2005), breastfeeding status (Tu et al., 2006), income (Scheyer and Urizar 2015), including age, body mass index (Straub et al., 2016), Socioeconomic Risk (Braren et al., 2019) and fasted (Bentele et al., 2021). While there have been many reports on how stress-related situations affect cortisol levels, few studies have examined the relationship between cortisol levels and psychological measures that have been shown to be related to postpartum psychological status. Adam et al. (2017) conducted a literature review on the association between physical and mental health and cortisol and found that the association is clear. Seth et al. (2016) also conducted a literature review on the association between depression and cortisol and provided clarification on the association. Scales that capture psychological states related to parenting stress and parenting anxiety in postpartum mothers include the General Health Questionnaire (GHQ; Chiba et al., 2020), the Mood State Profile (POMS; Kojima et al., 2019), and the State-Trait Anxiety Scale (STAI; Van, 1996; Wolff et al., 2006). In the present study, we examined the relationship between salivary cortisol levels and scores on the POMS and GHQ to clarify the relationship between salivary cortisol levels and mental and physical health and depression, which have not been previously suggested to be related to cortisol levels and

their effects on the psychological state of postpartum mothers. In addition, we examined the relationship between the maternal anxiety screening scale (MASS; Yoshida et al., 1999), which measures parenting anxiety, and the mother-infant bonding scale (MIS), which assesses feelings related to attachment formation with the child and is believed to be significantly involved in the psychological state of postpartum mothers. The relationship with MIBS; Yoshida et al., 2012) was also examined.

## METHODOLOGY

### Research Design

This study is a cross-sectional study.

### Subjects

Seventy mothers in the first month postpartum who delivered at the obstetrics department of a general hospital in T city, a prefecture adjacent to Tokyo, were included in the study. In consideration of the effect on biochemical indices, mothers with a smoking habit or chronic diseases accompanied by oral medication were excluded from the study. To obtain data from as standard a population as possible, mothers with a normal pregnancy, delivery, and postpartum period were selected in order to minimize the effects of physical stress caused by pregnancy and delivery and psychological stress factors. Mothers who had delivered by cesarean section were excluded from the study because they were considered to be in great physical distress.

### Recruitment and data collection

Participants were recruited at the 1-month postpartum checkup and included 70 consenting mothers at 1 month postpartum. We selected the subjects from the medical records of mothers who came to the hospital for a one-month postpartum checkup, and gave them a full explanation of the significance, purpose, and methods of the study, as well as their cooperation in the study. In addition, mothers were given an explanatory sheet containing the information and asked to sign a consent form if they agreed to participate in the study. After obtaining consent from the subjects, saliva was first collected to measure salivary cortisol levels, and they were asked to complete the Profile of Mood States (POMS), General Health Questionnaire (GHQ), Maternal Anxiety Screening Scale (MASS), and Mother-Infant Bonding Scale (MIBS) questionnaire.

### Psychological scales

#### *Profile of mood states*

The POMS is a standardized questionnaire developed by McNair et al. (1971) and translated into Japanese by Yokoyama et al. (1990) that has ensured reliability and validity. The content of the questionnaire does not measure personality tendencies, but rather the temporary mood and emotional state of the person under the conditions in which he or she is placed. A total of 65 items are rated on a 5-point scale to indicate the mood of the past week, across the following dimensions: T-A (Tension – Anxiety), D (Depression), A-H (Anger - Hostility), V (Vigor), F (Fatigue), and C (Confusion). The

higher the score, the stronger the emotion.

### **General health questionnaire**

The GHQ is a questionnaire-based screening test developed by Goldberg (1979) to identify, evaluate, and detect symptoms in neurotic patients. The Japanese version of the questionnaire has been rehabilitated and validated by Nakagawa (1985). The main content of the GHQ involves questions about whether healthy mental functioning can be sustained or whether there are new facts that distress the patient or subject. To reduce the burden on the subject as much as possible, the short version of the scale (GHQ30) was used in this study, which has 30 items across four factors: physical symptoms, anxiety and insomnia, social inactivity, and depressive tendencies. Scores range from 0–30, with a higher score indicating a more neurotic state. The classification (critical) score is 6/7.

