

## Salivary Composition, Gender and Psychosocial Stress

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*The purpose of the present study was to evaluate salivary composition as an indicator of psychological stress. A group of 26 new Israeli patients in a stress management clinic was studied in an attempt to assess both the effects of long standing trait-related stress as well as that of situational state related stress. Significantly higher state anxiety scores, blood pressure, pulse rate, and lower salivary flow rates were found in the patients before as compared to after the initial psychodiagnostic interview, illustrating the effect of a situational stress. Significantly lower salivary flow rates and elevated total protein concentrations were found in the patients after the interview as compared to healthy controls, illustrating the effect of sympathetic enhancement associated with more stable trait-related psychological stress. Salivary electrolytes (Na and K) and IgA did not differ significantly when the clinical sample was compared to the control group. Although male patients reported lower state anxiety than female patients did, their hemodynamic and salivary composition measures indicated a higher sympathetic arousal, possibly reflecting Israeli male patient difficulties in adapting their distress.*

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### INTRODUCTION

Interest in the physiological aspects of psychological stress is growing. Many studies on the effect of psychological stress on hemodynamic indices, on serum hormones, and on the immune system have been published.

Saliva is especially attractive for psychophysiological studies, as it is easily collected by non-invasive methods, enabling screening of populations outside of medical institutions. The rationale for using saliva as an indicator of stress is based on the fact that saliva is secreted by an active process regulated by the autonomic nervous system (ANS) (1). Various types of physical and psychological stressors have been found to cause a marked ANS response, with various effects on target organs attributable to activation of adrenergic receptors. Salivary secretion might also be used for monitoring altered ANS activity caused by stress.

An additional rationale for the involvement of salivary glands in stress detection might be associated with a possible direct action of stress hormones on specific receptors in the salivary glands. Among hormones altering salivary composition, the best known is aldosterone, however, sex hormones and cortisol also

have an effect on salivary glands (1). Another important reason for our interest in the potential use of saliva for stress detection is the feasibility of monitoring the levels of various hormones. The salivary concentrations of many steroid hormones have been found to be significantly correlated with their blood levels (2). Steroid hormones and autonomic intervention also influence the secretion of the electrolytes sodium and potassium in saliva (3).

Several studies on the effect of psychological stress on salivary secretion have been published. Bates and Adams (4) reported a significant decrease in salivary secretion in dental students during examinations. Morse et al. (5-7) have published several studies on the effect of environmental stress, such as dental treatment, on salivary secretion. Decreased level of immunoglobulin of the A class (IgA) in saliva during academic stress has also been reported (8,9). The IgA in saliva is a mixture of antibodies secreted from the plasma cells in the vicinity of the glands.

The purpose of the present study was to examine the relationship between the effects of a situational stressor as well as those associated with more stable trait-related psychological stress and several physiologic parameters in distressed persons who came for help to

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**Table 1.** Salivary composition, hemodynamic, and state anxiety variables of stress patients before and after their psychodiagnostic interview.

Measures	Before		After		Difference	
	Mean	S.D./√N	Mean	S.D./√N	Mean	S.D./√N
Rate of Flow ml/min	0.25	0.04	0.26	0.03	-0.02	0.04
IgA mg/100ml	12.23	2.02	13.04	1.94	1.48	1.69
Protein mg/100ml	241.82	39.81	228.54	18.38	25.00	36.48
Na mEq/L	5.71	0.57	5.94	0.63	0.03	0.41
K mEq/L	28.95	1.60	28.58	1.41	1.75	1.40
Rate x IgA mg/min	267	62	124	52	-31	48
IgA / Protein	0.06	0.01	0.06	0.01	0.01	0.01
SBP	122.72	2.99	114.88	2.62	8.04***	2.05
DBP	74.55	1.85	70.55	1.92	4.19	2.31
PR	83.68	2.99	74.23	2.36	9.20***	2.18
State Anxiety	46.17	2.43	40.44	2.31	5.83***	1.39

paired t test

\*\*\*  $p < 0.001$ 

Rate X IgA = the rate of IgA secretion and is calculated by multiplying the salivary flow rate and IgA concentration in each individual. This variable illustrates the total IgA secreted by the glands per minute.

a stress management clinic. The patients were examined both before and after their initial psychodiagnostic interview and then compared to healthy controls.

## METHODS

The sample studied consisted of a group of patients (14 males and 12 females) who came to a stress management clinic for treatment and were diagnosed as suffering from either psychosomatic symptoms of stress or a DSM III-R anxiety disorder. Their ages ranged from 16 to 35 years (26 + 3.5 years). Only patients not on any medication were included in this study. The control group consisted of healthy age and sex matched individuals not taking any medication.

