

# ORIGINAL ARTICLE

## The Relationship between Anxiety and Salivary Alpha-Amylase Levels in Mothers of Neonates Admitted to the Neonatal Intensive Care Unit

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### Abstract

#### Objectives

Anxiety is a significant health issue in mothers who give birth to unhealthy neonates. Different studies have investigated the relationship between anxiety and alpha-amylase. According to the necessity for psychological care of mothers whose infants are hospitalized in intensive care units and consequently the anxiety that is imposed on mothers due to the condition, this study aims to assess anxiety in these mothers and the relationship between this anxiety and salivary alpha-amylase.

#### Materials & Methods

This study was a cross-sectional study conducted at Besat Hospital in Hamadan in 2021. Thirty mothers were enrolled in the study through a census method sampling. The Hamilton questionnaire measured maternal anxiety during the children's hospitalization period. Salivary alpha-amylase samples were taken from all mothers according to the scientific method, and mothers' salivary alpha-amylase levels were measured. All the gathered data were analyzed using SPSS 21 software. The significant level was considered 0.05 in all comparisons.

#### Results

The mean age of the 30 mothers was 29.27+6.24 years. The mean score of maternal anxiety was 16.27 + 6.78, and the mean salivary amylase level was 33.02 ± 16.22 U / ml. Fourteen mothers obtained low anxiety scores, 14 had moderate anxiety, and two had severe anxiety. No significant relationship was found between the mean of salivary alpha-amylase at the three levels and anxiety. There was no

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## Introduction

Low birth weight and premature infants are high-risk groups in society and experience more physical and mental problems than normal infants. As a result, to survive and achieve normal growth, they need special care, including hospitalization in the neonatal intensive care unit (NICU) (1, 2).

Ample evidence was available that parents of an infant with health problems are more likely to experience social, economic, and emotional problems that are often restrictive, destructive, and pervasive. In such a situation, however, all members of the family and its function are harmed. Nevertheless, because of their traditional role of “caregiver,” take on more responsibilities towards their infant, leading to more psychological problems. Caring for a troubled infant puts parents, especially mothers, at risk for mental

significant relationship between anxiety level with parent location, age, mother’s level of education, infant gender, and child rank.

## Conclusion

According to the results of the present study, there was no significant relationship between the level of alpha-amylase and the level of anxiety in mothers of neonates admitted to the neonatal intensive care unit (NICU), so further researches in similar groups that are in anxious conditions seem necessary.

**Keywords:** Neonatal Intensive Care Unit; Anxiety; Alpha-Amylase, Saliva

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health problems (3, 4). The tensions, pressures, and psychological burdens of the mothers of these infants are significant and commonplace, but if left untreated, they can lead to decreased physical and mental health and reduced quality of life. The psychological burden of caring for a disabled or physically ill child can reduce the quality of care provided and endanger caregivers’ physical and mental health (5, 6).

Studies on anxiety, in addition to the psychological dimension from a neuropsychological perspective, are necessary because the knowledge of neuropathology and molecular biology of anxiety opens new ways to understand the causes and find more specific solutions for anxiety disorders. Biologically, the causes of anxiety can be attributed to the autonomic nervous system and neurotransmitters. Numerous studies have shown a strong positive relationship between the level of SAA (salivary alpha-amylase) and SNS (sympathetic nervous system) produced in response to anxiety. The measurement of SAA (salivary alpha-amylase) may be a non-invasive method for measuring anxiety (7, 8).

According to the importance of the effect of anxiety on maternal mental health, this study aimed to survey the level of anxiety and salivary alpha-amylase in mothers whose children were hospitalized in the NICU and to survey the relationship between anxiety and alpha-amylase levels.

### Materials & Methods

The present study was a descriptive-analytical cross-sectional study. The study samples were all mothers whose infants were admitted to the NICU ward of Besat Hospital in Hamadan in 2021. The sampling method was census. Finally, 30 mothers with hospitalized infants were included in the study. Inclusion criteria included mothers whose infants had to be hospitalized in the NICU for at least one week; infant patients' clinical conditions were equal. Exclusion criteria included mothers with a history of psychological or systemic illness, mothers who smoked, and mothers with acute infection.

