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Evaluation of salivary alpha-amylase level in patients undergoing surgical extraction of impacted lower wisdom teeth

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Abstract---Objectives: to evaluate changes in the level of salivary alpha amylase following surgical removal of impacted lower wisdom teeth. Methodology: this is a prospective cohort study, which included patients submitted for surgical extraction of impacted lower wisdom teeth, three samples of saliva were taken from each patient (preoperative, 48 hours, and 7 days postoperatively). The concentration of salivary amylase is assessed using Enzyme Linked Immunosorbent Assay (ELISA) kit, the difference between salivary amylase level pre- and postoperatively were assessed and correlated with the variables which are age, gender and side of impaction. Results: 36 impacted lower wisdom teeth were removed from 34 patients (19 females 55.88%, and 15 males 44.12%) with mean age of 24.85 years and a standard deviation of 4.93 years, 19 impacted lower wisdom teeth were in the right side (53%) and 17 (47%) were in the left side. The mean \pm SD of preoperative salivary amylase concentration was 126.2 ± 31.97 , 48 hours postoperatively it was 131.8 ± 48.99 , and 7 days postoperatively it was 127.0 ± 32.09 . The difference between pre- and postoperative salivary amylase level was non-significant. Conclusion: salivary alpha amylase level rises but non-significantly after surgical removal of impacted lower wisdom teeth.

Keywords---Salivary alpha amylase, impacted third molar, salivary enzymes, inflammatory markers, salivary biomarker.

1 Introduction

With a frequency that can reach as high as 70 percent of the population, impacted mandibular wisdom teeth are the most prevalent type of impacted teeth

seen in the mouth (Abdul Kareem & Al Hussaini, 2019). The lack of volume in the dental arches is the primary factor contributing to impaction (Juodzbaly & Daugela, 2013). The surgical extraction of impacted lower third molars with local anesthetic is still the most common surgical treatment done by oral and maxillofacial surgeons (Bui et al., 2003; Yuasa et al., 2002)

Saliva consists of 99% water; it's a much more serious, slightly acidic, clear fluid produced by the salivary glands. Other constituents of saliva are electrolytes, proteins, small organic molecules and hormones (Berkovitz et al., 2017; Edgar, 1992; Humphrey & Williamson, 2001). These components function as follows: (1) saliva's buffer system is provided by phosphates, bicarbonates, and urea; (2) enzymes, proteins and immunoglobulins provide antibacterial action; (3) modulation of remineralization and demineralization by the antisolubility action of phosphates, calcium and proteins; (4) plaque metabolism by mucins and proteins acting by cleaning, aggregating or attaching oral microorganisms (Humphrey & Williamson, 2001).

Salivary alpha-amylase (SAA) is one of the main enzymes (proteins) of saliva, making up 40% to 50% of salivary protein produced by salivary glands. Most SAA is synthesized in parotid glands (80%), and the submandibular gland produces the remainder (de Almeida Pdel et al., 2008). The primary function of SAA is the digestion of carbohydrates by hydrolyzing α -1,4 linkages in polysaccharides and converting them into simpler sugars (Baum, 1993); it also prevents bacterial growth and adherence contributing to mucosal immunity of the mouth (Scannapieco et al., 1993).

Several studies reported an increase in SAA levels and sympathetic function in response to psychological and physical stress conditions, suggesting that the control of SAA secretion is through the autonomic nervous system (ANS) (Filaire et al., 2010; Nater et al., 2006; Nater & Rohleder, 2009; Strahler et al., 2010). A few studies have examined the salivary level of alpha-amylase as a marker for dental anxiety following surgical removal of lower wisdom teeth (Robles et al., 2012). However, the impact of the acute inflammatory process after extraction on the biomarker's readings has not been investigated. This study aimed to see any change in the level of SAA, and if SAA might be used as an inflammatory biomarker after surgical wisdom teeth removal.

2 Materials and Methods

This prospective cohort clinical research was carried out at the University of Baghdad's College of Dentistry's Department of Oral and Maxillofacial Surgery from January 2022 to June 2022. It comprised individuals who had impacted mandibular third molars surgically extracted under local anesthesia. Medically fit patients aged 18 to 37 years old met the inclusion criteria. This study excluded patients with systemic diseases and patients who had associated pathology with the impacted tooth, such as tumors or cysts.

