

## Mental and Physical Stress of Pregnant Women and Work

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<https://doi.org/10.15017/21736>

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出版情報 : 九州大学, 2011, 博士 (看護学), 課程博士  
バージョン :  
権利関係 :

## Title Page

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Mental and Physical Stress of Pregnant Women and Work

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Key Words: Pregnancy, Working, Stress

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## Abstract

This study aimed to investigate the effects of working during gestation on pregnant women by evaluating their mental and physical stress. The subjects were 62 women during natural pregnancy with a singleton who were not in a state of illness. The survey was conducted by a Questionnaire to clarify their general background, daily activities, working status, mental stress (GHQ28) and measurement of antioxidant capacity (Potential Anti-Oxidant (PAO)) and oxidative stress urinary 8-hydroxydeoxy guanosine (8-OHdG) in early (12-16w) and late (32-36w) pregnancy.

Of the 62 females (primigravida:23, multigravida:39), 41 (66.1%) worked in early pregnancy and 17 (27.4 %) continued to work in late pregnancy, while the other, i.e. 24, discontinued working. Eight took maternal leave of absence and 16 resigned their jobs during the gestation period .

The total GHQ28 scores fell significantly in late pregnancy compared with early pregnancy, mainly caused by the relief of Somatic Symptoms. In addition, urinary 8-OHdG lowered significantly in late pregnancy whereas PAO rose significantly. The reduction of physical stress was recognized in late pregnancy. The urinary 8-OHdG of working pregnant women was lower than non-working pregnant women in early pregnancy. The PAO levels were not changed by working status. GHQ28, PAO and urinary 8-OHdG were not significantly related with life activities, such as going up/down, sleep, house-hold work and so on.

Mental and physical stress caused by pregnancy was high in early pregnancy and decreased as pregnancy progressed. Working during pregnancy did not adversely influence mental and physical stress of pregnant women, while working in early pregnancy reduced physical stress levels.

## Introduction

The current econo-industrial society cannot dispense with the female labor force. Females hold an important role in society by giving birth, besides industrial activities. Therefore, females in modern society are required to work even during gestation. This fact prompts the whole society to support working women by helping them give birth and raise their children, and this has become a major topic in occupational medicine.

Considering previous studies, many epidemiological studies have shown that working pregnant women are at a higher risk of coexisting illness such as threatened abortion, threatened preterm delivery and pregnancy-induced hypertension<sup>1)</sup>. According to the Labor Standards Act, pregnant women are protected from hazards of working; 1) ban on underground labor (article 64-2), 2) limitations on dangerous and injurious work (article 64-3), 3) leave work before and after childbirth (article 65), 4) avoidance of excess working hours, work on days off, and work at night if so requested by pregnant women (article 66)<sup>2)</sup>. The legal protection for working pregnant women is limited so that pregnant women are not completely taken care of. One of the reasons for this is that no risk assessment has been done as to the harmful factors involved in working during gestation, especially in terms of chemicals, newly developed jobs and so on.

Taking these facts into consideration, we studied mental and physical loads during gestation work. As a first step, this study reports analyze of the risks involved in getting pregnant and giving birth while working by measuring mental stress (The General Health Questionnaire: GHQ28) and oxidation stress markers (Urinary 8-hydroxydeoxyguanosine (8-OHdG) and Potential Anti-Oxidant (PAO)).

## Subjects and Methods

### 1. Study Subjects

Pregnant women within less than 10 weeks into their gestation periods were recruited at 5 medical facilities, including one university hospital, in K City after approving the purpose of this study and the confirmation of their fetuses' pulses. The recruitment criteria were as follows; 1) no current illness or medical treatment, 2) single pregnancy, 3) natural pregnancy.

### 2. Period of Study

Recruitment went on from January to March in 2010 with the survey at birth done by October, 2010.