### **Maternal anxiety screening scale**

This scale was developed by Yoshida et al. (1999) and its validity and reliability have been ensured in Japan. The scale consists of six subscales: childbirth satisfaction, husband's support, childcare anxiety, child-rearing ease, confident, and lack of confidence.

### **Mother-infant bonding scale**

Kumar (1997) stated that mothers with depressive symptoms were more likely to have severe bonding disorders and to be at risk for abuse and neglect. Suzumiya (2003) reported that postpartum depression was associated with impaired attachment formation on the part of mothers during infancy, or bonding disorders. In this study, we aimed to measure mothers' feelings toward their children to understand their psychological state after childbirth. MIBS, originally written by Marks and translated by Yoshida et al. (2012) was used in this study because the clinical usefulness and reliability, validity of the Japanese version have been examined. The questionnaire consists of 10 questions with scores of 0, 1, 2, and 3 for each question. The highest score is 30 points, and higher scores indicate stronger negative feelings toward the child.

### **Salivary cortisol**

#### **Sample collection**

Saliva was collected according to the method of Nakata et al. (2000). After rinsing the subject's mouth with distilled water, the subject rested for 5 min, swallowed the spilled saliva, and then used a saliva collection kit (salivette cotton, Funakoshi Co., Ltd.) to place the cotton in the kit under the tongue for 2 min, and then returned it to the tube to collect approximately 1.5 to 2.0 ml. Eating, drinking and brushing were prohibited for 30 min prior to saliva collection, and lipstick was avoided during collection. Considering the diurnal variation, we avoided collecting saliva samples in the early morning or at night, when the variation is high, and collected saliva samples between 10:00 and 14:00. Saliva samples were immediately transported to the laboratory in a refrigerated box maintained at 4°C after collection and stored at -30°C until analysis.

#### **Sample analysis**

At the time of analysis, the saliva was thawed, centrifuged at 8000 rpm for 20 min using a centrifuge (TOMY SRX-201), and the

supernatant was collected by pipette and quantified by competitive Enzyme Immunoassay (EIA) and cortisol Enzyme Immunoassay Kit "Assay Designs." The MTP-300 microplatereader (CORONA) was used for the assay. The lower limit of measurement was 105 pg/ml, the upper limit was 10000 pg/ml, and the coefficient of variation was 1.64–3.84%.

### **Statistical analysis**

Statistical analysis was performed using SPSS. ver27 for Windows. The relationship between salivary cortisol concentration and scores on the GHQ, POMS, MASS, and MIBS was analyzed by Pearson correlation coefficient. In addition, to analyze the influence factors of salivary cortisol, multiple regression analysis was conducted with salivary cortisol level as the dependent variable after deleting items that showed multicollinearity.

### **Ethical considerations**

A research plan was submitted to and approved by the Ethics Committee of A School, B University (Approval No. XXX) before starting this study.

## **RESULTS**

### **Subject background and measurement data**

Seventy postpartum mothers were included in this study (Table 1). The mean age was  $32.6 \pm 4.53$  years, all were married, 55 (78.5%) had nuclear families, and 47 (67.1%) were housewives. The mean delivery time was  $562.4 \pm 448.0$  hs, hemorrhage at delivery was  $369.0 \pm 244.7$  g, and child's birth weight was  $2993.5 \pm 353.1$  g. The mean values and ranges of salivary cortisol levels and scores on each scale are shown in Table 2.

### **Relationship between salivary cortisol levels and each scale**

Table 3 shows the results of the Pearson correlation coefficient analysis of the relationship between salivary cortisol levels and each scale. A significant association was found between salivary cortisol level ( $r = -0.028$   $P = 0.000$ ) and GHQ ( $r = 0.427$   $P = 0.000$ ); D of POMS ( $r = 0.263$   $P = 0.028$ ); childbirth satisfaction ( $r = -0.412$   $P = 0.000$ ), child-rearing ease ( $r = -0.412$   $P = 0.000$ ), confident ( $r = -0.338$   $P = 0.004$ ), and lack of confidence ( $r = 0.243$   $P = 0.043$ ) on the MASS; and the MIBS ( $r = 0.369$   $P = 0.002$ ).

