

Study of Pattern of Lactogenesis in Women with Chronic Stress by Analyzing Milk output in the First Week of Postpartum

Vandali Jyothi¹, Mohd Noorjahan Begum², D Rajashree³,
K Prashanth⁴, Neerja Shastri⁵

Authors Affiliation

¹Associate Professor, ²Assistant Professor, Department of Physiology, Malla Reddy Medical College for Women, Hyderabad, Telangana 500055, India. ³Professor & Head, ⁴Assistant Professor, Department of Physiology, Malla Reddy Medical College for Women, Suraram, Hyderabad, Telangana 500055, India. ⁵Professor & Head, Department of Physiology, Medical College, Nizams's Institute of Medical Sciences, Jaipur, Rajasthan 303121, India.

Corresponding Affiliation

Mohd Noorjahan Begum, Assistant Professor, Department of Physiology, Malla Reddy Medical College for Women, Hyderabad, Telangana 500055, India
Email: dr.noorie@gmail.com

Abstract

Introduction: Chronic stress is long term stress, generally lasting weeks to months to years. The concept of chronic stress is based on how frequently the stressors appear over period of time. Chronic & severe stressful events may alter hormonal balance & lead eventually to failure of homeostasis. Chronic stress response leads to pregnenolone steal, which causes production of huge amount of Cortisol (the principal stress hormone). So, serum cortisol level can be considered as an objective measure of chronic stress. Excessive Cortisol levels during pregnancy has been associated with adverse pregnancy outcomes. Keeping in view of this, present study was conducted to assess effect of chronic stress on the pattern of milk output in women of reproductive age.

Materials and methods: A longitudinal cohort study was conducted in pregnant women in the reproductive age between 21 and 45 years. Holmes and Rahe stress scale was used to measure stress. In our study we have included anthropometric parameters (Height, Weight, BMI), physiological parameters (BP, HR) and hormonal assay (for cortisol, prolactin). Volunteers were screened in 1st, 2nd and 3rd trimesters as well as during postpartum upto 7days. Milk volumes were measured for 7 days by baby test weighing method.

Results: We observed that mean cortisol levels in 1st trimester were 14.15±1.91, 20.44±1.33, 23.10±2.53 µg/dl in mild, moderate and severely stressed women respectively. The levels were in increasing order in 2nd, 3rd trimester and postpartum. Mean prolactin levels were in decreasing order in women with mild to severe stress. It was found that in moderately and severely stressed women, prolactin levels were in decreasing order from 1st day to 5th day postpartum when compared with mildly stressed women. It was observed that mildly stressed women produced ~ 700ml of milk per day, moderately stressed women produced ~ 425 ml/day and severely stressed women produced ~ 360 ml/day. Milk volumes were reduced by ~ 40% in moderately stressed women and ~ 50% in severely stressed women when compared with milk volumes of mildly stressed women.

Conclusion: Serum Cortisol levels of moderate and severely stressed women were significantly raised in comparison with mildly stressed women. Cortisol levels were high after delivery also. This indicates chronically stressed women remain in the same stressed state even in the postpartal period. In our study, serum prolactin levels were decreased in moderate as well as in severely stressed groups when compared with mildly stressed group, and has shown its effect on milk volume. Decreased milk volume in moderate and severely stressed women indicates that cumulative effects of changing life events and daily hassles with failure of coping reflex leads to chronic stress which causes decreased lactation.

Keywords: Chronic stress; Cortisol; Milk volume; Prolactin.

