Salivary Alpha Amylase, Dental Anxiety, and Extraction Pain: A Pilot Study

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The primary intention of this study was to determine whether salivary alpha-amylase (sAA) factors or the Dental Anxiety Scale (DAS) was a better predictor of dental extraction pain. This study followed a cross-sectional design and included a convenience sample ($n = 23$) recruited from an outpatient oral surgery clinic. While waiting for their scheduled appointments, consenting patients completed both basic demographic/medical history questionnaires and Corah's DAS as well as submitted sublingual saliva samples. After their extractions, patients marked visual analog scales (VAS) to indicate the intensity of their intraoperative discomfort. Results of this study confirm that there is a relationship between a patient's dental anxiety and intraoperative extraction pain ($r[21] = .47, P = .02$). This study did not find that preoperative sAA factors (concentration and output rate) were related to either VAS extraction pain or DAS score. A strong positive relationship was observed between the concentration of sAA and the rate of sAA output ($r[21] = .81, P < .001$). Based on the results of our study, we conclude that dental anxiety has a moderate but significant correlation with intraoperative dental pain. Factors of sAA do not appear to be predictive of this experience. Therefore, simply assessing an anxious patient may be the best indication of that patient's extraction pain.

Key Words: Salivary enzymes; Dental anxiety; Dental extraction; Alpha amylase.

Salivary biomarkers have received special attention because they are readily accessible and easily obtained.^{1,2} Salivary alpha-amylase (sAA) is a metallo-enzyme produced by the parotid, submandibular, and sublingual glands for the purpose of hydrolyzing α -1,4 linkages of starch into glucose and maltose.³ Its diurnal rhythm exhibits a pronounced decrease within 60 minutes after awakening and a steady increase in activity throughout the day. Salivary alpha-amylase is released from preformed granules located within acinar cells that are innervated by both parasympathetic and sympathetic branches of the autonomic nervous system. Although it does not demonstrate a 1:1 relationship with plasma catecholamine levels, sAA secretion is regulated by alpha- and beta-adrenergic receptors and is still directly correlated with catecholamine release.³

Recently, sAA has emerged as a reliable biomarker for objectively assessing patient pain.^{4,5} Separate studies have also found a relationship between Corah's Dental Anxiety Scale (DAS) and patient pain.^{6,7} Pain percep-

tion is not simply a physiological experience. Emotional states can modulate pain, and there exists a reciprocal interplay among dental fears, dental pain, and the body's physiological response.⁷ The primary intention of this study was to determine whether sAA variables and Corah's DAS are reliable predictors of dental pain during extraction. To date, no study has assessed and compared the correlations of both pretreatment sAA variables and dental anxiety with intraoperative pain in the context of a dental treatment. Oral surgery is associated with a high expectancy of pain, and portable salivary biomarkers have the potential to help clinicians objectively ascertain anxiety levels and anticipate pain-control issues. This study hypothesized that both preoperative sAA salivary concentrations and DAS scores are significantly correlated with extraction pain.

MATERIALS AND METHODS

This study followed a cross-sectional design and included a convenience sample recruited from the waiting room of the outpatient oral surgery clinic at Columbia University, College of Dental Medicine. Data

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Figure 1. Patient partitioning. Data were excluded from analyses if patients had eaten 1 hour prior to saliva collection, were scheduled for intravenous sedation, had been diagnosed with xerostomia, were taking beta-blockers, or were taking antidepressant and/or antipsychotic medications.

collection was performed over a 3-week period between the hours of 1 and 4 PM to account for the diurnal rhythm of sAA secretion. All patients were older than 18 years and had a willingness to participate in the study following signed informed consent.

The same trained research fellow performed all data collection. While waiting for their scheduled appointments, consenting patients were asked to complete both a basic demographic/medical history questionnaire and Corah's DAS as well as submit a saliva sample. The DAS scale includes 4 specific questions about dental situations. Each question has 5 possible responses, ranging from 1 (*no anxiety*) to 5 (*high anxiety*), and total scores range from 4 to 20. An individual is considered normal when the total score is between 4 and 8, moderately anxious when the total score is between 9 and 12, highly anxious when the total score is between 13 and 14, and severely anxious when the total score is between 15 and 20.