### **Factors affecting salivary cortisol levels**

A multiple regression analysis (forced imputation method) was conducted with salivary cortisol level as the dependent variable and each scale as the independent variable (Table 4). D of the POMS showed a strong

**Table 1.** Characteristics of the participants (N=70).

Variable	Mean±SD	Min-Max
Age (year) mean±SD	32.6±4.53	[23-43]
Marital status (has a partner)	70.0(100.0)	
<b>Family form</b>		
Nuclear family	55.0(78.5)	
Extended family	15.0(21.5)	
<b>Occupation</b>		
Homemaker	47.0(67.1)	
Full-time employment	21.0(30.0)	
Part-time job	2.0(2.9)	
Delivery time (minute) mean±SD	562.4±448.0	[87-2370]
Bleeding during childbirth (ml) (mean±SD)	369.0±244.7	[84-1395]
Birth weight (g)	2993.5±353.1	[2105-3865]

Age, delivery time, bleeding during childbirth, and birth weight are shown as mean and standard deviation (mean±SD), delivery time, bleeding during childbirth, birth weight are shown as (%), range.

**Table 2.** Variable data (N=70).

Variable	Mean±SD	Min-Max
Salivary cortisol level	371.0±197.7	[105.0-760.0]
GHQ	4.3±3.7	[0-15]
<b>POMS</b>		
T-A	6.8±5.6	[0-24]
D	4.2±6.0	[0-28]
A-H	5.3±6.3	[0-29]
V	12.0±5.8	[0-27]
F	7.4±5.5	[0-20]
C	5.8±3.8	[0-16]
<b>Maternal anxiety screening scale</b>		
Childbirth satisfaction	28.2±4.0	[12-32]
Husband's support	21.7±5.9	[4-28]
Childcare anxiety	17.3±4.9	[10-34]
Child-rearing ease	14.9±3.3	[6-20]
Confidant	10.7±2.1	[3-12]
Lack of confidence	9.5±3.0	[5-20]
Total score	26.9±7.2	[16-50]
Mother-infant bonding scale	1.9±2.4	[0-8]

Mean and standard deviation (mean±SD).

correlation with T-A ( $r=0.808$   $P=0.000$ ) and A-H ( $r=0.831$   $P=0.000$ ), and the total score on the MASS. In addition, the MASS total score was strongly correlated with childcare anxiety ( $r=0.943$   $P=0.000$ ) and lack of confidence ( $r=0.844$   $P=0.000$ ), and therefore, considering

multicollinearity, the POMS T-A and A-H and the MASS total score were excluded from the variables in the analysis. The results showed that only the GHQ score was affected ( $P=.039$ , 95% confidence interval: .961-35.749).

**Table 3.** Correlation between cortisol levels and GHQ, POMS, maternal anxiety screening scale, and mother-infant bonding scale (N=70).

Variable	Salivary cortisol level	
	r	p-value
GHQ	0.427**	0.000
<b>POMS</b>		
T-A	0.121	0.321
D	0.263*	0.028
A-H	0.219	0.068
V	-0.087	0.472
F	0.113	0.350
C	0.153	0.207
<b>Maternal anxiety screening scale</b>		
Childbirth satisfaction	-0.412**	0.000
Husband's support	-0.105	0.387
Childcare anxiety	0.189	0.117
Child-rearing ease	-0.242*	0.043
Confidant	-0.338**	0.004
Lack of confidence	0.243*	0.043
Total score	0.232	0.054
Mother-Infant Bonding Scale	0.369**	0.002

Spearman's rank correlation coefficient \*\*P&lt;0.01 \*P&lt;0.05.

**Table 4.** Factors affecting salivary cortisol(N=70)..

Variable	$\beta$	p-value	95% confidence interval	
			d - dw	d + dw
GHQ			0.0961	35.749
<b>POMS</b>				
D	0.249	0.214	-4.821	21.102
V	0.198	0.143	-2.335	15.735
F	-0.296	0.117	-23.737	2.701
C	-0.031	0.882	-22.910	19.737
<b>Maternal anxiety screening scale</b>				
Childbirth satisfaction	-0.213	0.198	-26.281	5.581
Husband's support	0.131	0.320	-4.357	13.113
Childcare anxiety	-0.135	0.464	-20.090	9.276
Child-rearing ease	0.024	0.861	-14.497	17.293
Confidant	-0.280	0.440	-51.787	-0.67
Lack of confidence	-0.161	0.321	-31.089	10.36
Mother-infant bonding scale	0.331	0.052	-0.215	53.938

Multiple regression analysis using forced entry method/Variables of TA, AH, and Maternal Anxiety Screening Scale (total score) were excluded due to multicollinearity. Object variable: salivary cortisol level.