### 3. Survey Contents

#### 1) Early survey in the gestation period (between the 12th and 16th week)

The survey was conducted with questionnaire sheets (age, family members, pregnancy history, current state of illness, smoker/non-smoker, drinker/non-drinker, residential environment, and breakdown of daily hours: sleep, housework, child-raising, resting time and so on) and GHQ28, as well as collection of biospecimens (4ml of blood and 3ml of urine). In addition to these, the basic items on the medical records of the subjects (their current state of illness, treatment, pregnancy history, and body mass index before pregnancy) were copied.

## 2) Late survey in the gestation period (between the 32nd and 36th week)

A Questionnaire (smoking and drinking in pregnancy, career and employment status, breakdown of daily hours: sleep, housework, child-raising, resting time) and GHQ28, as well as collection of biospecimens (4ml of blood and 3ml of urine) were conducted late in pregnancy.

## 3) Survey at birth

The survey was conducted with questionnaire sheets. In addition to this, the items on the medical records of the subjects (remarks on their pregnancy, childbirth delivery and newly-born babies) were copied.

## 4. Ethical Compliance

Informed consent to the surveys was given by the subjects after proper explanation of the purpose of this study in the early stage of pregnancy. Permission to access to their medical records was also given. Biospecimens of blood and urine were taken in combination with the ordinary collection of blood and urine at medical checkups for pregnant women.

This study was approved by Kyushu University Institutional Review Board for Clinic Research (approval number; 21-119) and Ethics committee of Medicine and Medical Care, University of Occupational and Environmental Health, Japan (accepted number; 08-91).

## 5. Measurements of Biospecimens

Blood samples were frozen in storage at -80 °C as serum within four hours after sampling. Urine samples

were frozen for storage at -80 °C after being aliquoted.

#### 1) Measurements of Urinary 8-OHdG

Urinary 8-OHdG levels were measured with an ELISA kit made by the Japan Institute for the Control of Aging. Since the samples were spot urine, the measurement values were corrected by urinary creatinine levels (8-OHdG/Cre) or urine - specific gravity (8-OHdG/mL) in order to adjust for daily fluctuations.

#### 2) Measurements of Antioxidant Capacity in Blood

Antioxidant capacity was measured by an ELISA kit for Potential Anti-Oxidant(PAO) (made by the Japan Institute for the Control of Aging). The mixture of the sample and a  $\text{Cu}^{++}$  reagent generated  $\text{Cu}^+$  due to a reduction reaction by antioxidant materials in the samples.  $\text{Cu}^+$  together with a color-producing reagent formed a compound, which absorbed light at 490 nm producing  $\text{Cu}^+$ . The amount of produced  $\text{Cu}^+$  showed the antioxidant capacity of the samples.

#### 6. Statistical Analysis

Paired t-test, multiple linear regression analysis and multiple logistic regression analysis were carried out using a statistical analysis application SPSS19.0J.

## Results

### 1. Characteristics of the Subjects and their Employment Status

Eighty-four subjects gave their consent to participation in the surveys. Twenty-two subjects were excluded from the survey due to lack of biospecimens. Finally, 62 subjects satisfied the recruiting criteria and could be followed-up by both of biospecimens and questionnaires. Of the 62 subjects, 23 were primigravida and 39 were multigravida, with no significant differences in age. Moreover, no significant differences in age or working status were observed in the early and late surveys in the gestation period. 41 subjects (66.1%) were working in early pregnancy. Of them, 17 (27.4 %) continued working in late pregnancy, while 24 stopped working during the gestation period (Table 1). Of the 24, 8 took maternal leave of absence and the rest (16 people) resigned their jobs during the gestation period (the dropout rate was 39%). The reasons for resigning were as follows; 2 complained of somatic symptoms, such as feelings of weariness and/or nausea, 4 found it difficult to cope with their working conditions, such as handling heavy objects and/or driving for a lengthy period, 6 registered reasons concerning their employment status and system, such as the termination of an employment period and the absence of maternal leave and/or a child-care leave, and 4 gave no reason. No one experienced a career change during the surveillance period.

### 2. General Health Questionnaire (GHQ) 28 in early and late pregnancy

The results of GHQ28 in early and late pregnancy are shown in Table 2. The total scores in late pregnancy were significantly lower than those in early pregnancy ( $p<0.01$ ). This could be attributed to the



relief of Somatic Symptoms as the greatest factor ( $p<0.01$ ). The scores for Anxiety and Insomnia, Social Dysfunction, and Severe Depression tended to be lower in late pregnancy.

