Original research papers

Salivary Alpha-amylase Activity Level in ST-segment elevation myocardial infarction Patients

Abstract

Background and Objective: Sudden death is the main cause of mortality and disability in patients with coronary artery disease or myocardial infarction. The aim of this study was to evaluate the activity level of salivary alpha-amylase to predict malignant ventricular arrhythmias in ST-segment elevation myocardial infarction (STEMI) patients.

Material and Methods: In this analytical cross-sectional study, 42 patients with STEMI who referred to Imam Reza Hospital participated. First, salivary amylase was taken from all STEMI patients and then these patients were divided into two groups of patients with malignant ventricular arrhythmia or without malignant ventricular arrhythmia during 72 hours.

Results: A total of 42 patients were included in the study out of which 30 (71.4%) were females and 12 (28.6%) males. The average salivary amylase in patients was 118/41 ± 96/87. There was no significant difference in the frequency of diabetes, blood pressure, blood lipids, ischemic heart disease, and involvement severity in both groups with arrhythmia and lack of arrhythmias (P> 0.05). Also there was no significant difference in systolic and diastolic blood pressure, respiratory rate, heart rate, oxygen saturation, blood glucose, temperature and severity of infarction (P> 0.05). However, the two groups were different in terms of salivary amylase levels. Salivary amylase levels were significantly higher in arrhythmic group than in the non-arrhythmic group (P< 0.001).

Conclusions: Our result shows that there is a difference in the concentrations of salivary Alpha-amylase activity level in with and without ventricular arrhythmias groups.

Key words: Ventricular arrhythmia, salivary amylase, STEMI.

Introduction

Sudden death is the main cause of mortality and disability in patients with coronary artery disease or myocardial infarction [1]. In spite of the rapid and complete treatment of patients with acute myocardial infarction (AMI), mortality is still high in these patients [2]. Ventricular arrhythmias include ventricular fibrillation (VF) and ventricular tachycardia ((V-tach or VT) are the most commonly reported causes of death in AMI patients, Therefore, it is very clear that preventive and therapeutic strategies should be implemented in such diseases [3,4]. The prevalence of ventricular arrhythmias in patients with AMI was reported to be "between" 9.1% to 10.2%, which is also higher in the early hours after the MI. Although the pathogenesis and stimuli of the development of ventricular arrhythmias are very different according to the incidence, but the autonomic nervous system activity can be a

very important factor in the initiation of ventricular arrhythmias during AMI [5,6]. Previous studies have shown that the effect of increasing the activity of the sympathetic system on the heart and subsequently stimulating the secretion of catechol amines and the formation of ischemia can be one of the important factors in the development of ventricular arrhythmia, which has an any arrhythmic effect on the ventricles in the presence of parasympathetic system [7,8]. Epidemiological studies have indicated that about 25% of patients with coronary artery disease have suddenly died of psychological stress. Because acute psychological stress can act as an activator of the sympathetic system and cause myocardial ischemia, left ventricular dysfunction, rhythm disorder, and consequently facilitate sudden death [9]. The interest in measuring oral fluids for the diagnosis of diseases has recently increased because of its non-invasiveness and convenient sampling. Neuroendocrine markers such as salivary alpha-amylase (SAA) play a key role as an indicator of the human body in the face of an acute stressful event [10]. The salivary gland contains beta-adrenergic receptors where norepinephrine, secreted from the sympathetic nerve endings, promotes the activity of the salivary glands on adrenergic receptors. As a result, it increases the ratio of protein to fluid in the saliva, from which salivary alpha-amylase is produced by salivary gland cells [11,12]. Alpha amylase activity is a reflection of changes in catechol amines. It can therefore be used as a non-invasive and easy-to-measure sympathetic system activity marker. The aim of this study is to determine whether the level of salivary alpha-amylase activity is a predictor of malignant ventricular arrhythmias in STEMI patients.

Material and Methods

This analytical cross-sectional study was performed on patients referred to Imam Reza Hospital. Therefore, patients with STEMI diagnosis were selected for sampling. Patient satisfaction has been incorporated into study, the saliva was taken and immediately sent to the laboratory for measuring its alpha amylase. In addition to the initial assessment of vital signs, the presence of diabetes and smoking, patients were next followed up. Individuals was classified as having malignant ventricular arrhythmias and divided into 2 groups:

1. Patients who develop malignant ventricular arrhythmias within the first 72 hours of admission.

2. Patients who do not have malignant ventricular arrhythmias within the first 72 hours of admission.

Whenever illness occurs in a group of malignant arrhythmias, it was tried to have two matched controls for cigarette, diabetes, age, sex, initial blood pressure and initial heart rate from a group that has no malignant arrhythmia. As a result, the sample size was obtained from the cases and controls and finally the data were analyzed as blind.

