

## Relationship between Stress and Alpha Amylase Levels among Presenters

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### ABSTRACT:

**Background:** Stress occurs when the internal homeostasis is disrupted by external environmental stressors. Salivary Alpha Amylase (sAA) levels are increased in response to stress, and it is frequently used as a biomarker of stress.

**Aims:** To determine the relationship between stress and alpha-amylase levels among presenters by the difference in levels scored in pre and post presentation.

**Material and Methods:** A cross-sectional descriptive study was conducted to achieve the objectives of the study and was achieved to determine the association between stress and salivary alpha amylase levels for presenters during the period from 10<sup>th</sup>. Jan, 2022 to Jan, 20<sup>th</sup>. Mar, 2022. The study was conducted by selecting a typical form of non-probability (Purposive) sample. The size of the sample of 70 participants was included in the study.

**Results:** The relationship between stress and the level of alpha amylase concentration was low in post presentation in comparison with pre presentation ( $0.11\pm 0.03$ ) and ( $0.13\pm 0.02$ ) respectively. This result was statistically highly significant according to paired t-test and the P-value was  $< 0.001$ . Furthermore, the level of Alpha amylase activity was low in post presentation in comparison with pre presentation ( $289658.5\pm 76490.7$ ) and ( $321693.9\pm 52219.2$ ) respectively. This result was statistically highly significant according to paired one group t-test and the P-value was  $\leq 0.0001$ .

**Conclusion:** It was concluded Stress was elevated in the post-presentation in compare with the pre-presentation. and the levels of alpha amylase were affected by the presenters' age groups.

**KEYWORDS:** Stress, alpha amylase, Presenters and HPA.

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### INTRODUCTION

Stress is described as a state where the internal homeostasis is interrupted as a result of real or perceived external environmental pressures. This could be environmental, biological, emotional, social, or psychological factors that cause the body to attempt to reestablish homeostatic equilibrium, a process known as stress reaction [1]. Stress occurs when an individual perceives an incompatibility between the psychological or physical demands of a situation and his or her psychological, biological, or social resources. Furthermore, it occurs when the individual believes that environmental demands are onerous or outstripping his or her ability to adapt [2]. Salivary alpha-amylase has been determined as a possible biomarker for the autonomic nervous system's response to stress, particularly sympathetic activity [3]. The sympathetic component of the stress reaction Although salivary AA, a digestive enzyme included principally in the degradation of starch molecules in the oral cavity, is not a direct by-product of the somatic nervous system, several studies have reported increased levels of salivary AA in response to physical stress (heat, cold and daily exercise). Furthermore, it was found that these sAA. levels are linked to general NE alterations in response to stress in psychological (written assessments).. Additionally, some research has investigated the activity of sAA in response to laboratory stressors such as the Trier Social Stress Test (TSST) and the Cold Pressor Task (CPT) [4]. Teaching speaking, particularly for university students, is viewed as an "interesting and difficult task." Indeed, maintaining students' "engaged in speaking" throughout the session needs a number of tactics. The author highlights that one of the most effective ways to support students in reaching the lesson's objectives is a thorough introduction of the new language at the lesson's early phase, dubbed "the presentation phase." Following that, pupils require "ample activities" to aid them in practicing the new language; this is referred to as the "practice phase." Lastly, pupils must practice communicating with one another in their newly acquired language: this is referred to as "the production phase." [5]. Public speaking is a common and necessary skill in today's business; ideas are shared, people are persuaded, and facts are presented in many aspects of professional life . Stress is linked to a wide array of health problems, including infectious infections, cardiovascular disease, hypertension, diabetes, obesity, , cancer atherosclerosis, and

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neurological diseases. Speaking stress results in communication breakdowns, which affect an individual's personal, social, and emotional well-being [6]. The goal of the current study: to determine the relationship between stress and alpha-amylase levels among presenters by the difference in levels scored in pre and post presentation and to measure the alpha-amylase, and Stress Levels among Presenter before (20) minute Presentation.

### METHODOLOGY

A cross-sectional descriptive study was conducted to achieve the objectives of the study and was conducted to determine the association between stress and alpha amylase levels for presenters during the period from 10<sup>th</sup>. Jan, 2022 to 20<sup>th</sup>. Mar, 2022. The study was conducted by selecting a typical form of non-probability (Purposive) sample. The size of the sample of 70 participants was included in the study. The setting of the study Continuous Education Center at University of Kufa in AL-Najaf AL-Ashraf City.

#### Salivary Alpha Amylase:

**A. Principle:** Alpha Amylase catalyzes the hydrolysis of alpha -1,4-glycosidic bonds in starch, resulting in the formation of reducing sugars such as maltotriose, glucose, dextrin and maltose. The activity of amylase is determined by the rate at which the absorbance at 540 nm increases. Although -AL is thermostable, it can be passivated for 15 minutes at 70°C.

