Study of Salivary Alpha-amylase Activity Level for Predicting Malignant Ventricular Arrhythmias in STEMI 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 STEMI patients.

Material and Methods: In this Nested case-control 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: Considering the higher salivary amylase in the arrhythmic group, it can be concluded that salivary amylase is a valuable marker in predicting the incidence of arrhythmia in STEMI patients. It is also easy, non-invasive, inexpensive and fast, and easily accessible in an emergency.

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. The salivary gland contain 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 [10-12]. 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 nested case control (NCC) study was performed on patients referred to Imam Reza Hospital. Therefore, patients with STEMI diagnosis were selected for sampling. Patient satisfaction have 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

80 below zero, and kept there until use. Alpha Amylase activity is quantitatively measured by a
81 specific kit.

82 Inclusion and Exclusion criteria

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

86 Data analysis

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

94 Ethical considerations

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

100 **Results**

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

107 The mean of oxygen saturation ($91.71 \pm 3.75\%$), blood glucose (171.88 ± 34.54), and
108 temperature ($36.86 \pm 0.27 \, °C$) were determined. The average salivary amylase in patients
109 was 118.41 ± 96.87 μl / ml. The frequency of diabetes, hypertension and high blood lipids
110 was 59.5% (25 patients), 40.5% (17 patients) and 71.4% (30 patients) respectively.
111 Furthermore, 27 (64.3%) patients suffered from ischemic heart disease and 57.1% (24
112 people) were smokers. The most common places for the infarction were determined
113 including the lower limbs (42.1%; 16 patients), lateral (21.1%; 8 cases), posterior (21.1%; 8
114 cases) and anterior (15.8%; 6 cases). The location of the infarction was unknown in 4
115 patients (Diagram 4-3). Moreover, the severity of infarction was based on the number of
116 affected areas in patients where an area with a frequency of 63.2% (24 people) and two
117 regions with frequency of 33.3% (14 cases) were determined in the study. Four people
118 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 group with arrhythmia, 63.6% were male and women (14 cases) and 36.4% (8 cases), respectively. 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 ($91.3 \pm 3.85\%$), 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 ($92.09 \pm 3.7\%$), 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

Considering the higher salivary amylase in the arrhythmic group, it can be concluded that salivary amylase is a valuable marker in predicting the incidence of arrhythmia in STEMI patients. 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.

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