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Introduction

Chronic stress is long lived stress, generally lasting weeks to months to years. So, the physiological response does not have effective resolution to the stressors.¹ Pregnancy is recognized as one of the stressful events in woman's life. It is a time of physiological change that needs huge psychological adjustments.² Women's health care providers have investigated and are increasingly aware that the chronic stressors i.e. ongoing perceived stress/threat and anxiety are due to poverty, intimate partner violence and experiences of racism. They are associated with an increased incidence of preterm birth and low birth weight of the baby in United States.³ Presence of major events in the life affects the daily activities of a person. Some of the studies have shown significant association of stressful life events during pregnancy on reducing age of gestation. Presence of two or more major life events during pregnancy has been linked to birth of baby with prematurity and low birth weight.^{4,5} Chronic stress response leads to pregnenolone steal, which causes production of huge amount of cortisol, the principal stress hormone. So, serum cortisol level can be considered as an objective measure of chronic stress. Excessive Cortisol levels during pregnancy has been associated with adverse pregnancy outcomes.^{6,7} Some animal experiments have demonstrated suppression of lactation when animals were exposed to certain types of stressful stimuli.^{8,9,10,11} In humans, lactation insufficiency may be due to the stress imposed by preterm delivery, infant medical condition, maternal life style or life events.^{9,10} Duration of stress and its sites of action lead to decrease in milk synthesis or ejection which causes the suppression of lactation.^{12,13} As per the human experiment done by Newton and Newton, various types of acute stressful stimuli decreased the milk ejection reflex.¹⁴ Udea T et al have shown that mental stress and noise stress have reduced milk volume.¹⁰ These studies were demonstrated during established lactation and not during lactogenesis. This research indicate the effect of acute stress on lactation i.e. Milk volume was measured before the induction of stress, during the time of stress and after the removal of stress. Milk volume was normal before induction of stress, volume was reduced during the time of stress as the milk ejection was affected and normal milk

volume was regained after recovery from the stress. As the effects of chronic stress were not yet known, we have conducted a study to assess the effect of chronic stress on milk output in humans.

Materials and methods: The present study was undertaken to assess the milk volume output in chronically stressed women. Pregnant women attending the antenatal clinic at Prathima Institute of Medical Sciences, Nagunur, Karimnagar were included.

Inclusion criteria: Pregnant women were in the reproductive age between 21-45 yrs, Women who fulfilled the selection criteria as per Holmes and Rahe stress scale, Women who agreed to be in the study group till 7 days postpartum.

Exclusion criteria: Pregnant women with history of non lactogenesis, Women with no intention to breastfeed, Women who were HbSAg or HIV positive, Alcoholics, smokers, known diabetics, with h/o major endocrinal abnormality. Participants were divided into 5 age groups. 21-25 years, 26-30 years, 31-35 years, 36-40 years, 41-45 years. A total of 128 pregnant women were included. With 95% confidence level and margin of error of $\pm 10\%$, a sample size of 96 subjects were allowed to participate in the study.

Volunteers were screened at visits 1-10. Out of 10 visits, 3 were held before delivery and 7 were held during postpartum period. It is a follow-up study. The pregnant women were screened in 1st, 2nd and 3rd trimesters as well as during postpartum up to 7 days.

Parameters included:

1. To measure stress according to the Holmes and Rahe Stress Scale, the number of "Life Change Units"(LCU) that apply to events in the past year of an individual's life^{15,16,17,18,19,20,21}, were added and the final score gave a estimate of how stress affects health. Participants were assessed for the level of stress by asking questionnaire as per Holmes and Rahe stress scale. Questionnaire was in regard of life events which occurred in past one year like death of spouse, divorce, marital separation, imprisonment, pregnancy, marriage, marital reconciliation, sexual difficulties, dismissal from work,

change in financial state, change in sleeping or eating habits etc. to estimate the LCU score. Accordingly we have categorized participants into mildly, moderately, severely stressed groups.

Mild stress; LCU<150, moderate stress; LUC150-299, severe stress; LCU= 300.

2. Anthropometric measurements were recorded.
 - a. *Body weight:* Body weight was measured (to the nearest 0.5 Kg) with subject wearing light clothing without shoes, standing motionless on the weighing scale and with the weight distributed equally on both legs.
 - b. *Height:* Height was measured in cms (to the nearest 0.5cm) with the subject standing in an erect position against a vertical scale and with the head positioned so that the top of the external auditory meatus was level with the inferior margin of the bony orbit (Frankfurt's plane).
 - c. *Body mass index (BMI):* BMI of each participant was calculated as body weight in kilograms divided by square of body height in meters. $BMI = kg/m^2$
3. Recording of physiological parameters:
 - a. *Heart rate (HR) :* Heart rate of individual participant was recorded in sitting posture. The radial pulse is examined by compressing the radial artery against the head of radius. For elucidation of the pulse, the forearm of the subject should be semipronated and the wrist was slightly flexed. Heart rate was recorded as beats/min.
 - b. *Blood pressure (BP):* Blood pressure was measured as per WHO guidelines using a standard mercury sphygmomanometer in sitting posture. Three recordings were taken for each subject. Average of 3 measurements was taken for calculation.
4. *Hormone assay:*
 - a. Objective measurement was done by analyzing the serum cortisol levels in all the three trimesters of pregnancy and postpartum. Blood samples (5-7ml) were collected by venipuncture between 8.30am - 10.30am with all sterile precautions, placed in evacuated tubes and then taken to laboratory in ice box, centrifuged at 1300xg for 20 min at 4°C in a refrigerated table top centrifuge. Serum samples for hormone assays were frozen at -20°C until analyzed by electrochemiluminescence immunoassay (Roche Elecsys 1010/2010).