### 3. Potential Anti-Oxidant (PAO) and Urinary 8-OHdG in early and late pregnancy

The time course of PAO levels after blood collection was analyzed (Fig 1). No decreases in PAO levels were observed for 4 hours after sampling. PAO significantly elevated in late pregnancy ( $p<0.01$ ). Urinary 8-OHdG significantly fell in late pregnancy both after creatinine correction ( $p<0.01$ ) and after specific gravity correction ( $p<0.05$ ) (Fig 2).

Urinary 8-OHdG did not elevate in either early or late pregnancy for the above-mentioned 4 women who answered in their questionnaires that they had resigned their jobs because of working conditions/ such as handling heavy objects and/ or driving for a lengthy period.

### 4. Factors Influencing the Stress-Related Conditions of Mothers

#### 1) Factors Influencing GHQ28

Using the GHQ total scores as dependent variables (6 or more with some problems and 5 or less in a normal state) and items such as working status, personal factors, and breakdowns of daily hours as independent variables, multiple logistic regression analysis was performed. As a result, no variables influencing the GHQ total scores were recognized in either early or late pregnancy.

## 2) Factors Influencing Potential Anti-Oxidant (PAO)

The indication of a logarithmic normal distribution for PAO led to a multiple linear regression analysis after a logarithmic conversion. BMI was the only variable that influenced PAO ( $p < 0.001$ ) in early pregnancy (Table 3). No influential variable was seen in late pregnancy (Table 3).

## 3) Factors Influencing Urinary 8-OHdG

According to multiple linear regression analysis (Stepwise Method), the continuation of work was only influential ( $p < 0.05$ ) in early pregnancy. Urinary 8-OHdG was higher in non-working pregnant women. As for Urinary 8-OHdG after specific gravity correction, the working state and daily sleeping time were influential variables in early pregnancy ( $p < 0.001$  respectively). Urinary 8-OHdG specific gravity was affected by age in late pregnancy. In late pregnancy, urinary 8-OHdG was lower for older subjects ( $p < 0.05$ ) (Table 3).

## 5. Relation between working status and pregnancy complications

Nine subjects were diagnosed with threatened preterm delivery and treated with internal medicine, while 2 subjects were diagnosed with pregnancy-induced hypertension. The development of these diseases had no bearing on whether these 11 subjects continued working or not.

## 6. Effects on Health Conditions of Newly-born Babies

Sixty-two subjects all delivered babies via normal delivery ( in-womb period: from 37 weeks and five days through 41 weeks and five days) and the average weight of newly-born babies was  $3,031 \pm 350\text{g}$  (N=62).

Given the body weight, height, head circumference, chest circumference and placenta weight as dependent variables and also items such as working status, personal factors, and breakdowns of daily hours as independent variables, a multiple linear regression analysis was performed, but no influential variable was found.

## Discussion

The General Health Questionnaire (GHQ) is a screening test developed by Goldberg et al.<sup>3)</sup> and is effective in detection and evaluation neurosis symptoms. It has been modified and translated into a Japanese language version by Nakagawa et al.<sup>4)</sup>, and is now in use as a Japanese shorter version (GHQ28).

The reliability and applicability of GHQ28 have been validated, and the results of GHQ28 are evaluated by total scores and subscores of four factors: A) Somatic symptoms, B) Anxiety and Insomnia, C) Social Dysfunction, D) Severe Depression disorder. The total score ranges from 0 to 28 points, and the higher the score, the more serious the mental health problems are. The cut-off point to decide the mental health condition was from 5 to 6.