#### B. Kit Salivary Alpha Amylase Components

Components	Character
Reagent I:40 mL×1	If yellow crystals have precipitated, they should be gently heated to dissolve before using.
Reagent II: Powder×1	When the solution is to be used, add 20 mL distilled water, place in room temperature water, heat with frequent agitation, and bring to a boil to completely dissolve the powder..
Standard: Powder×1,10mg anhydrous glucose.	When the solution is to be used, add 1 mL of distilled water to create a 10 mg/mL glucose standard solution. Dilute the glucose standard solution to 0.2, 0.1, 0.05, 0.025, 0.0125, 0.00625 mg/mL with distilled water.

#### C. Procedure:

- 250 µl of the sample was boiled for 5 min and used as a control tube.
- Add reagents as follows:

Reagent (µL)	Control tube (C)	Test tube (T)	Standard tube (S)	Blank tube (B)
Stock solution of alpha amylase	50 (Boiled sample)	250	-	-
Distilled water	-	-	-	250
Standard Solution(mg/mL)	-	-	250	-
Incubate for 15 minutes in a 70°C water bath, then cool.				
Reagent II	-	250	-	-
Incubate for 5 minutes in a 40°C thermostat water bath.				
ReagenI	500	500	500	500
Reagent II	250	-	250	250

\*Appreivation  $A_{\text{control tube}}=A(C)$ ,  $A_{\text{blank tube}}=A(B)$ ,  $A_{\text{test tube}}=A(T)$ ,  $A_{\text{Standard tube}}=A(S)$ ,  $\Delta A(T)=A(T)-A(C)$ ,  $\Delta A(S)=A(S)-A(B)$ .

- Mix thoroughly, place in a 90°C water bath for 10 minutes, and then use a spectrophotometer to determine the absorbance at 540 nm.

### RESULTS

(Table 1.) shows that the highest percentage of the age group (30-39) years old with (48.57%), females participants (58.57 %), ,who are married (64.29%) ; and who are living in urban area (97.14 %) ; with bachelor education level (51.43 %). (Table 2.)

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shows that the about (10%) of presenters in the current study have high stress in the pre presentation, while about (42.86%) of them have high stress in the post presentation. (Table 3.) shows that stress assessment in Post presentation was higher than those in Pre presentation for overall stress items, this result was statistically high significant according to paired t-test and P-value was  $\leq 0.001$ . (Table 4.): shows that the level of alpha amylase concentration was low in post presentation in comparison with pre presentation ( $0.11 \pm 0.03$ ) and ( $0.13 \pm 0.02$ ) respectively. This result was statistically highly significant according to paired t-test and the P-value was  $< 0.001$ . Furthermore, the level of Alpha amylase activity was low in post presentation in comparison with pre presentation ( $289658.5 \pm 76490.7$ ) and ( $321693.9 \pm 52219.2$ ) respectively. This result was statistically highly significant according to paired t-test and the P-value was  $\leq 0.0001$ .

**Table 1. Frequency distribution of studied sample according to Demographic Data.**

Demographic data		Freq.	%
Age groups (Years)	$\leq 29$	25	35.71
	30 – 39	34	48.57
	40 – 49	8	11.43
	50 and more	3	4.29
	Mean $\pm$ SD(Range)	32.59 $\pm$ 7.8(22-65)	
Gender	Males	29	41.43
	Females	41	58.57
Marital status	Single	22	31.43
	Married	45	64.29
	Divorced	3	4.29
	Widowed	0	.00
	Separated	0	.00
Monthly income	Enough	49	70.00
	Enough to some extent	15	21.43
	Not enough	6	8.57
Residence	Urban	68	97.14
	Rural	2	2.86
Educational level	Bachelor	36	51.43
	High Diploma	0	0.00
	Master	17	24.29
	Doctorate	15	21.43
	Post-doctoral	2	2.86
Total		70	100%

**Table 2. Frequency distribution of Stress overall Questionnaire Items pre and post presentation.**

Stress overall Questionnaire Items		Pre Presentation		Post Presentation	
		Freq.	%	Freq.	%
Stress Assessment	Low stress	63	90.00	40	57.14
	High stress	7	10.00	30	42.86
Total		70	100%	70	100%

\*Cutoff value=2; when mean of score  $\leq 2$  stress is low, when mean of score  $> 2$  stress is high

**Table 3. Comparisons of Overall Stress Questionnaire Items between pre and post presentation using paired t test.**

Overall Stress Questionnaire Items	Pre Presentation			Post presentation			T-test (df=69)	P-value (Sig.)
	Mean	SD	Assess.	Mean	SD	Assess.		

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Overall Stress Items	1.74	0.29	Low	1.89	0.36	Low	-4.933	0.001**
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Abbreviations: \*\*=significant at the p-value  $\leq 0.01$ .