- b. Antenatal and postpartum blood samples (5ml) were collected from 1st to 5th day with standard aseptic precautions. Quantitative determination of Serum Prolactin ($\mu g/L$) was done by electrochemiluminescence immunoassay.
5. Measurement of milk volume: Women were encouraged to express breast milk 10 times per day from day 1 to day 7 postpartum. Milk volumes (ml) were measured by test weighing method on electronic digital weighing scale to the nearest of 0.01g[22]. Test weighing method: Goldtech digital baby weighing scale was used to measure the weight of the baby.

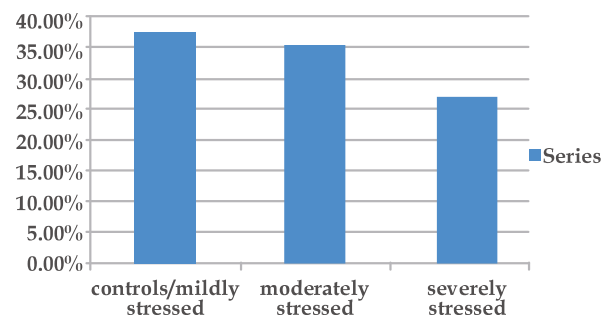
Results were recorded and data analysis was done. All characteristics were summarized descriptively. For all continuous variables, the summary statistics of N, mean and standard deviation (SD) were used. For all categorical data, numbers and percentages (%) were used in the data summaries. If the p-value was < 0.05, then the results were considered to be statistically significant otherwise it was considered as not statistically significant. The SPSS software v.23.0. was used for data analysis.

Results

Table 1: Categorization of women of different age groups into different stress levels.

Age groups (yrs)	Control or mildly stressed (<150)	Moderately stressed (150-299)	Severely stressed (>300)	Total
20 - 25	14	7	7	28
26 - 30	8	9	5	22
31 - 35	8	9	6	23
36 - 40	4	7	3	14
41 - 45	2	3	4	9
	36(37.5%)	35 (36.5%)	25 (26%)	96

Graph 1 : Categorization of participants(%) into different stress levels.



As per Holmes and Rahe stress scale 96 participants were categorized into different stress levels. As pregnancy itself is a stressful event in women’s life, mildly stressed women were considered as controls. We found 37.5% were mildly stressed, 36.5% were moderately stressed and 26 % were severely stressed among total participants.

Table 2: Mean and SD of cortisol levels of study subjects in 3 trimesters & postpartum at different stress levels.

Stress levels (LCU)	Cortisol in 1st tri (µg/dl)	Cortisol in 2nd tri (µg/dl)	Cortisol in 3rd tri (µg/dl)	Postpartum cortisol (µg/dl)
<150	14.15±1.91	22.16±1.99	26.48±1.85	26.50±1.91
150-299	20.44±1.33	25.91±1.79	29.72±2.95	30.79±3.39
>300	23.10±2.53	27.56±3.09	33.06±2.97	34.15±3.19

Graph 2: Mean and SD of cortisol levels of all study subjects in 3 trimesters, postpartum at different stress levels.

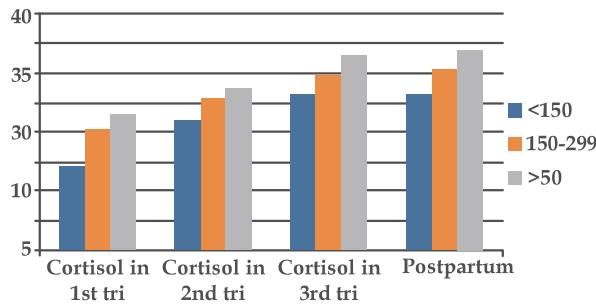


Table 4: Mean and SD of Prolactin (Prl) levels of all study subjects before delivery, for first 5 days postpartum (PP) at different levels of stress.