## DISCUSSION

### Subject characteristics

The average age of the mothers in this study was 32.6

years ( $\pm 4.53$  years), and there were 70 subjects in total. According to birth order in FY2018, the mother's age was 30.7 years for the first child and 32.6 years for the second child (Vital Statistics, Ministry of Health, Labor and Welfare, 2018); thus, the age of the subjects in this study

is estimated to be slightly higher, but it is not considered as a significant difference from the population. The number of weeks of delivery was calculated for all subjects. The number of weeks of delivery was normal in all cases, the time required for delivery and the amount of blood loss during delivery were within the normal range, although they varied from case to case, and the weight of the baby was also normal in all cases. A total of 78% of the subjects had nuclear families, and 67% were housewives. According to the 2019 census, the number of nuclear family households with children aged under six years is increasing year by year in Japan, 59.8% of all households are nuclear families, and 47.5% are nuclear families consisting of a couple and unmarried children (national population census on 2019). It can be said that the subjects of this study have a high percentage of nuclear families. The employment rate of mothers with 0-year-old children is 49.8% (Ministry of Health, Labour and Welfare, National Survey of Family Life 2019), indicating that the mothers in this study were a group with a somewhat low employment rate.

### **Psychological state as reflected by psychological scales**

The mean POMS scores were T-A  $6.8 \pm 5.6$ , D  $4.2 \pm 6.0$ , A-H  $5.3 \pm 6.3$ , V  $12.0 \pm 5.8$ , F  $7.4 \pm 5.5$ , and C  $5.8 \pm 3.8$  on the POMS. Matsuoka and Sasaki (2000) reported that mothers' POMS scores during the first 3 to 10 days after childbirth were T-A  $11.1 \pm 7.13$ , D  $4.6 \pm 7.30$ , A-H  $2.0 \pm 3.80$ , V  $14.2 \pm 7.16$ , F  $6.3 \pm 4.26$ , and C  $4.3 \pm 3.82$ . In the postpartum period, anger and hostility tended to be lower and liveliness tended to be higher. Although there are no previous reports on POMS scores in the first postpartum month, and it is difficult to compare them because of the different measurement periods, the subjects in this study had POMS scores similar to those in previous studies. The GHQ score was  $4.3 \pm 3.7$ , which was slightly higher than the GHQ score of  $4.0 \pm 3.5$  reported by Kanaoka (2011), but similar to the GHQ score of mothers with infants. On the MASS, the scores were  $28.2 \pm 4.0$  for childbirth satisfaction,  $21.7 \pm 5.9$  for husband's support,  $17.3 \pm 4.9$  for childcare anxiety,  $14.9 \pm 3.3$  for child-rearing ease,  $10.7 \pm 2.1$  for confidant, and  $9.5 \pm 3.0$  for lack of confidence. The total score on the MASS was  $26.9 \pm 7.2$ , indicating a similar degree of child-rearing anxiety as that in the study by Miyaoka et al. (2015), who obtained a total score of  $27.3 \pm 7.9$  in mothers at one month postpartum. These previous studies indicate that negative feelings toward babies tend to be lower among the subjects in this study.

### **Psychological state as reflected by salivary cortisol**

Salivary cortisol level was associated with scores on the

GHQ; D of POMS; childbirth satisfaction, child-rearing ease, confidant, and lack of confidence on the MASS; and the MIBS ( $r=0.369$   $P=0.002$ ). Infant Bonding Scale ( $r=0.369$   $P=0.002$ ). There were particularly strong associations with the GHQ ( $r=0.427$   $P=0.000$ ), childbirth satisfaction subscale of the MASS ( $r=-0.412$   $P=0.000$ ), and MIBS ( $r=0.369$   $P=0.002$ ) scores. The results suggest that salivary cortisol is a good indicator of psychological state, as measured by the GHQ. In addition, among the factors of child-rearing anxiety, there was a rather strong correlation with the childbirth satisfaction subscale, suggesting that mothers who feel that their children are difficult to raise need more careful follow-up, and that salivary cortisol levels may be utilized as a tool to understand mothers who need such support and their psychological state. Furthermore, salivary cortisol levels could be used to identify mothers with more negative feelings toward their children, or in other words, mothers who are having trouble in establishing attachment, and that this could be used for early coping support and care. In the multiple regression analysis, the only factor that affected salivary cortisol was the GHQ score. Mihara et al (2019) found that GHQ was associated with salivary cortisol in college students. In the present study, we can say that similar results were obtained in postpartum mothers. The use of salivary cortisol, which is strongly related to GHQ, an index of mental health, may allow us to easily understand the mental state of mothers.