After obtaining written consent, the Hamilton questionnaire measured mothers' anxiety during the infant's hospitalization period. Then saliva samples were taken from all individuals according to the following method. The method of saliva collection in these mothers was non-stimulatory. To avoid 24-hour changes, the researchers collected samples between 9 and 10 a.m. Mothers were prohibited from eating, drinking, mouth washing, and brushing for 2 hours prior to saliva collection and were instructed to refrain from talking while collecting saliva. Each mother was then asked to rinse their saliva directly into the graduated test tube for 5 minutes after rinsing their mouth with water and swallowing saliva. After recording the onset of time, Each subject sat in a sitting

position at a 45-degree angle to the head and neck, swallowed saliva, then closed his lips and poured saliva into an empty container 1 to 2 times per minute to 5 minutes. Finally, saliva samples were sent to the laboratory to check the level of salivary alpha-amylase in mothers.

The present study used a questionnaire to investigate the objectives of the study. This questionnaire consisted of two parts. The first part included demographic information, such as the mother's age, mother's education level, child's gender, place of residence, number of children in the family, and child's birth rank. The second part included the Hamilton questionnaire.

The Hamilton Scale includes 14 items, each related to specific anxiety symptoms. The researcher scores this test; each item is evaluated by a 5 points Likert scale, scored from zero to four depending on the severity of the symptoms. Zero indicates the absence of anxiety, and four equivalents indicate very severe anxiety in the patient. The reliability of the Hamilton questionnaire, using the test-retest, is reported at 0.81 during other studies.

After summing all the scores of 14 questions, a general score is obtained based on which this study determined the severity of maternal anxiety (score zero to 14 mild anxiety, score 14 to 28 moderate anxiety, score 28 to 42 severe anxiety, scores 42 to 56 very severe anxiety) (9, 10).

Demographic characteristics of mothers, along with the Hamilton questionnaire results and salivary alpha-amylase level were entered in the checklist prepared. SPSS software version 21 was used for data analysis. In order to analyze the data, correlation, independent t-test, and one-way analysis of variance (ANOVA) were used. A statistically significant level of 5% was considered.

**Results**

This research investigated thirty mothers. The mean age of the participants was  $29.27 \pm 6.24$  years. Among the mothers, twenty lived in the city (66.66%), and ten lived in the village (33.33%). The mothers' education levels were: 14 (46.66%) undergraduate, eight (26.33%) diplomas, three (9.99%) above diploma, four (13.33%) bachelor, and one (3.33%) higher than a bachelor's.

Fifteen of the studied infants were male, and 15 infants were female. Six infants (20%) were the first child, 15 (50%) were the second child, eight were the third child (26.66%), and one (3.33%) was the fourth child and more.

Anxiety levels in 14 mothers (46.66%) were mild (score 0 to 14), in 14 mothers (46.66%) were moderate (score 14 to 28), and in two mothers (6.66%) were severe (score 29 to 42), and none of the mothers got very severe score (Figure 1).

The mean level of salivary amylase based on anxiety intervals in the Hamilton questionnaire was as follows: According to the one-way ANOVA test, no statistically significant difference was found in different anxiety intervals ( $P = 0.38$ ) (Table 1).

Moreover, no statistically significant relationship was between anxiety and variables of residence, level of education, age range, infant sex, and infant rank.

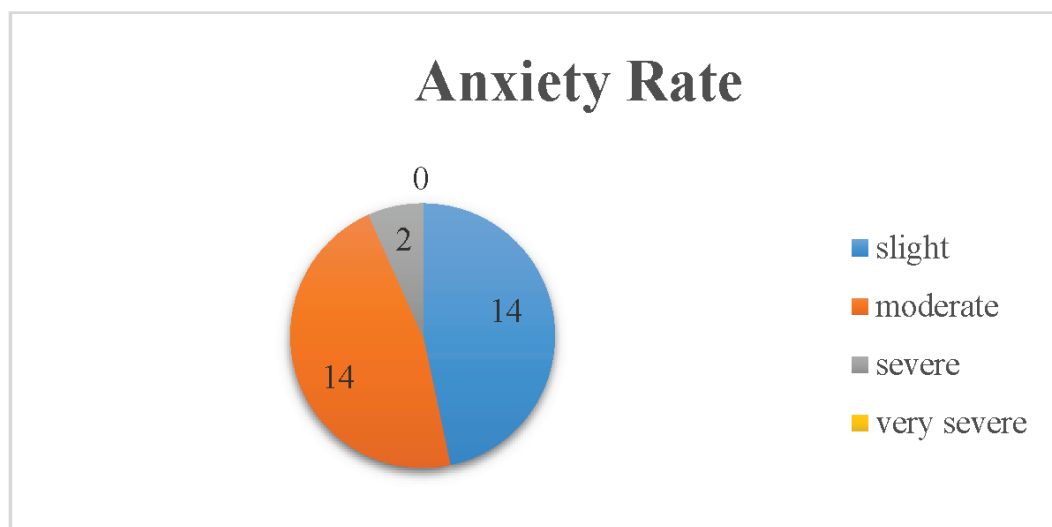


Figure 1: Frequency of participants in terms of anxiety

Table 1. Mean salivary amylase levels based on anxiety intervals

		Count	(U/L) Mean	SD	P Value
Score Range	14-0	14	52.31	49.15	38.0
	28-15	14	32.32	48.17	
	42-29	2	49.48	99.44	
	All	30	02.33	22.16	