Sampling method:

The saliva samples were collected by spitting inside the special tubes from the mouth water that is secreted without stimulation. Before collecting samples, the patient was asked to wash mouth with water. Only one sampling at the time of arrival of the patients was done in

the emergency department. The specimens were sent to the laboratory at a temperature below zero, and kept there until use. Alpha Amylase activity is quantitatively measured by a specific kit.

Inclusion and Exclusion criteria

Inclusion criteria: All patients who were diagnosed with STEMI.
Exclusion criteria: 1. Any oral illness 2. Patients diagnosed with ACS at the other centers and then referred to the center for treatment.

Data analysis

Data were entered into SPSS software after proper design and descriptive analyzes were performed to measure the mean and incidence in each group using parametric t-test. If needed, a nonparametric test (Mann-Whitney) was used to compare the mean in different groups. Possible confounding factors such as cigarette smoking, diabetes and primary vital signs were evaluated in two groups and then, if observed, significant differences wereobserved, they were adjusted by multivariate analysis. P <0.05 was considered statistically significant with 95% confidence interval.

Ethical considerations

Because the taking of a salivary sample is not invasive, the patient only receives oral satisfaction. To protect the patient's secrets after collecting information, the patient's name was removed from the checklist header, where only the patient's file number was remained available for data retrieval. The executives were committed to the 26 moral codes in all phases of the investigation.

Results

In this research, 42 patients were enrolled in Imam Reza Hospital with STEMI. 71.4% (30 patients) were male and 28.6% (12 patients) were female. The mean age of patients was 67.1 ± 9 years with a minimum of 47 and a maximum of 80 years. The mean systolic and diastolic blood pressure of patients at the time of referral was 145.05 ± 14.64 and 85.26 ± 8.13 mm Hg, respectively. Also, the mean respiratory rate and heart rate in patients were 16 ± 3 and 84 ± 13, respectively.

The mean of oxygen saturation (91.71 ± 3.75%), blood glucose (171.88 ± 34.54), and temperature (36.86 ± 0.27 ° C) were determined. The average salivary amylase in patients was 118.41 ± 96.87 µl / ml. The frequency of diabetes, hypertension and high blood lipids was 59.5% (25 patients), 40.5% (17 patients) and 71.4% (30 patients) respectively. Furthermore, 27 (64.3%) patients suffered from ischemic heart disease and 57.1% (24 people) were smokers. The most common places for the infarction were determined including the lower limbs (42.1%; 16 patients), lateral (21.1%; 8 cases), posterior (21.1%; 8 cases) and anterior (15.8%; 6 cases). The location of the infarction was unknown in 4 patients (Diagram 4-3). Moreover, the severity of infarction was based on the number of affected areas in patients where an area with a frequency of 63.2% (24 people) and two

regions with frequency of 33.3% (14 cases) were determined in the study. Four people lacked enough information on the severity of the infarction.