**Table 4. Comparison of Alpha amylase Concentration, and Alpha amylase Activity between pre and post presentation using paired t test.**

Saliva laboratory parameters	Pre presentation (n=70)	Post presentation (n=70)	T-test (df=47)	P-value (Sig.)
	Mean±SD	Mean±SD		
Alpha amylase Concentration (mg/ml)	0.13±0.02	0.11±0.03	3.617	0.001
Alpha amylase Activity (IU/ml)	321693.9±52219.2	289658.5±76490.7	3.655	<0.0001

## DISCUSSION

Throughout the course of the current research's data analysis, the results in (Table 1.) showed that most participants in the age group (30-39) years old had (48.57%). A previous study conducted in USA of America discovered that the age group (18-65) years was largely male [7]. Another study was conducted at Florida State University found that 31% of Americans aged (55-64) years were comfortable with public speaking, contrast to only 13% of 18-24-year-olds. However, this difference could be due to the sample selection procedure used [8]. In terms of gender, the current study included mostly males (41.43%) and females (58.57%) who were married (64.29%). A recent study in University of the West of England/ Continuing Education Center, reported in which 7 males (15%), 33 females (72%), and 6 participants (13%) did not report their gender, that selection of the sample with the highest percentage was female in studied [9]. Furthermore, the highest percentage in the studies above is agreed between the percentages, since the selection of the sample for both gender ranges from females to the highest percentage in the selection of samples, but the difference in the number sample size. For residence, presenters were living in urban area (97.14 %); with bachelor education level (51.43 %), this results agreed with, survey about underscores the critical need for additional models and a theoretical framework that explain the non-linear, bi-directional, and dynamic nature of stress and PA interactions. Currently, there are either a dearth of theoretical models of stress and behavior or they are highly contextualized (e.g., work environments, urban life)[10]. According to (Table 2.), approximately 10% of presenters in the current study had high levels of stress before presentations, and Post-presentation stress was equally high (42.86 %). A previous study of University of Murcia in Spain, reported that the stress in the post presentation was high stress. These result was consistent with current study, which showed a real stressful situation after public oral presentations the speech was part of the final examination, and participants had to speak in a different language [11], and another previous study, conducted at Sweden's Institute of Stress Medicine/Department of Medical and Health Sciences, revealed that stress-related exhaustion developed as a result of long-term stress exposure to reflect general levels of psychological stress, and that overall stress ratings were categorized into low and high stress [12]. (Table 3.) shows that post-presentation stress assessments were greater than pre-presentation stress assessments for overall stress items, this result was significant by using the paired one group t-test. According to a previous study reported, Under conditions of stress, cold or sweaty hands and feet, shaking hands, legs, or body, frequent blush, sweating, excessive thirst, and difficulty swallowing can develop. Muscle may be unable to relax. Headaches, shoulder and back pain, repetitive motion, paralysis, awkward movements, stomach pain, and body aches are all symptoms of tight muscles. Over time, this can trigger an unhealthy cycle in which individuals stop exercising and rely on pain medication for relief. These findings corroborate those from a previous study in Iraq, where participants reported being constantly stressed, experiencing sweaty or cold hands and feet, frequent blushing, dry mouth, sweating, and difficulty swallowing. Muscles may be unable to relax. Muscle tension contributes to headaches, back and shoulder discomfort, and general aches [6]. While another study conducted in Iraq found that students who are fearful of public speaking frequently exhibit a number of symptoms during a public performance setting, including palpitations, perspiration, gastrointestinal discomfort, diarrhea, muscle tension, and confused states [13]. (Table 4.) shows that the level of concentration of Alpha amylase was lower in Post presentation than in Pre presentation (0.11±0.03) and (0.13±0.02), respectively. According to the paired t-test, this result was statistically high significant, with P-value was  $< 0.001$ . Furthermore, the level of Alpha amylase activity was significantly lower post presentation than pre presentation (289658.5±76490.7) and (321693.9±52219.2), respectively. According to the paired t-test, this result was statistically highly significant, with P-value was  $\leq 0.0001$ , the current findings were consistent with previous study in Spain, which revealed that when both groups were compared, sAA levels were significantly higher in the speech group at 5 minutes ( $P < .05$ ) than in the control group. sAA increased at 0 minutes and peaked at 1.58-fold soon after the presentation in the speech group (5 minutes). Then, during the resting time (20 and 35 minutes), sAA

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activity reduced considerably [11]. The response in time sAA (peaking at 5 minutes) seen in this study implies a distinct activation pattern for the SNS and HPA axis during the stress response. The SNS responds more rapidly than the HPA axis, which is consistent with earlier published research. It's worth noting that sAA was already elevated (0.42-fold) before the presentation in comparison to the resting time. Similarly to the cortisol data, these findings could be interpreted by the fact that students were already stressed due to their prior understanding of the task [11].

### CONCLUSION

It was concluded that oral presentation of the participants has increased their stressful situation. Alpha amylase levels were elevated after doing the presentation compared to its levels before presentation.

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**CONFLICT OF INTEREST:** Nil

### ETHICAL APPROVAL:

This case-control study was approved by the medical ethics committee in the Faculty of Medicine/Kufa University (Reference#: MEC-15 on November 8, 2021).

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**AUTHOR CONTRIBUTIONS:** Ali Majid Hassana : Corresponding author, data collection, manuscript concept, writing, results analysis. Ali J. Eidan : manuscript submission, revision and gallery proof. Muradha Kanim Adea : Data collection, manuscript concept and writing.

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