As part of the standard of care at the Columbia University oral surgery clinic, all patients were verified for adequate anesthesia prior to extraction with the aid of a No. 9 molt periosteal elevator. No additional tests were performed to test the effectiveness of local anesthesia, and no data were collected regarding the difficulty of each extraction or length of time for each procedure. There were no complications reported with any of the extractions performed for this study.

After their extractions, patients were asked to mark 100-mm visual analog scales (VAS) to indicate the

Table 1. Summary of Demographics and Anxiety Statuses of Both the Total Sample and Final Included Sample for Analyses*

	Total (n = 41)	Included (n = 23)
Age, y	38.69 ± 15.23	41.26 ± 13.56
Male	18 (43.9%)	12 (52.2%)
Hispanic	19 (46.3%)	9 (39.2%)
DAS anxiety score		
Normal (4–8)	15 (36.6%)	9 (37.5%)
Moderate (9–12)	18 (43.9%)	9 (37.5%)
High (13–14)	4 (9.8%)	2 (8.3%)
Severe (15–20)	4 (9.8%)	4 (16.7%)
sAA output rate, U/min		36.46 ± 49.93
sAA concentration, U/mL		132.55 ± 169.03

* sAA factor averages are provided for the final included sample only. DAS indicates Dental Anxiety scale, sAA, salivary alpha-amylase.

intensity of their intraoperative pain. The VAS is a continuous, unidimensional measure of pain intensity that is scored by measuring the distance in millimeters from the left boundary of the line to a point marked by the patient. Each saliva sample was collected using a timed sublingual swab held in position for a minimum of 60 seconds to ensure adequate volume. Swabs were then transferred to labeled tubes and stored on ice until they could be moved to a -20°C freezer space. Once data collection was completed, the samples were packaged with dry ice and submitted to Salimetrics Lab Testing Services for sAA concentration analysis. Patients were excluded from the study if they had eaten or drank 1 hour before their appointment, were scheduled for intravenous sedation, had been diagnosed with xerostomia, were currently taking beta-blocker medications, or were currently taking antidepressants or antipsychotics. All statistical analyses were performed using SAS software version 9.3. Columbia University Institutional Review Board approval for this study was received on July 1, 2014 (protocol IRB-AAAN4505).

RESULTS

A total of 41 patients were enrolled in this study. At the time of saliva analysis, 2 samples were excluded on account of insufficient saliva volume. Fifteen samples were disqualified after saliva analysis on account of exclusion criteria. One sample was excluded during data analysis on account of improper VAS scoring. The final sample included 23 subjects (Figure 1).

Demographic summaries can be found in Table 1. Excluded patients were not significantly different from

Figure 2. Stratification of responses for each component of the Corah Dental Anxiety Scale.

included patients with respect to age, gender, or DAS scores. In both total and included samples, most patients presented with normal (total = 36.6%, included = 37.5%) or moderate (total = 43.9%, included = 37.5%) dental anxiety. Breakdowns of individual question responses are shown in Figure 2.

Of the included patients ($n=23$), the mean DAS score was 10.00 and the mean VAS score was 26.48. Pearson correlation coefficient was performed to test the correlation between VAS and DAS scores. A statistically significant correlation ($r = .47$, $P = .02$) was observed between these variables, and their relationship (Table 2) was classified as moderately strong (Figure 3). The 23 included patients were further categorized by 2 variables, DAS (normal/moderate anxiety or high/severe anxiety) and VAS (<50 or ≥ 50), into a 2×2 contingency table (Table 3). Pearson chi-square test for

Table 2. Linear Correlation Coefficients for Various Paired Variables*

	Pearson Coefficient, r	P Value
VAS; DAS	.47	.02
VAS; sAA conc	.22	.32
VAS; sAA rate	.14	.52
VAS; saliva rate	.18	.42
DAS; sAA conc	<.01	.98
DAS; sAA rate	.01	.64
DAS; saliva rate	<.01	.98
sAA rate; sAA conc	.81	<.001

* No correlations were corrected for outliers. A P value $<.05$ indicates that a statistically significant relationship is present between that pair of variables. The Pearson coefficient, r , indicates the strength of the linear relationship. $.3 < |r| < .5$ demonstrates a moderate relationship, $.5 < |r| < .8$ a strong relationship, and $.8 < |r| < 1.0$ a very strong relationship. conc indicates concentration; DAS, Dental Anxiety Scale; sAA, salivary alpha-amylase; VAS, visual analog scale.