In this study, we analyzed GHQ28 results by the cut-off point values of total scores and the average values on each factor's scale. The total scores of GHQ in this study decreased significantly in late pregnancy compared with early pregnancy( $p<0.01$ ). Looking at the subscores of respective factors, the subscores of Somatic symptoms significantly fell and those of the other three factors tended to decrease in late pregnancy. These results prove that the mental stress levels of pregnant women are higher with the somatic symptoms in early pregnancy and decrease as pregnancy progresses. In a report by the Japan Association for The Advancement of Working Women in 2006<sup>5)</sup>, the greatest number of pregnant workers complained of physical anxiety like emesis as a stress factor during gestation. Emesis in pregnancy generally appears during the 5th to 6th week of pregnancy and is generally relieved during the 12th to 16th week; nevertheless, it varies greatly between individual to individual<sup>6)</sup>. Because the early survey in the gestation period in this study was made during the 12th to 16th week when pregnancy emesis is relatively relieved but continuing, this presumably reflects the higher degrees of somatic symptoms caused by emesis.

On the other hand, the total scores of GHQ12 in the study of Matsuzaki et al.<sup>7)</sup> did not recognize any significant differences between early pregnancy (the 12th to 13th week), mid-pregnancy (the 22nd to 26th week), and late pregnancy (the 30th to 34th week). Matsuzaki et al.<sup>7)</sup> used GHQ12, in which the number of questions in the questionnaire was less than half that of GHQ28. The results of GHQ12 showed a problem that the values of the standard deviation were larger than the average values of the total scores. Furthermore, they selected different pregnant women at the three stages, that is, the subjects in early pregnancy, mid-pregnancy and late pregnancy were not the same. In this study, we analyzed the

working and individual factors as possible variables that may affect the total scores of GHQ28; nevertheless, no such variables were significant either in early pregnancy or late pregnancy.

Among the various methods for measuring antioxidant capacity, we used a copper reduction assay with a PAO measurement kit for this study. Copper has advantages over iron for antioxidant assays in that all classes of antioxidants, including thiol, are detected with little interference from reactive radicals, while the copper reaction kinetics are faster than iron. The assay requires only 3 min; the assay is complete in minutes for ascorbic acid, uric acid, gallic acid, and quercetin, but requires 30-60 min for more complex molecules. Thus, the copper reduction assays have similar problems with a complex mixture of antioxidants in terms of selecting an appropriate reaction time<sup>8)</sup>.

We also studied the changes in PAO levels after blood collection. The results showed that the PAO levels stayed stable more than four hours after blood collection (Fig 3). Therefore, we separated serum from blood samples and kept them at -80°C within four hours of blood collection. The decrease in PAO due to smoking was proved in the study by Dietrich et al.<sup>9)</sup> and the decrease was especially significant in the umbilical cord blood<sup>10)</sup>. However, in this study, no decrease in the PAO level due to smoking was observed either in early or late pregnancy; the reason for this is yet to be confined. Some kind of connection was recognized between body mass index (BMI) and the PAO values in early pregnancy, but no prior report on this phenomenon has been published to date. Since PAO is said to be affected by the intake of anti-oxidant substances in food<sup>11)</sup>, a further study of the connection between BMI and diet on PAO levels of pregnant women may be necessary.

8-OHdG is marker of oxidative DNA damage in whose structure guanine has been oxidized by reactive oxygen<sup>12)</sup>. Since urinary 8-OHdG can reflect oxidative stress non-invasively, it is now widely used as an oxidative stress marker. It was reported that urinary 8-OHdG levels of female shift workers were significantly higher than those of female part-time workers<sup>13)</sup>. Moreover, the urinary 8-OHdG of male shift workers was reported to increase compared with male day-time workers<sup>14)</sup>. Urinary 8-OHdG is widely used as an oxidative stress marker to study the influence of working, but no report has yet been done concerning pregnant women.

Both values of 8-OHdG after creatine correction and that after specific gravity correction in late pregnancy were significantly lower than in early pregnancy ( $p<0.01$ ,  $p<0.05$ ), giving the same result as for mental stress that was reduced in late pregnancy. It has been reported that females have increased oxidative stress levels as a result of mental stress<sup>15)16)</sup>. The dual decreases in the scores of mental stress (GHQ) and the values of oxidative stress markers in late pregnancy support those preceding studies.