### **Issues in application of salivary cortisol level to screen for postpartum depression**

This study reveals the possibility that measurement of salivary cortisol can be used as a tool to understand the psychological state of mothers in the first month postpartum. However, it should be taken into account that salivary cortisol levels also reflect factors other than psychological state. First, postpartum women are in a period of high physical stress due to childbirth (posterior labor pain, breast pain, perineal wound pain, etc.). In addition, Tsukahara et al. (2006) reported a significant decrease in Visual Analog Scale scores in the period up to two days postpartum and after four days postpartum after stitches were removed. She also stated that the REEDA score (a five-point scale indicating redness, edema, subcutaneous hemorrhage, secretions, and healing status) decreased significantly from one to four days postpartum after suture removal. However, Nakazawa et al. (2006) stated that 58.5% of mothers in the first week after childbirth and 55.3% in the first month after childbirth had low back pain, and most of them had continuous pain from pregnancy. It is clear that postpartum back pain is a continuous physical pain from the pregnancy period, and that it is a long-term physical pain after childbirth, with no significant reduction during the early postpartum period to one month. Nakayama

(2007) also reported that 83% of mothers complained of at least one of the following pains in the first four days postpartum: back pain, perineal pain, pubic pain, and hemorrhoidal pain. From these reports, it is clear that the majority of mothers have some kind of pain symptoms in the early postpartum period due to the effects of childbirth. In addition, while pain that has persisted since pregnancy, such as back pain, remains for a long time after childbirth, pain that has been noticeable since immediately after childbirth, such as perineal pain, pubalgia, and hemorrhoidal pain, is relieved in the early postpartum period. This suggests that the salivary cortisol level in the early postpartum period does not directly reflect psychological state. However, Koyasu and Matsui (2018) reported that the physical distress of mothers during the postpartum period is associated with the EPDS; therefore, mothers who have persistent back and breast pain and feel distress even after the early postpartum period may develop postpartum depression triggered by their physical distress. Cortisol levels have also been found to reflect sleep status. However, although there are no reports on the relationship between sleep and cortisol levels in mothers during the puerperium period and using saliva samples, Inoue et al. (2019) found that blood cortisol levels in middle school students were associated with sleep duration. Fujioka et al. (2016) reported the results of a survey of mothers in the first month after childbirth regarding their primary sleep time (the time they slept the longest), total sleep time, and number of sleeps. They found that during the puerperium, mothers were not able to get enough sleep, even for short periods of time, and that there was a relationship between primary sleep duration and EPDS. This indicates that many mothers during the postpartum period do not get enough sleep, and that individual differences in sleep status are also associated with postpartum depression. In conclusion, salivary cortisol levels, which reflect physical distress and sleep status as well as psychological status, may contribute to the early detection of postpartum depression.

As various studies have reported factors associated with salivary cortisol levels, such as age, body mass index (Straub et al, 2016), breastfeeding status (Tu et al, 2006), income (Scheyer and Urizar 2015), education (Maureen, 2005), and smoking (Jack et al., 2002), it is necessary to consider these factors when assessing the psychological state of postpartum mothers.

## Conclusion

Maternal salivary cortisol levels in the first postpartum month were associated with scores on the GHQ; D of POMS; childbirth satisfaction, child-rearing ease, confidant, and lack of confidence on the MASS; and the MIBS. The results suggest that cortisol levels are associated with mental health, depression, childbirth satisfaction, child-rearing ease, confidant, lack of

confidence, and attachment with the infant. In addition, salivary cortisol levels were found to be influenced by the psychological state captured by the GHQ, suggesting that measurement of salivary cortisol may be a method to screen for postpartum depression in mothers in the first month after childbirth.

## Limitation

This study is limited in its generalizability due to the limited study area and subjects.

## CONFLICT OF INTERESTS

The authors have not declared any conflict of interest.

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