Figure 3. (A) Scatter plot of salivary alpha-amylase (sAA) output rate (y axis) against sAA concentration (x axis) summarizing the results. $r(21) = .81$, $P < .001$. (B) Scatter plot of visual analog scale score (y axis) against Dental Anxiety Scale score (x axis) summarizing the results. $r(21) = .47$, $P = .02$.

independence was statistically significant, $\chi^2(1, n = 23) = 3.81$, $P = .05$, demonstrating that DAS and VAS are dependent variables.

Although DAS and VAS were correlated with each other, neither DAS nor VAS was found to be significantly correlated with any of the salivary variables assessed (sAA concentration, sAA output rate, saliva flow rate; Table 2).

Of the included patients, the mean sAA output rate was 36.46 U/min and the mean sAA concentration was 132.55 U/mL. Pearson correlation coefficient was performed to test the correlation between sAA output

Table 3. Dental Anxiety Scale (DAS) Category Partitioned Into Those With Normal or Moderate Anxiety and Those With High or Severe Anxiety*

		DAS Category	
		Normal/Moderate	High/Severe
Visual analog scale	<50	15	2
	≥50	3	3

* $\chi^2(1, n = 23) = 3.81$, $P = .05$.

rates and sAA concentrations. A statistically significant correlation ($r = .81$, $P < .001$) was observed between these variables, and their (Table 2) relationship was classified as strong (Figure 3).

DISCUSSION

In oral surgery, third-molar extraction is the most fearful experience for patients treated in the outpatient setting.⁸ Although most routine procedures in oral surgery are followed by relatively short recovery periods, they still have the potential to be stressful events. Pretreatment dental anxiety is correlated with difficulty of extraction,⁹ and patients with high trait or dental anxiety may require longer surgery times and have poorer postoperative recovery.¹⁰ Intravenous sedation largely serves to address these issues, but in patients for whom intravenous sedation is contraindicated, the surgeon would benefit from being able to preoperatively triage anxiety levels. In those cases, anxiety variables that are correlated to measures of intraoperative pain, operation time, and difficulty of extraction would be the most clinically meaningful.

DAS and VAS

The Corah DAS has historically been the most commonly used scale to measure dental anxiety.¹¹ It was adopted for use in this study because it has been shown to have high validity and is easy to administer clinically. Results of this study confirm that there is a relationship between a patient's dental anxiety and intraoperative extraction pain. Other authors who reported similar findings likewise demonstrated significant correlations with modest Pearson coefficients.^{6,7}

sAA Variables and DAS; sAA Variables and VAS

The scientific literature heavily supports the use of sAA concentration as an objective assessment of

physical stress.³ In our study, sAA was used as a biomarker for sympathetic activity on account of the powerful role the autonomic nervous system plays in sAA secretion and the acceptance within the published literature of sAA as a sympathetic biomarker.^{4,5,12–15} The net result of sympathetic stimulation on the salivary glands is a thicker, protein-rich saliva, in contrast to the watery saliva produced by parasympathetic activity. Multiple peer-reviewed studies have validated the application of sAA concentration as a noninvasive biomarker for both objective pain intensity and emotional status in a stressful environment.^{3–5}

This study did not find that sAA factors (both sAA concentration and sAA output rate) were related to either VAS or DAS score. These findings corroborate those of Sadi et al,¹⁴ who also found no significant correlation between DAS and sAA factors. The mean sAA concentration (132.55 U/mL) in our study was greater than that reported by Sadi et al¹⁴ (73.73 U/mL). The study by Sadi et al¹⁴ was conducted in the context of a dental hygiene appointment, while our study was conducted in the context of a dental extraction. The higher sAA concentrations found in our sample suggest that the sympathetic activity of our patients was elevated, presumably in anticipation of dental extraction. However, the elevations were not linearly correlated with DAS or VAS scores.