On the other hand, Matsuzaki et al. reported that urinary biopyrrin, an oxidative stress marker, significantly increased in late pregnancy compared with early or mid-pregnancy<sup>7)</sup>. Furthermore, in a study by Tateoka et al., cortisol concentration, another stress indicator, significantly increased during early (the 8th to 12th week of gestation) and mid pregnancy (the 22nd to 26th week), and even more significantly in late pregnancy (the 30th to 34th)<sup>17)</sup>. Those reports indicated opposite results to this study. 8-OHdG is a DNA oxidative damage marker involved not only in oxidative stress, but also DNA

repair enzyme activities. The reasons why 8-OHdG levels were lower in late pregnancy in this study are thought to be an increase in defense systems such as PAO and DNA repair.

Urinary 8-OHdG in non-working pregnant women was significantly higher than in working women ( $p < 0.05$  after creatinine correction,  $p < 0.001$  after specific gravity correction) in this study. According to a study by Shinkawa et al.<sup>18)</sup>, the occurrence of minor pregnancy problems occurred more often in non-working women than working women, Yubune et al.<sup>19)</sup> also reported lower degrees of depression and physical stress in working pregnant women. A study by the above-mentioned Japan Association for The Advancement of Working Women<sup>5)</sup> reported that non-working pregnant women scored higher in all stress check items than working pregnant women, indicating that the results of this study were the same as those of these preceding studies.

In this study, we examined the effects of continuation of work during pregnancy on the mental and physical condition of pregnant women. The hypothesis that continuation of work early in the gestation period was harmful to pregnancy was rejected because of the lower oxidative stress marker levels in working pregnant women. Instead, the study recognized lower physical stress when working during pregnancy. Yamada et al.<sup>20)</sup> reported higher rates of headaches with full-time housewives than with working women; the reason was that the former have little satisfaction or fulfillment and greater stress from their activities, while working women have greater satisfaction and fulfillment by receiving individual recognition and rewards. Besides, other studies of working women also often report fulfillment and

self-satisfaction by working<sup>22)23)</sup>. Although these studies were conducted with only non-pregnant or child-raising women, the same results are feasible.

Eight pregnant working women took maternity leave in early to late pregnancy, and another 16 resigned their jobs. Those who resigned their jobs in early pregnancy were compared with those who continued working up to late pregnancy, and no significant differences were recognized between them on GHQ28, PAO, or urinary 8-OHdG either in early or late pregnancy. Although working during pregnancy did not have any adverse effects on pregnant women in this study, it should be remembered that some hard work is really harmful to pregnant women.

It is necessary to construct social environments where women feel they can work easily, get pregnant, have babies, and raise children. In a further study, an increased number of study subjects and a diverse, long-term viewed analytical study of working and living environments and various oxidative stress markers to evaluate mental and physical stress of pregnant women are necessary.

## Conclusion

We examined the working, life-style, environment, and mental stress (GHQ28) by questionnaires , and the PAO in blood serum and urinary 8-OHdG in 62 pregnant women who were free of any basic diseases and were pregnant by natural insemination with a single fetus, first in early pregnancy (the 12th to 16th week) and then in late pregnancy (the 32nd-36th week). The values of mental stress were higher in early



pregnancy and decreased as the pregnancy advanced. Also, the values of urinary 8-OHdG, a marker of physical oxidative stress, decreased, as the values of antioxidant capacity (PAO) increased, in late pregnancy. No relation was detected between working and threatened preterm premature delivery, pregnancy hypertension or the condition of the delivered babies. Working during pregnancy did not increase the mental or physical stress of pregnant women, but rather reduced physical stress.

#### Acknowledgements

This work was supported in part by Grants-in-Aid from the Ministry of Education, Culture, Sport, Science and Technology of Japan (23593360) and UOEH Research Grant for Promotion of Occupational Health.