Saliva collection was done between 9 a.m. and 12 noon, at least 1 hour after a meal. The patients' blood pressure and pulse rate were taken before and after the interview. On both these occasions the patients responded to the state version of the State-Trait Anxiety Inventory, a set of 20 anxiety related items concerning how the respondent feels at a particular moment (10). This instrument was used as an evaluation of the current manifest stress level. The adaptation of the questionnaire was to the Hebrew language following the guidelines set by Spielberger and Diaz-Guerrero (11). Data on parameters of validity and reliability of the instrument were widely reported. Subjects were asked to evaluate each indicator of the inventory on a 4 point scale. A single score of state anxiety was obtained by summing the items. Scores of the inventory may vary from a minimal score of 20 to a maximal score of 80. It has been hypothesized that individuals seeking help in a stress management clinic would feel more stressed

than healthy controls, and that their situational anxiety level would be higher before they entered their psychodiagnostic interview than following it.

Whole, resting saliva was collected by the spitting method both before and after the interview. The samples were kept at 4°C until analyzed. The volume of saliva was measured, and salivary secretion rate was calculated (12). The samples were centrifuged at 3,000 rpm for 10 minutes. Na and K concentrations were measured by flame photometry, total protein by the method of Lowry et al. (13), and IgA by the method of Mancini et al. (14) on low-level Kallestad plates using serum IgA as standard. IgA secretion rates were calculated by multiplying IgA concentration by flow rate.

Student's t-test was used for comparison of the patients' data, with that of the controls, while the paired t-test was used to assess changes in patients following the psychodiagnostic interview.

## RESULTS

Table 1 compares the patients' data before and after a situational stressor: the psychodiagnostic interview. Following the interview significant decreases in state anxiety scores (SA), pulse rates (PR), and systolic blood pressure measures (SPB) were found. No significant changes in saliva variables were noted. However, when we divided the patient group by gender we noticed an interesting difference between male and female patients relevant to their salivary flow rate. While males showed a significant increase of salivary flow after the interview ( $p = 0.04$ ) the females showed a statistically non-significant opposite trend.

**Table 2:** Salivary composition in stress patients following their psychodiagnostic interview as compared to healthy controls.

Measures		Patients		Controls		p (t-test)
		Mean	S.D./√N	Mean	S.D./√N	
Rate of Flow	ml/min	0.26	0.03	0.37	0.03	<0.01
IgA	mg/10ml	13.04	1.94	9.98	1.05	NS
Protein	mg/100mi	228.54	18.38	131.96	12.28	< 0.001
Na	mEq/L	5.94	0.63	7.67	1.62	NS
K	mEq/L	28.58	1.41	29.17	2.83	NS
Rate x IgA	mg/min	324	52	338	33	NS
IgA /Protein		0.06	0.01	0.08	0.01	< 0.05

The results of the salivary parameters measures in patients following their intake interview as compared to healthy controls are given in Table 2. Significantly lower salivary flow rates were detected in the clinical sample which also had significantly higher concentrations of total salivary protein. No significant differences were found between stress patients and controls as far as Na, K, and IgA concentrations or IgA secretion rates were concerned. The IgA-to-protein ratio, however, was lower in patients as compared to controls. This ratio represents the proportion of IgA antibodies within the total proteins secreted by the salivary glands.

Control group data concerning state anxiety and hemodynamic data were not available for comparison. The post interview variables of the patient group, however showed hemodynamic measures within normal limits. The lower post interview levels of the patients' state anxiety, on the other hand, were still high and ranked an average above the 85th percentile when measured against norms of healthy adults, re-

flecting the more stable character of experienced stress in this group.

Several gender differences within the stress patients were noted (see Table 3).

Male patients had higher protein, Na, K, Rate x IgA, and SBP measures, marginally higher IgA and PR rates and lower state anxiety scores. Although no significant Time x Gender interactions were found, a trend towards some gender differences in response to the stressor were noted. For example, male patients tended to increase their rate of salivary flow rates following the intake interview from 0.24 ml/min to 0.31 ml/min. Female patients, on the other hand, had a mean flow rate of 0.26 ml/min before the interview which decreased to 0.21 ml/min following it. The mean of those differences for male patients was 0.08 ml/min (S.D. / √N = 0.03), indicating again an increase. When analyzed by a paired t-test this difference showed a p level of 0.04. A non significant opposite trend was found among female patients.

**Table 3.** Mean salivary hemodynamic, and state anxiety variables of stress patients by time of measurement in relation to the psychodiagnostic interview and by gender.