General anxiety is distinct from dental anxiety and can be categorized as either state or trait anxiety. State anxiety evaluates how one feels in the moment and is measured using subjective feelings of apprehension, tension, nervousness, worry, and activation/arousal of the autonomic nervous system.¹⁶ Trait anxiety evaluates one's overall susceptibility to anxiety.¹⁶ Thus, classifications for trait anxiety are relatively stable, whereas classifications for state anxiety can vary in the presence of anxious stimuli. The DAS score reflects dental anxiety, not general anxiety. sAA variables measure sympathetic activity and therefore may better reflect general state anxiety.¹³ The State-Trait Anxiety Inventory (STAI) is a superior instrument for the purposes of assessing general state and trait anxiety. Although the Corah-DAS has been traditionally used as an indicator of anxiety in dentistry, recent studies show that the STAI-State actually appears to be a more sensitive measurement of anxiety in oral surgery.¹⁷ Nonetheless, because they measure different phenomena, both scales (STAI and DAS) are recommended for evaluating anxiety in dentistry.¹⁷

There is some debate as to whether or not the DAS and STAI-State actually measure a similar phenomenon in the context of a dental procedure. Lago-Méndez et al,¹⁸ Kvale et al,¹⁹ Hakeberg et al,²⁰ and Tarazona et al²¹ all found a significant correlation between STAI-S and

DAS scores. Two other studies^{22,23} found no such correlation. Our study did not find a significant correlation between sAA variables and DAS (Table 2) and therefore would support the evidence that DAS measures a distinct phenomenon from general state anxiety.

It was hypothesized that anticipation of dental extraction would raise baseline sAA variables linearly with extraction pain.¹³ This was not found to be the case. Campos et al²⁴ also reported no correlation between sAA concentration and pain intensity. Those patients had a significant and progressive increase of sAA concentration during orthodontic treatment, but this increase was likewise not linearly correlated with pain intensity. Robles et al²⁵ found that sAA levels were lower prior to extraction than compared to consult and follow-up visits. Robles et al²⁵ hypothesized that both the anticipation of extraction and the effort expended to arrive at the clinic (ie, rushing to be on time) could have led to an increase in autonomic activity. It was thought that this increase would then decrease over time as the patients arrived at the clinic and were able to acclimate to the setting.

sAA Rate and sAA Concentration

A strong positive relationship was observed between the concentration of sAA and the rate of sAA output. This finding has been well-documented elsewhere.^{14,26,27} Rohleder et al²⁷ demonstrated that stress-induced cohorts had both increased sAA concentrations and sAA output rates from baseline. Sadi et al¹⁴ demonstrated that saliva of unstressed subjects showed a positive correlation between sAA concentrations and sAA output rates. This concentration-rate correlation suggests that sAA secretion is decoupled from the secretion of other saliva components.

Limitations

Given the study time frame and resources, a convenience sample was used to achieve the necessary sample size. It is likely that patients with severe dental anxiety refused participation in this study, were more likely to receive intravenous sedation, or were less inclined to seek treatment at a teaching clinic. Confounding factors to saliva analysis such as eating or drinking 1 hour before collection and current medications were all self-reported. Although at the end of the procedure patients were instructed to report VAS scores reflecting intraoperative pain, some may have reported

scores reflecting posttreatment or perioperative pain or discomfort. In addition, VAS scores do not reflect or correct for multiple tooth extractions, complicated or impacted extractions, and interoperator variability in anesthesia technique.

CONCLUSIONS

Based on the results of our study, we conclude that dental anxiety has a moderate but significant correlation with intraoperative dental extraction pain. Baseline sAA concentration and output rate do not appear to be related to this experience. Other contributing factors such as salivary cortisol are yet to be assessed in the context of a dental treatment. Future studies should identify additional baseline predictors; namely, they should assess both STAI-State and STAI-Trait anxiety in addition to the DAS.

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