Patients were divided into two groups without arrhythmia (20 cases, 47.6%) and arrhythmia (22 cases, 52.4%) based on the presence or absence of arrhythmia. In the group without arrhythmia, the frequency of men and women was 80% (16 cases) and 20% (4 cases) respectively. While in the groups with arrhythmia, 63.6% (14 cases) was male and 36.4% (8 cases) was women. The x^2 test showed no significant difference between the two genders ($p > 0.05$). In the group without arrhythmia, the mean age of the patients was 65.7 ± 7.78 years with a minimum of 55 and a maximum of 79 years. The mean systolic and diastolic blood pressure of the patients at the time of referral were estimated to be 142.7 ± 11.22 and 85.3 ± 5.37 mmHg, respectively. Furthermore, the mean respiratory rate and heart rate in patients were determined as 15 ± 3 and 84 ± 10, respectively. In addition, the mean oxygen saturation (91.3 ± 3.85%), blood glucose (172.32 42.32), and temperature (36.78 ± 0.16) were determined. The average salivary amylase in patients was 59.32 ± 44.13 µ/ ml. Frequency of diabetes, high blood pressure and high blood lipids was 75% (15 patients), 40% (8 patients) and 65% (13 people), respectively. Also, 10 patients (50%) suffered from ischemic heart disease and 40% (8 patients) consumed cigarette. In this group, the severity of infarction was based on the number of affected areas in the patients, including an area with a frequency of 55.6% (10 patients) and two areas with 44.4% frequency (8 patients). Two patients did not have enough information about severity of infarction. In the group with arrhythmia, the mean age of the patients was 88.36 ± 10.14 years with a minimum of 47 and a maximum of 80 years. The mean systolic and diastolic blood pressure of the patients at the time of referral were 147.18 ± 17.16 and 85.23 ± 10.16 mm Hg, respectively. Moreover, the mean respiratory rate and heart rate of patients were 16 ± 3 and 84 ± 16, respectively. The mean oxygen saturation (92.09 ± 3.7%), blood glucose 171.36 ± 26.26), and the temperature (36.94 ± 0.33) were calculated in the present study. The average salivary amylase in patients was 174.55 ± 100.56 µ/ ml. The frequency of diabetes, hypertension and high blood lipids was 45.5% (10 subjects), 40.9% (9 patients) and 77.3% (17 patients), respectively. Furthermore, 17 patients (77.3%) suffered from ischemic heart disease and 72.7% (16 patients) were registered as smokers. In this group, the intensity of the infarction was determined based on the number of affected areas in the patients, including an area with a frequency of 70% (14 individuals) and two regions 30% (6 individuals).

Chi square test showed that there was a significant difference in the frequency of diabetes ($P = 0.06$), blood pressure ($P = 0.60$), blood lipids ($P = 0.49$), cardiac ischemic disease ($P = 0.1$), smoking ($P = 0.06$), and intensity of conflict ($P = 0.5$) between two groups with/or without arrhythmia. There was no significant difference in age between two groups with/or without arrhythmia using t-test ($P > 0.05$).

Moreover, t-test showed that there was no significant difference in terms of systolic and diastolic blood pressure, respiratory rate, heart rate, oxygen saturation, blood glucose, temperature and severity of involvement between two groups with arrhythmia/or without arrhythmia ($p > 0.05$). However, the two groups were different in terms of salivary amylase levels using t-test. Salivary amylase levels were significantly higher in arrhythmia group

than group without arrhythmia (p = 0.001). Moreover, salivary amylase was significantly higher in women as compared to men (p = 0.02) (Table 1).

Discussion

Nowadays, heart attack is a major cause of death and disability in Iran and other countries. STEMI is a very serious type of heart attack, in which one of the main arteries of the heart is blocked [13]. Diagnostic and acute STEMI care systems have been developed worldwide [14]. An important factor in the development of ventricular arrhythmias is the increase in the activity of the sympathetic system with an effect on the heart [15]. Saliva has been introduced as a diagnostic fluid for many years, and has been introduced in many research studies. Biomarkers have been discovered in the saliva that can detect diseases such as malignancies, connective tissue diseases, oral and dental diseases, and systemic diseases. One of these biomarkers is salivary amylase that can predict sympathetic activity in the body [16-19]. In this study, salivary amylase measurement was used to predict ventricular arrhythmia in patients with myocardial infarction. Patients were divided into two groups including ventricular arrhythmia and without ventricular arrhythmia. In this study, there was no difference in sex between groups and the gender variable did not because errors among other analyzes. The results showed that there was no significant difference in age between two groups with/ or without arrhythmia. As a result, the two groups were divided equally in terms of age. There was no significant difference in systolic and diastolic blood pressure, respiratory rate, heart rate, oxygen saturation, blood glucose, temperature, and severity of infarction in arrhythmic and non-arrhythmic groups, which indicates these variables have no predictive role in arrhythmias and cannot alter the results of alpha amylase. Salivary amylase levels were significantly higher in arrhythmic group than in the non-arrhythmic group, which proves the hypothesis of the role of salivary amylase in predicting the occurrence of ventricular arrhythmias in patients with STEMI. In a study conducted by Shen and colleagues in Taiwan in 2011, salivary alpha-amylase was used for prognosis of malignant arrhythmias and its short-term prognosis, after myocardial infarction, by changing the ST segment. These results were consistent with the present study. Due to the occurrence of a dangerous ventricular arrhythmia followed by sympathetic neuropathic activity, the release of myocardial cytokines and the introduction of alpha amylase as one of the sympathetic activity markers, alpha-amylase was used to prognoses the occurrence of a dangerous ventricular arrhythmia using salivary alpha-amylase. In this study, 91 patients with ST segmental myocardial infarction were divided into two groups including arrhythmic (9 patients) and non-arrhythmic (82) groups and salivary alpha-amylase was then measured. In the group with salivary amylases arrhythmia was significantly higher than the group without arrhythmia (P = 0.04). Moreover, the use of logistic regression test showed that salivary amylase plays an independent role in prognosis of malignant arrhythmias and therefore has a high diagnostic value [20]. The results of the present study are consistent with the results of Shen et al., which confirms the value of salivary amylase in early diagnosis of ventricular arrhythmia. In a prospective cohort study, Shen and colleagues (2012) have reconsidered their previous findings obtained on this topic. They have considered the predictive value of salivary amylase for the diagnosis of acute ventricular infarction in patients with chest pain. In this prospective study, 473 patients with chest pain