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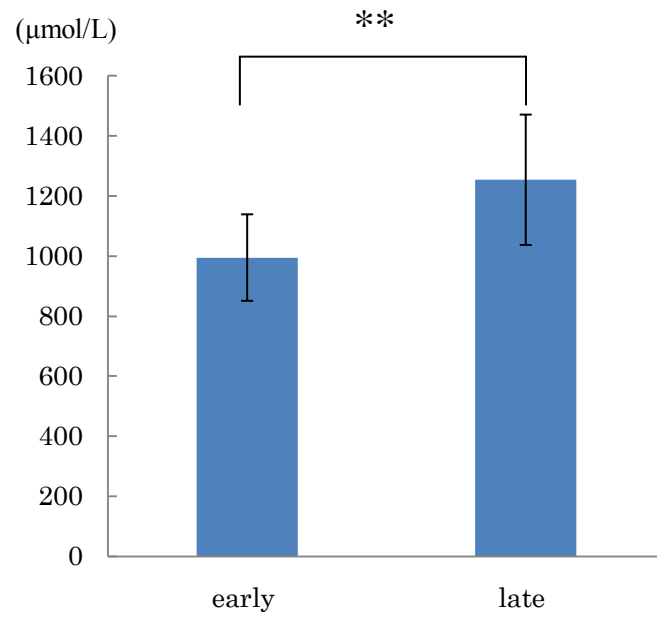


Fig. 1. Serum potential antioxidant (PAO) levels in early(12-16w) and late(32-36w) pregnancy  
\*\*p<0.01(paired t-test)

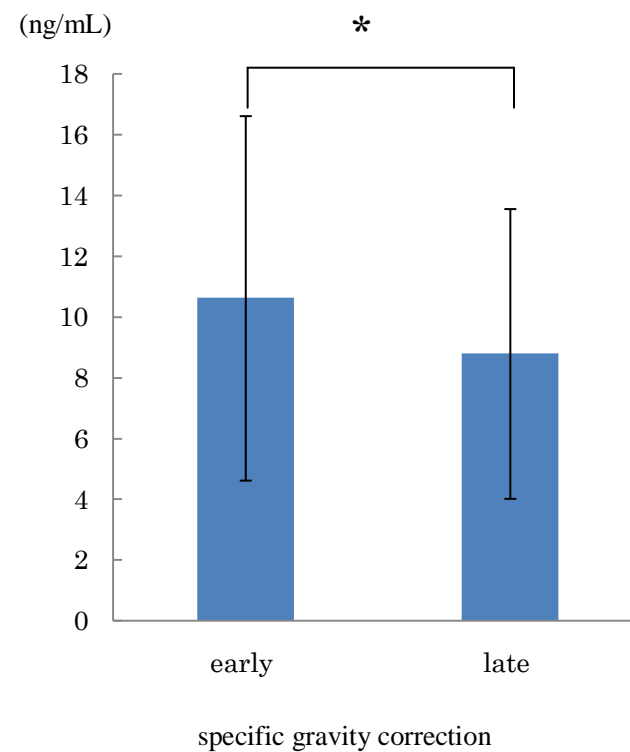
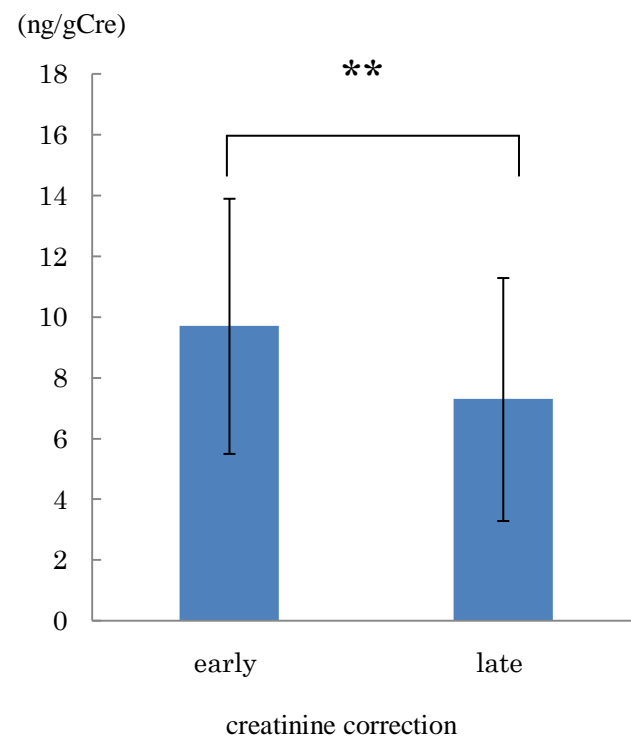