Stress levels	Prl Before delivery	PP Prl on D- 1	PP Prl on D-2	PP Prl on D- 3	PP Prl on D-4	PP Prl on D-5
<150	175.58±29.30	172.08±32.68	183.75±33.05	197.44±31.50	215.27±38.44	240.44±41.84
150-299	148.85±22.19	141.44±21.41	151.85±20.50	162.97±15.52	179.52±14.94	204.5±15.55
>300	133.73±21.98	125.11±23.66	137.11±22.41	152.30±20.02	170.46±17.20	191.92±12.79

Graph 3: Mean and SD of Prolactin(Prl) levels of all study subjects before delivery, for first 5 days postpartum(PP) at different levels of stress.

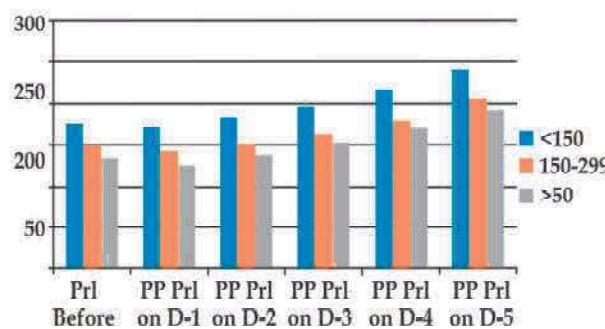


Table 3: Comparison of age, stress level and mean Cortisol level (µg/dl) for 3rd trimester & Postpartum.

Age group (years)	Stress level (LCU)	Cortisol level (µg/dl)		t value	p value
		(Mean±SD)			
		3rd trimester	Post Partum		
21-25	Mild	26.2±1.5	26.2±1.4	-0.5	0.62
	Moderate	29.5±3.2	31.7±3.3	-5.2	0.002*
	Severe	32.4±2.3	34.6±3	-2.9	0.027*
26-30	Mild	26.5±1.7	26.5±1.9	0.0	0.979
	Moderate	30±2.8	31±3.6	-3.4	0.009*
	Severe	32.4±2.4	33.7±3.1	-1.8	0.152
31-35	Mild	27.2±2.4	27±2.5	2.1	0.077
	Moderate	29.7±2.7	30.4±2.8	-3.0	0.016*
	Severe	33.8±3	34.5±3.2	-2.7	0.043*
36-40	Mild	25.9±2	25.8±2.2	0.8	0.507
	Moderate	29.9±3.2	30.8±3.8	-2.3	0.062
	Severe	35±4.8	35.2±5	-1.2	0.362
41-45	Mild	26.6±3.6	27.5±3.4	-7.2	0.088
	Moderate	29.3±4.6	29.7±5	-1.8	0.214
	Severe	33.2±4.1	33.3±4	-1.5	0.241

Mean cortisol level in 3rd trimester of mildly stressed and postpartum were between 25-27.2 µg/dl and 25-27.5µg/dl respectively. In 3rd trimester of moderately stressed and postpartum were between 29-30 µg/dl and 29-31 µg/dl respectively. In 3rd trimester of severely stressed and postpartum were between 32-35 µg/dl and 33-35.2 µg/dl respectively. So we found no significant difference between 3rd trimester and postpartal cortisol levels.

Table 5: Comparison of age group, stress levels & mean Prolactin levels during antenatal and on day 5 after delivery.

Age group (years)	Stress level (LCU)	N	Prolactin level (Mean±SD)		t value	p value
			Ante natal Prolactin level	Post partum day 5		
			21-25	Mild		
21-25	Moderate	7	167.9±19.7	202.1±13.8	-3.8	0.004*
	Severe	7	158.7±17.2	170.7±17.4	-1.3	0.221
26-30	Mild	8	180.8±26.3	243.8±58.8	-2.8	0.022*
	Moderate	9	168.9±22.6	204.9±20	-3.6	0.003*
	Severe	5	166.6±22.8	179.4±9.8	-1.2	0.301