were evaluated for salivary amylase up to 4 hours after referral to the emergency department. In this study, salivary amylase was significantly higher in the group with myocardial infarction than in the non-arrhythmic group. In addition, regression analysis indicated that salivary amylase has an independent predictor role. The study also found that the cutting point of 197/7 had the best sensitivity and specificity in predicting the occurrence of an infarction. Salivary amylase as an independent variable can play an effective role in predicting acute myocardial infarction [21]. The second study is also consistent with the results of the first study and confirms our results. Regarding the higher salivary amylase in the arrhythmic group, the results of this study showed that salivary amylase is an important marker in predicting the incidence of arrhythmia in STEMI patients. The use of this method is also easy, non-invasive, inexpensive and fast and can be easily employed in an emergency department. It can also be used to predict the risk of cardiac arrhythmias in these patients and to monitor these patients more precisely in risk groups. However, it is suggested that a prospective cohort study be conducted on focusing on the role of salivary alpha-amylase in early diagnosis of ventricular arrhythmia in patients with STEMI. It is also suggested that additional studies in the semen of heart and serum biomarkers should be performed to determine the diagnostic accuracy of salivary amylase. By determining the cut-off point, the sensitivity and specificity of this method can be obtained so that it will be used as an efficient, non-invasive, cheap and fast method in the future in emergency department of the hospital.

Conclusion

Our result shows that there is a difference in the concentrations of Salivary Alpha-amylase Activity Level in both groups. It is also easy, non-invasive, inexpensive and fast, and easily accessible in an emergency. However, in order to confirm the findings of this study, a prospective cohort study with a higher sample size is needed and that other potential confounders must also be taken into account.

References

1. Henkel DM, Witt BJ, Gersh BJ, et al. Ventricular arrhythmias after acute myocardial infarction: a 20-year community study. American heart journal. 2006;151(4):806-812.
2. Campbell R, Murray A, Julian DG. Ventricular arrhythmias in first 12 hours of acute myocardial infarction. Natural history study. British heart journal. 1981;46(4):351.
3. Thompson CA, Yarzebski J, Goldberg RJ, et al. Changes over time in the incidence and case-fatality rates of primary ventricular fibrillation complicating acute myocardial infarction: perspectives from the Worcester Heart Attack Study. American heart journal. 2000;139(6):1014-1021.
4. Moghadam AJ, Azizinejad S. Study of high sensitive c-reactive protein (HS-CRP) after cardiac rehabilitation program in patients undergoing isolated CABG. International journal of biomedical science: IJBS. 2016;12(4):143.
5. Daniłowicz-Szymanowicz L, Figura-Chmielewska M, Ratkowski W, et al. Effect of various forms of physical training on the autonomic nervous system activity in patients with acute myocardial infarction. Kardiologia Polska (Polish Heart Journal). 2013;71(6):558-565.