The organization committee for research ethics has authorized this research (project No. 395121); Information about the study is explained to all patients, and informed consent is obtained from them to participate in the study.

Preoperatively, patients were instructed to give 1-2 ml saliva via the passive drooling method to test the preoperative salivary level of α -amylase; the patients were seated in the upright position with their heads tilted forward, allowing saliva to be collected on the floor of the mouth and given a sterile container to spit the accumulated saliva into the container, then the collected saliva is transferred into Eppendorf tube and placed in Eppendorf rack for storage in the freezer at -20°C , until being tested.

A single operator performed all of the surgical operations with local anesthesia (Lidocaine hydrochloride 2% with epinephrine 1:80000) block of inferior alveolar nerve and infiltration for the long buccal nerve. A full-thickness mucoperiosteal flap is reflected, then the surgical extraction was proceeded by using elevators alone or with the removal of bone and sectioning of the tooth through the use of a straight surgical handpiece high-speed turbine, respectively, under copious saline irrigation. After tooth removal is achieved, the area is carefully rinsed and inspected for any sharp margins and then sutured and given a gauze to bite on. Then patients are given instructions not to rinse for 24 hrs. And to take Amoxicillin 500mg cap. Three times daily for five days (if the patient is allergic to penicillin, azithromycin 500mg capsules are given once daily for three days instead of Amoxil), Metronidazole 500mg tab three times daily and analgesics (paracetamol 1g tab and ibuprofen 400mg tab when needed).

Follow-up is performed at forty-eight hours and seven days postoperatively; in each follow-up visit, a sample of saliva was taken from the patients to determine the level of α -amylase by using the same technique for saliva collection described above. The concentration of SAA in preoperative and post-operative samples was measured using an Enzyme-Linked Immunosorbent Analysis (ELISA) kit (Demeditec Diagnostics GmbH, Germany).

Predictor variables were the preoperative SAA concentration and age, sex and impaction side, while; the outcome variable was the post-operative SAA levels measured at 48 hours and seven days after removing the impacted lower wisdom teeth. Version 9 of Graphpad prism windows software (GraphPad Software, San Diego, CA, USA) is used for statistical analysis. Descriptive analysis included percentages (%), or mean \pm standard deviation (S.D.). The differences were considered significant at $P < 0.05$.

3 Results and Discussions

34 patients (19 females {55.88%} and 15 males {44.12%}) underwent surgical extraction of 36 impacted lower wisdom teeth.

The mean \pm standard deviation of the age was 24.85 ± 4.93 years. The distribution of age is shown in figure (1.1).

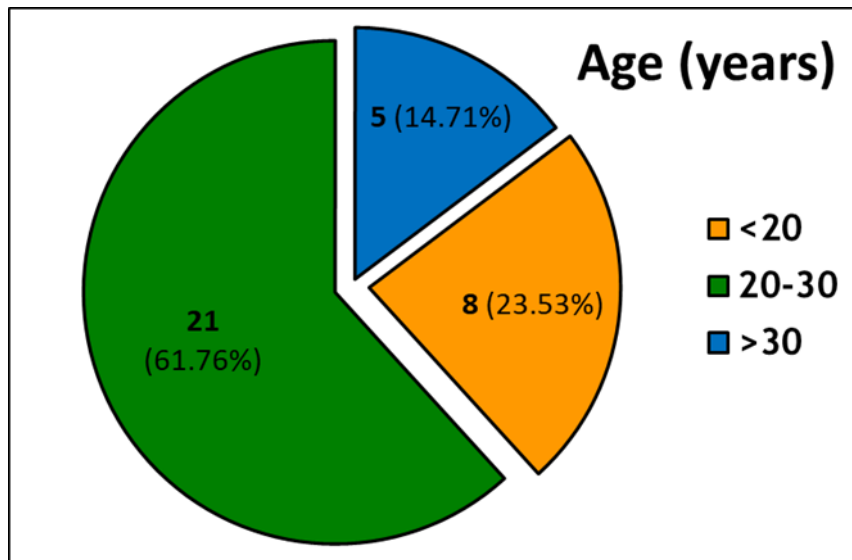


Figure (1.1): age distribution

Of the 36 surgically removed wisdom teeth, 19 were on the right side, representing 53%, and 17 were on the left, representing 47%. The concentration of SAA (preoperatively, 48 hours and seven days postoperatively) are described in table 1.1. There was a non-significant increase in SAA as shown in Table 1.1

Table 1.1: concentration of SAA and the difference between preoperative, 48 hrs. And seven days post-operative

Preoperative	SAA level U/ml		P-value
	48 hrs. post-operative	Seven days post-operative	
126.2 ± 31.97	131.8 ± 48.99	127.0 ± 32.09	0.636 [NS] a

a: Repeated measure ANOVA

NS: non-significant

The correlations of different variables with the changes in SAA level are summarized in Table 1.2.