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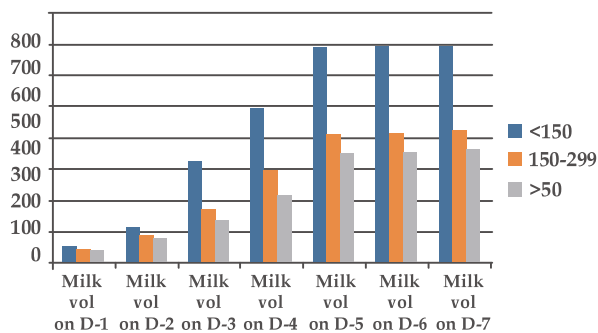
31-35	Mild	8	157±25.2	226.4±21.3	-5.9	<0.001*
	Moderate	9.0	145.1±19.8	172.2±16.3	-3.2	0.006*
	Severe	6	139±27.2	170.2±10.4	-2.6	0.039*
36-40	Mild	4	151.3±21.3	200±13.3	-3.9	0.012*
	Moderate	7.0	147.1±28.2	172.7±7.5	2.3	0.059
	Severe	3	128.3±16.8	170.3±6.8	-4.0	0.057
41-45	Mild	2	171.5±40.3	228±17	-1.8	0.319
	Moderate	3.0	159±31.2	172.3±14	-0.7	0.570
	Severe	4	149.3±27.5	170.5±6.5	-1.5	0.230

In all the age groups of antenatal and day-1 postpartum, the difference in mean prolactin levels were very slight and up to day 5 postpartum there was no significant increase. The serum prolactin levels were not significantly altered in moderate and severely stressed groups when compared with mildly stressed group and the effect was observed on milk volume.

Table 6: Mean and SD of milk volumes of all study subjects for first 7 days postpartum at different stress levels.

Stress level	Milk vol on D-1	Milk vol on D-2	Milk vol on D-3	Milk vol on D-4	Milk vol on D-5	Milk vol on D-6	Milk vol on D-7
<150	49.43±3.77	108.68±14.80	326.77±50.76	505.10±66.65	689.45±31.56	692.76±30.41	699.89±29.77
150-299	43.60±3.28	85.89±5.15	172.81±18.59	294.54±20.53	409.83±42.80	413.20±43.02	423.09±41.47
>300	38.82±4.89	78.26±7.51	134.44±16.13	217.70±49.22	349.53±39.38	353.76±40.30	361.65±36.85

Graph 4: Mean and SD of milk volumes of all study subjects for first 7 days postpartum at different stress levels.



Total milk volumes of all participants were measured at different stress levels. It was observed that mildly stressed women produced ~ 700ml of milk, moderately stressed women produced ~ 425ml and severely stressed women produced ~ 360 ml of milk. Milk volumes were reduced by ~ 40% in moderately stressed women and ~ 50% in severely stressed women when compared with milk volumes of mildly stressed women.

Discussion

Serum Cortisol levels of moderate and severely stressed women were significantly raised in comparison with mildly stressed women (table-2). Similar results were observed by Dallman et al²³, Harvaline et al²⁴ and Obel et al²⁵ and also by few other studies.^{26,27} The difference in the 3rd trimester and postpartal Cortisol level was insignificant indicating that chronically stressed women remain in the same stressed state even in the postpartal period (table-3). In our study, serum Prolactin levels were not significantly altered in moderate and severely stressed groups when compared with mildly stressed

group (table-4). Total milk volumes of all subjects were measured at different stress levels. It was observed that mildly stressed women produced ~ 700ml of milk, moderately stressed women produced ~ 425ml and severely stressed women produced ~ 360 ml of milk. Milk volumes were reduced by ~ 40% in moderately stressed women and ~ 50% in severely stressed women when compared with milk volumes of mildly stressed women (table-6) by baby test weighing method. Similar studies were done by Savenije et al, Arthur et al.^{28,29}

Ueda et al in 1994 conducted a similar study in breastfeeding women assigned with noise stress and mental stress. They also found the same results[10]. Most of the studies were performed during established lactation, not during lactogenesis. So, this study is conducted to assess the effect of chronic stress on milk output in humans. Conclusion: 62.49 % of Indian women are living with stress, of which 35.41% are moderately and 27.08% are severely stressed. Chronically stressed women remain in the same stressed state even in the postpartal period. Chronic stress of different levels did not show much effect on serum prolactin levels during pregnancy and postpartum. Moderate stress of up to 299 on Holmes and Rahe stress scale in younger age group (21-30 years) suppresses the initial phase (first two days) of lactation, but lactation improves from third day. Severe stress of >300 in the same age group has deleterious effect on all phases of lactation (even up to seventh day). In the women of middle and elderly age group (above 30 years), moderate and severe stress has compounding negative effect leading to difficulty in initiation and establishment of lactation. Moderate and severe stressful life events reduce milk volume output in humans. So it is essential to bring awareness in

population about deleterious effects of stress on lactation. It is essential to assess the stress in the 1st trimester and treat those women with proper measures to reduce the stress level. It may partially nullify the effects of chronic stress on lactogenesis and normal lactation can be established.

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