6. Sharma AD, Corr PB. Adrenergic factors in arrhythmogenesis in the ischemic and reperfused myocardium. European heart journal. 1983;4(suppl_D):79-90.
7. Du X-J, Cox HS, Dart AM, et al. Sympathetic activation triggers ventricular arrhythmias in rat heart with chronic infarction and failure. Cardiovascular research. 1999;43(4):919-929.
8. Podrid PJ, Fuchs T, Candinas R. Role of the sympathetic nervous system in the genesis of ventricular arrhythmia. Circulation. 1990;82(2 Suppl):I103-113.
9. Blumgart HL, Schlesinger MJ, Davis D. Studies on the relation of the clinical manifestations of angina pectoris, coronary thrombosis, and myocardial infarction to the pathologic findings. American Heart Journal. 1940;19(1):1-91.
10. Maruyama Y, Kawano A, Okamoto S, et al. Differences in salivary alpha-amylase and cortisol responsiveness following exposure to electrical stimulation versus the Trier Social Stress Tests. PLoS One. 2012;7(7):e39375.
11. Miller C, Foley III J, Floriano P, et al. Utility of salivary biomarkers for demonstrating acute myocardial infarction. Journal of dental research. 2014;93(7_suppl):72S-79S.
12. Ramasubbu N, Paloth V, Luo Y, et al. Structure of human salivary α-amylase at 1.6 Å resolution: implications for its role in the oral cavity. Acta Crystallographica Section D: Biological Crystallography. 1996;52(3):435-446.
13. Cecchi E, D'Alfonso MG, Chiostri M, et al. Impact of hypertension history on short and long-term prognosis in patients with acute myocardial infarction treated with percutaneous angioplasty: comparison between STEMI and NSTEMI. High Blood Pressure & Cardiovascular Prevention. 2014;21(1):37-43.
14. Solla DJF, de Mattos Paiva Filho I, Delisle JE, et al. Integrated Regional Networks for ST-Segment–Elevation Myocardial Infarction Care in Developing Countries: The Experience of Salvador, Bahia, Brazil. Circulation: Cardiovascular Quality and Outcomes. 2013;6(1):9-17.
15. Yu L, Zhou L, Cao G, et al. Optogenetic Modulation of Cardiac Sympathetic Nerve Activity to Prevent Ventricular Arrhythmias. Journal of the American College of Cardiology. 2017;70(22):2778-2790.
16. Ghallab NA. Diagnostic potential and future directions of biomarkers in gingival crevicular fluid and saliva of periodontal diseases: Review of the current evidence. Archives of oral biology. 2017.
17. Schumacher S, Kirschbaum C, Fydrich T, et al. Is salivary alpha-amylase an indicator of autonomic nervous system dysregulations in mental disorders?—A review of preliminary findings and the interactions with cortisol. Psychoneuroendocrinology. 2013;38(6):729-743.
18. Kaur J, Jacobs R, Huang Y, et al. Salivary biomarkers for oral cancer and pre-cancer screening: a review. Clinical oral investigations. 2018:1-8.
19. Thoma MV, Kirschbaum C, Wolf JM, et al. Acute stress responses in salivary alpha-amylase predict increases of plasma norepinephrine. Biological psychology. 2012;91(3):342-348.
20. Shen Y-S, Chan C-M, Chen W-L, et al. Initial salivary α-amylase activity predicts malignant ventricular arrhythmias and short-term prognosis after ST-segment elevation myocardial infarction. Emerg Med J. 2011;28(12):1041-1045.
21. Shen Y-S, Chen W-L, Chang H-Y, et al. Diagnostic performance of initial salivary alpha-amylase activity for acute myocardial infarction in patients with acute chest pain. Journal of Emergency Medicine. 2012;43(4):553-560.

294 **Table 1:** Comparison of mean of two groups with and without arrhythmias

arrhythmia		Number	Deviation from criterion	Average	P-value
Amylase	Positive	19	44.13	59.32	**< 0.001**
	Negative	20	100.56	174.55	
age	Positive	20	78.7	65.70	**< 0.16**
	Negative	22	10.14	68.36	
Systolic blood pressure	Positive	20	11.22	142.70	**< 0.16**
	Negative	22	17.16	147.18	
Diastolic blood pressure	Positive	20	5.37	85.30	**< 0.15**
	Negative	22	10.16	85.23	
Number of breaths	Positive	20	2.96	14.85	**< 0.67**
	Negative	22	3.18	16.27	
Heart rate	Positive	20	9.73	83.85	**< 0.09**
	Negative	22	16.55	84.23	
Oxygen Saccharification	Positive	20	3.85	91.30	**< 0.79**
	Negative	22	3.70	92.09	
Blood glucose	Positive	20	42.32	172.30	**< 0.09**
	Negative	22	62.26	171.36	
Temperatures	Positive	20	0.16	36.78	**< 0.24**
	Negative	22	0.33	36.94	

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