## Discussion

This study aimed to survey the level of anxiety and salivary alpha-amylase in mothers whose children were hospitalized in the NICU and to survey the relationship between anxiety and alpha-amylase levels.

According to the results of this study, the highest mean score of maternal anxiety according to the Hamilton questionnaire was in mothers aged 20 to 24 years, and the lowest rate of anxiety was in mothers aged 25 to 29 years. This difference was not statistically significant, meaning no relationship existed between mothers' age and their level of anxiety. According to the present study's findings, the highest anxiety was in mothers with undergraduate education (18.07%) and the lowest in above-diploma mothers (9.33%). This difference was not statistically significant. In other words, no relationship existed between the mother's level of education and their level of anxiety during their child's hospitalization. Anxiety was  $15.00 \pm 6.35$  in mothers with male infants and  $17.53 \pm 7.18$  in mothers with female infants. From this study's findings, it can be concluded that the sex of infants admitted to the NICU does not affect the anxiety of the mothers of these infants. In a similar study that examined the relationship between anxiety and alpha-amylase levels among patients with dental problems, no statistically significant relationship was observed between alpha-amylase and demographic variables such as age, sex, and level of education (11). In another similar study examining the relationship between salivary alpha-amylase and cortisol with social anxiety among children, no statistically significant relationship was observed between salivary variables of age, sex, and stage of puberty with salivary alpha-amylase, which was consistent with the present study (12). However,

in one study, the level of anxiety among students was examined. There was a statistically significant relationship between the demographic variables of age and gender, and anxiety level. The differences in the objectives of the two studies could cause this discrepancy in the results (13).

The mean anxiety score in the Hamilton questionnaire in mothers living in urban areas was  $16.00 \pm 6.32$  and in rural mothers was  $16.80 \pm 7.96$ , which was not statistically different. Correspondingly, in examining the child's rank in the family and its relationship with anxiety, the highest mean score of maternal anxiety was based on the Hamilton questionnaire in the third child (16.88) and the lowest in the fourth child and above (13.00). However, based on the findings, this difference was not statistically significant, and it can be said that there is no relationship between the rank of infants in the family and their mother's anxiety. Finally, this study's results showed no statistically significant relationship between all demographic variables and anxiety.

In the present study, according to the Hamilton questionnaire, 46.66% of mothers had slight anxiety, 46.66% had moderate anxiety, and 6.66% had severe anxiety. The highest salivary alpha-amylase level was in the group, with severe anxiety with a mean of  $48.49 \pm 44.99$ , and the enzyme in the group with slight anxiety was  $15.49 \pm 31.52$ , not statistically significant despite a clear difference in mean. Overall, the mean anxiety score in the Hamilton questionnaire was  $16.27 \pm 6.78$ , and their mean salivary amylase level was  $16.22 \pm 33.02$  U / L, which were not statistically significant (Pearson correlation coefficient) ( $N=30$ ,  $P=0.399$ ,  $r=0.16$ ). This finding is consistent with Yorbik et al. (2016) study. Yorbik's study found no association between anxiety scores and salivary alpha-amylase (14).

Contrary to the results of the present study, Laura Capranica et al. (2016) stated that salivary alpha-amylase levels could be a reasonable indicator of pre-competition stress and strain in adolescent taekwondo practitioners (15). Similarly, Afrisham et al. (2015) in their study stated that stressful conditions such as testing increase salivary alpha-amylase levels (16). These findings were also in contradiction with studies that have been performed to investigate the relationship between stress and salivary amylase levels, and therefore this difference in results may be due to demographic differences in the study samples (17-19).

### **In Conclusion**

According to the results of the present study, no significant relationship existed between the level of alpha-amylase and the level of anxiety in mothers of neonates admitted to the NICU. Thus, further studies in similar populations in anxious conditions seem necessary to obtain comprehensive results. Analytical studies and clinical trials with more samples in similar populations are recommended to prove the causal relationship between anxiety and increased alpha-amylase enzyme.