						P	
		Before		After		(GLM procedure)	
		m	f	m	f	Time of Measurement	Gender
Rate of flow	ml/min	0.24	0.26	0.31	0.21	NS	NS
IgA	mg/100mi	13.82	10.64	16.08	9.73	NS	NS
Protein	mg/100ml	289.55	194.09	276.15	172.27	NS	<0.05
Na	mEq/L	6.45	4.97	7.09	4.44	NS	<0.01
K	mEq/L	31.60	26.29	31.27	25.09	NS	< 0.005
Rate x IgA	mg/min	320	216	438	199	NS	<0.05
IgA / Protein		0.05	0.07	0.06'	0.06	NS	NS
SBP		126.36	118.09	117.76	111.50	<0.05	NS
DBP		75.00	74.00	71.18	69.67	NS	NS
PR		87.43	78.91	76.43	71.67	<0.05	NS
State Anxiety		42.33	50.82	37.23	43.92	NS	<0.05

## DISCUSSION

Following the intake interview, significant decreases in state anxiety scores, pulse rates and blood pressure measurements were noted, demonstrating the effects of terminating a stressor event. The new patients who were facing the unknown as far as their treatment was concerned, might have been also experiencing some expectational anxiety with regard to their performance and experience within the interview itself. The decrease in situational stress following the interview was also related to an increase change in salivary flow rate among males. This data corresponds with findings reported by Morse et al. (5,6) concerning stress and relaxation in clinical endodontic patients. However, while Morse et al. reported elevated total protein concentration among subjects experiencing examination induced stress (7), we did not find such changes within a gender combined group of persons suffering from a more long standing stress (the patients' ongoing psychological stress) who were also exposed to a situational stress (the expectational anxiety in anticipation to the psychodiagnostic intake interview). The possibility of a gender dependent salivary flow response to the interview should be considered here.

We assumed that the differences between the more relaxed patients who were relieved from the situational stressor and healthy controls, would help us understand some of the impacts of a long standing more trait-related stressor on salivary composition. We found significant lower salivary flow rates in the post intake patient group compared to healthy controls. The difference could not be explained by a lingering arousal associated with the interview because all three hemodynamic measures (SBP, DBP and PR) indicated a functioning within normal limits. This difference could reflect a chronically enhanced sympathetic activity among people seeking help due to persistent distress. We think that such activity affects adrenergic receptors in the salivary glands. It is also feasible that a decrease of blood flow to the glands due to vasoconstriction caused by the constantly aroused sympathetic system would also contribute to a lowered salivary flow rate.

No significant difference in salivary IgA concentration or IgA secretion was found when the patients group was compared to controls. However, the IgA to protein ratio was found to be significantly lower in the patients group (see Table 2). A lowered IgA/protein ratio such as we found in the patient group

might represent either the elevated protein or the lowered IgA concentration. The data on salivary IgA in psychoimmunology research is, thus, still somewhat controversial (15).

Jemot and McClelland (16), on the other hand, argued that salivary IgA levels are stable over time and have been shown to be related to future illness. Our data on IgA saliva may be in agreement with Mouton, et al. (17) who found a weak negative correlation between the level of salivary IgA and stress rating. A recent series of studies looked at the effects of motive-related personality variables on NK cell numbers and activity, another set of immunologic parameters (18) and suggested that more stable trait-related variables may be important mediators of the effects of stress on some aspects of immune function. Because some immunologic diseases have a prolonged development and course we are currently exploring the relationships between stable personality dimensions and altered salivary immune response to stress.

Significant differences in salivary composition were found when the male patients were compared to female patients (Table 3). No such differences were present when healthy populations were investigated (19). The significantly higher concentrations of total protein, sodium, potassium and rate of IgA secretion found in saliva of male stress patients might indicate an enhanced sympathetic activity expressed not only in salivary composition, but also in a trend towards more elevated systolic blood pressure and pulse rate measures (*p* values 0.06 and 0.08 respectively). Interestingly, the self reported state of anxiety scores were significantly lower among male patients. We can suggest, then, that although males afflicted with stress and anxiety disorders tend to report lower state anxiety levels when initially presenting themselves to treatment, their physiological stress responses tell a different story, and appear to be more pronounced than that of female patients. It is conceivable that such under-reporting of anxiety would also be manifested outside the clinic and result in less emotional support for males, thus, further increasing their stress.

Whether or not this finding represents a universal gender issue associated with potential consequences of male needs to deny vulnerability and weakness remains to be clarified through replication studies. To rule out specificity of these findings to the Israeli man-oriented war-torn culture, further studies conducted in different sociopolitical milieus are called for.

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## Index Terms

saliva, stress.

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