Fig. 2. urinary 8-OHdG levels after creatinine correction and specific gravity correction in early (12-16w) and late (32-36w) pregnancy

\*\*p<0.01, \*p<0.05 ( paired t-test)

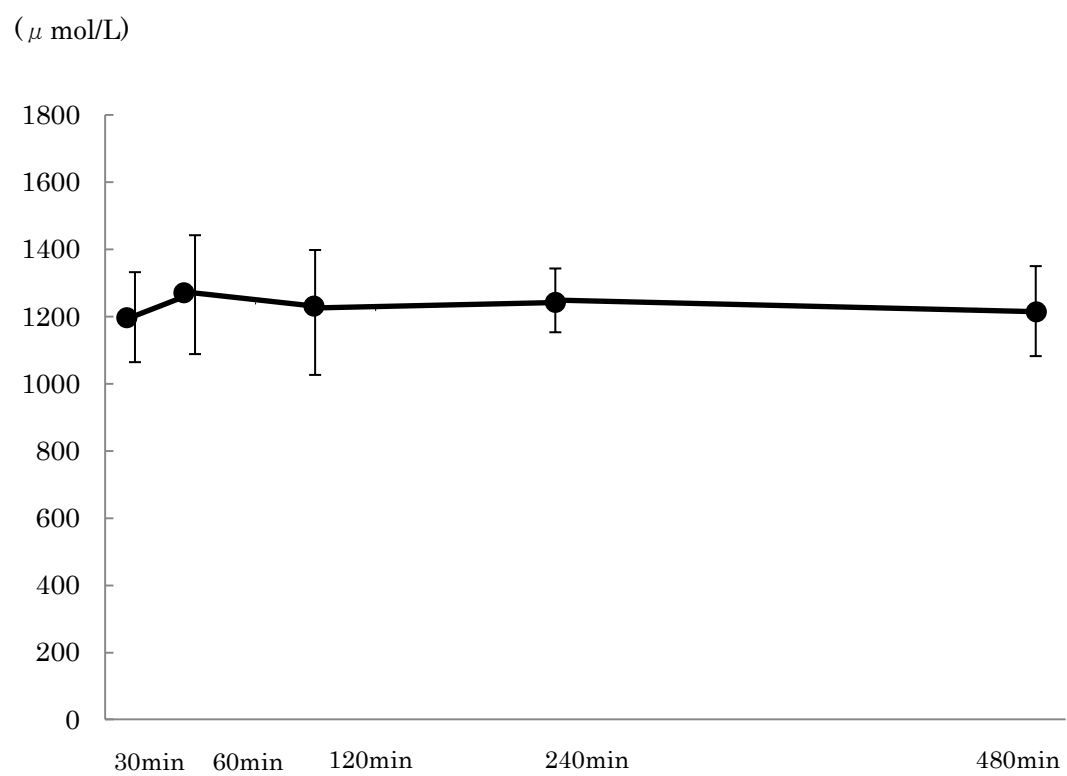


Fig. 3. Change of Potential anti-oxidant (PAO) levels in serum after blood collection.  
Blood samples are kept at  $+4^{\circ}\text{C}$  untill serum separation

Table1. Characteristics of the subjects

	Number of subjects (%)	Age (Mean±SD)
History of delivery		
Primigravida	23 (37.1)	29.2±4.0
Multigravida	39 (62.9)	31.3±4.7
Working status		
Early pregnancy (12-16w)		
Working	41 (66.1)	31.5±4.3
Out of working	21 (33.9)	28.4±4.3
Late pregnancy (32-36w)		
Working	17 (27.4)	31.8±4.3
Out of working	45 (72.6)	30.0±4.3