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### **Author's contribution**

Fatemeh Eghbalian: Head

Mohammad Ali Seif Rabiei: Data Analysis

Mohammad Ahmadpanah: Scientific Advisor

Parya Khadem, Mohammad hasan Saati: Data Gathering

### **Conflict of interest**

None

### **References**

1. De Rouck S, Leys M. Information needs of parents of children admitted to a neonatal intensive care unit: A review of the literature (1990–2008). *Patient education and counseling*. 2009;76(2):159-73.
2. Peyrovi H, Mosayebi Z, Mohammad-Doost F, Chehrzad M-M, Mehran A. The effect of empowerment program on “perceived readiness for discharge” of mothers of premature infants. *The Journal of Maternal-Fetal & Neonatal Medicine*. 2016;29(5):752-7.
3. Gath A. The impact of an abnormal child upon the parents. *The British Journal of Psychiatry*. 1977;130(4):405-10.
4. Johnston C, Mash EJ. Families of children with attention-deficit/hyperactivity disorder: review and recommendations for future research. *Clinical child and family psychology review*. 2001;4(3):183-207.
5. Goodman JH. Paternal postpartum depression, its relationship to maternal postpartum depression, and implications for family health. *Journal of advanced nursing*. 2004;45(1):26-35.
6. Kuo DZ, Houtrow AJ, Arango P, Kuhlthau KA, Simmons JM, Neff JM. Family-centered care: current applications and future directions in pediatric health care. *Maternal and child health*

- journal. 2012;16(2):297-305.
7. Scannapieco FA, Torres G, Levine MJ. Salivary  $\alpha$ -amylase: role in dental plaque and caries formation. *Critical Reviews in Oral Biology & Medicine*. 1993;4(3):301-7.
  8. Nater UM, Rohleder N. Salivary alpha-amylase as a non-invasive biomarker for the sympathetic nervous system: current state of research. *Psychoneuroendocrinology*. 2009;34(4):486-96.
  9. Thase ME, Hersen M, Bellack AS, Himmelhoch JM, Kupfer DJ. Validation of a Hamilton subscale for endogenomorphic depression. *Journal of Affective Disorders*. 1983;5(3):267-78.
  10. Kavyani H, Mossavi A, Mohit A. *Psychological Scales and Interviews (Farsi)*. Tehran, Iran: Sana Publications. 2001:179-205.
  11. Jafari A, Pouramir M, Shirzad A, Motalebnejad M, Bijani A, Moudi S, et al. Evaluation of salivary alpha amylase as a biomarker for dental anxiety. *Iran J Psychiatry Behav Sci*. 2018;12(1):e9350.
  12. Payne LA, Hibel LC, Granger DA, Tsao JC, Zeltzer LK. Relationship of salivary alpha amylase and cortisol to social anxiety in healthy children undergoing laboratory pain tasks. *Journal of child and adolescent behavior*. 2014;2.
  13. Bayram N, Bilgel N. The prevalence and socio-demographic correlations of depression, anxiety and stress among a group of university students. *Social psychiatry and psychiatric epidemiology*. 2008;43(8):667-72.
  14. Yorbik O, Mutlu C, Ozturk O, Altinay DK, Tanju IA, Kurt I. Salivary alpha amylase levels in youths with anxiety disorders. *Psychiatry research*. 2016;235:148-53.
  15. Capranica L, Condello G, Tornello F, Iona T, Chiodo S, Valenzano A, et al. Salivary alpha-amylase, salivary cortisol, and anxiety during a youth taekwondo championship: An observational study. *Medicine*. 2017;96(28).
  16. Afrisham R, Sadegh-Nejadi S, SoliemaniFar O, Abromand M, Kooti W, Najjar Asl S, et al. Evaluating the salivary alpha-amylase level under psychological stress and its relationship with rumination and the five personality traits. *Journal of Mazandaran University of Medical Sciences*. 2015;25(126):22-33.
  17. Ng V, Koh D, Wee A. Salivary alpha amylase levels under conditions of extreme examination stress. *Psychological reports*. 2008;103(2):455-8.
  18. Schoofs D, Hartmann R, Wolf O. Neuroendocrine stress responses to an oral academic examination: No strong influence of sex, repeated participation and personality traits. *Stress*. 2008;11(1):52-61.
  19. Bosch JA, Brand HS, Ligtenberg TJ, Bermond B, Hoogstraten J, Amerongen AVN. Psychological stress as a determinant of protein levels and salivary-induced aggregation of *Streptococcus gordonii* in human whole saliva. *Psychosomatic medicine*. 1996;58(4):374-82.

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