Table 1.2: correlations of variables with SAA level

Variables	SAA level U/ml			P-value
	preoperative	48 hrs. post-operative	Seven days post-operative	
Gender				
Male	129.5 ± 26.82	134.0 ± 41.00	130.2 ± 26.98	0.1955 a [N.S]
Female	122.1 ± 37.95	129.0 ± 58.79	122.9 ± 38.06	
Age (years)				
<20	124.0 ± 41.38	127.5 ± 34.22	124.7 ± 41.58	0.4918 b [NS]
20-30	128.8 ± 27.23	133.0 ± 45.02	129.5 ± 27.35	

>30	117.8 ± 41.59	121.2 ± 61.44	118.8 ± 41.71	
Side of impaction				
Right	127.4 ± 25.15	134 ± 46.87	128 ± 25.38	0.501
Left	124.8 ± 38.99	129.4 ± 52.61	125.7 ± 39.04	c [N.S]

a: Mann Whitney test.

b: Kruskal-Wallis test.

c: Unpaired t-test.

NS: not significant.

Discussion

The surgical removal of impacted wisdom teeth is one of the most routine surgeries carried out in the mouth (Ravikumar & Narayanan, 2018). The post-operative sequels are pain, swelling and limited ability to open the mouth. Rare complications include alveolar osteitis, nerve damage, infection, or fracture of the mandible.

(Chauhan et al., 2015; Degala & Bathija, 2018) Mentioned that the inflammatory reactions developed postoperatively and reached their maximum values on the second-day post-operation and started to decline to get the seventh day. (López Carriches et al., 2006) Also found that inflammatory markers increased after wisdom tooth surgery and remained high for one week postoperatively.

The present study has more attendance of females than males, which is in accordance with other studies (Saravanakumar et al., 2019; Zafar et al., 2019). Most of the patients in the current study were in the age range of 20-30 years. This is in accordance with other studies (Ayaz, 2012; Krishnan et al., 2009), that revealed that patients in their third decade of life were the most common age group; this could be explained by the positive correlation between the time of emergence of impacted wisdom teeth and the beginning of problems that appear when it has partially erupted.

A rise in SAA levels was recorded as early as 1979 by Gilman et al., who triggered it by exercise (Gilman et al., 1979). Research in 1996 revealed a significant correlation between plasma norepinephrine and SAA following exercise, thus suggesting that SAA can be used as a marker of the activity of the sympathetic nervous system (Chatterton et al., 1996). Since then, many studies have been conducted and reviled the response of SAA to psychological stress and physical exercise, providing more evidence about the association between SAA and SNS (Filaire et al., 2010).

In the current study, there is a non-significant increase in SAA level following lower wisdom tooth surgical removal; which is the contrary to (Gutiérrez-Corrales et al., 2017), who showed a significant increase in SAA levels 2 hours postoperatively. This could be explained by the time of sampling, Gutierrez-Corrales took samples 1 hour, and 2 hours postoperatively, while in this study the samples were taken 2 days postoperatively.

Another study by AlMaummar et al. (2019), showed significant increase (difference) in SAA level between control and anxious patient groups undergoing

dental treatment (AlMaummar et al., 2019). The surgical removal of IMTM produce trauma that create a unique metabolic reaction leading to rise in circulating stress hormones and stimulating HPA which in turn increase SAA secretion (Al-Adili, 2016), and as emphasized earlier, the secretion of SAA is under the control of ANS, which is activated in response to inflammation and stress, leading to higher levels postoperatively. This could be the explanation for the (insignificant) increase in SAA levels postoperatively.

4 Conclusion

SAA levels increase but non significantly after surgical removal of impacted lower wisdom teeth which indicate that it cannot be used as an inflammatory marker; this increase has no relation to age and gender.

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