Table2. Results of GHQ28 in early and late pregnancy

		Mean ± SD			≤5	≥6		
Total Scores					n(%)	n(%)		
	Early pregnancy	7.37±4.57	]	* *	25(40.3)	37(59.7)	]	*
	Late pregnancy	5.44±3.61			33(53.2)	29(46.8)		
Subscores								
(A)	Somatic Symptoms							
	Early pregnancy	3.13±1.95	]	* *				
	Late pregnancy	1.90±1.59						
(B)	Anxiety and Insomnia							
	Early pregnancy	2.58±1.63	]	n.s				
	Late pregnancy	2.24±1.64						
(C)	Social Dysfunction							
	Early pregnancy	1.55±1.92	]	n.s				
	Late pregnancy	1.19±1.14						
(D)	Severe Depression							
	Early pregnancy	0.11±0.32	]	n.s				
	Late pregnancy	0.10±0.43						

N=62

\*\*p&lt;0.01 ( paired t-test)

\*p<0.05 (  $\chi^2$  test)

**Table3.** Stepwise multiple linear regression analysis of the urinary 8-OHdG ,PAO: Early pregnancy and late pregnancy

Early pregnancy (12-16w)	log(PAO) <sup>a</sup>					8-OHdG (Creatinine correction)					8-OHdG (Specific gravity correction)				
	partial	regression	standard	partial	p value	partial	regression	standard	partial	p value	partial	regression	standard	partial	p value
	coefficient	coefficient	regression	coefficient	(p)	coefficient	coefficient	regression	coefficient	(p)	coefficient	coefficient	regression	coefficient	(p)
Primigravida/multigravida	—		—		—	—		—		—	—		—		—
Working/non-working	—		—		—	−2.70		−0.31		0.017	−4.56		−0.37		0.003
Smoker/non-smoker	—		—		—	—		—		—	—		—		—
Drinker/non-drinker	—		—		—	—		—		—	—		—		—
Age	—		—		—	—		—		—	—		—		—
Body mass index	0.006		0.320		0.01	—		—		—	—		—		—
Times of going up and down stairs per day	—		—		—	—		—		—	—		—		—
Working hours per day	—		—		—	—		—		—	—		—		—
Sleeping hours per day	—		—		—	—		—		—	1.32		0.28		0.021
Household work hours per day	—		—		—	—		—		—	—		—		—
Child-raising hours per day	—		—		—	—		—		—	—		—		—
Resting hour per day	—		—		—	—		—		—	—		—		—
Intercept	2.85				0.00	11.43				0.00	13.49				0.00
	R-square 0.21 p-value <0.001					R-square 0.09 p-value <0.05					R-square 0.22 p-value <0.001				
Late pregnancy (32-36w)	log(PAO) <sup>a</sup>					8-OHdG (Creatinine correction)					8-OHdG (Specific gravity correction)				
	partial	regression	standard	partial	p value	partial	regression	standard	partial	p value	partial	regression	standard	partial	p value
	coefficient	coefficient	regression	coefficient	(p)	coefficient	coefficient	regression	coefficient	(p)	coefficient	coefficient	regression	coefficient	(p)
Primigravida/multigravida	—		—		—	—		—		—	—		—		—
Working/non-working	—		—		—	—		—		—	—		—		—
Smoker/non-smoker	—		—		—	—		—		—	—		—		—
Drinker/non-drinker	—		—		—	—		—		—	—		—		—
Age	—		—		—	—		—		—	−0.293		−0.282		0.032
Body mass index	—		—		—	—		—		—	—		—		—
Times of going up and down stairs per day	—		—		—	—		—		—	—		—		—
Working hours per day	—		—		—	—		—		—	—		—		—
Sleeping hours per day	—		—		—	—		—		—	—		—		—
Household work hours per day	—		—		—	—		—		—	—		—		—
Child-raising hours per day	—		—		—	—		—		—	—		—		—
Resting hour per day	—		—		—	—		—		—	—		—		—
Intercept											17.71				0.00
											R-square 0.08 p-value <0.05				

<sup>a</sup>Logarithm of PAO(μmol/L)

