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# THE USE OF SNYDER'S CARIES ACTIVITY TEST IN ORAL HYGIENE EFFECTIVENESS MONITORING

by

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# SUMMARY PAGE

### THE PROBLEM

The evaluation of the effectiveness of dental caries preventive procedures ultimately depends upon a decrease in the caries incidence. Since at least a year is required to make a meaningful caries rate assessment, interim caries activity tests are needed. The Snyder's caries activity test seemed a promising tool for the assessment of oral hygiene effectiveness, but it required evaluation.

### FINDINGS

The Snyder's caries activity test results were extremely reproducible for each individual subject. No significant changes were obtained, however, even with the most careful toothbrushing practices.

### APPLICATIONS

The Snyder's test is recommended as a highly reliable test for the characterization of the caries activity of an individual; however, its usefulness as a tool for assessing hygiene effectiveness seems limited.

### ADMINISTRATIVE INFORMATION

This investigation was conducted as a part of Bureau of Medicine and Surgery Research Work Unit MR005.19-6042—Study of Preventive Dental Principles and Methods in Military Populations. This report has been designated as Submarine Medical Research Laboratory Report No. 559. It is report No. 4 on this Work Unit, and was approved for publication as of 13 January 1969.

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# ABSTRACT

Dental caries is a rather slowly progressing disease. At least a year is required to evaluate a preventive measure by means of a reduction in the caries incidence. The Snyder's caries activity test, which is based on the acid production by oral bacteria, seemed an attractive short-term assessment tool for hygiene effectiveness.

Six subjects used three different hygiene methods and their "caries activity" was measured before and after the hygiene procedures. The results were essentially negative with regard to any hygiene method significantly reducing the caries activity. In all cases, however, an extreme degree of reproducibility was noted in the individual's caries activity. It is concluded that the test is a useful tool for characterizing an individual's caries activity, but the usually employed hygiene methods are not reflected in a changed activity.

# THE USE OF SNYDER'S CARIES ACTIVITY TEST IN ORAL HYGIENE EFFECTIVENESS MONITORING

### INTRODUCTION

Dental caries is a chronic, slowly progressing disease. This fact, coupled with the apparently multiple causes, has made the study and the control of this disease very difficult. This difficulty is obvious when it is considered that it takes approximately one year for the average young Navy man to develop two discernible new carious lesions. In this period of time it is recognized that many variables could have played a part in the disease and could only be discovered by a most painstakingly precise longitudinal analysis.

For this reason, various caries activity tests have been devised with which caries activity may be assessed on a more immediate and short term basis rather than on the basis of having to wait for an enumeration of actual lesions. The attractiveness of such tests is readily apparent in that they could allow for the immediate assessment of preventive measures.

Among the tests devised, the Snyder's caries activity test is probably one of the best known. In epidemiological studies it has been shown to be well correlated with other bacteriological counts and with actual carious lesion enumerations.<sup>1,2</sup> The test employs selective bacteriological medium for acidogenic microorganisms. A sample of saliva is inoculated into tubes of media and color changes of bromcresol green in the media are read after 24, 48 and 72 hours. The results are read qualitatively as caries activity, either none, slight, moderate or marked.

The difficulty of relating oral bacterial tests to individual caries activity assessments has been previously recognized<sup>3</sup>; however, they have been used to demonstrate the effect of caries elimination on the oral flora<sup>4,5</sup> and recently<sup>6</sup> demonstrated the effectiveness of an electric toothbrush in reducing caries activity as measured by the Snyder's test.

The idea of using this caries activity test as a tool for assessing the effectiveness of hygiene methods was very attractive. Some preliminary studies using the standard Snyder's test at this laboratory, however, were inconclusive. It was therefore deemed appropriate to attempt some modification of the standard Snyder's test in order to see if it could be used as a monitoring tool for oral hygiene effectiveness.

### MATERIALS AND METHODS

Some preliminary studies at this laboratory had given the impression that changes in the Snyder's caries activity test were too gross to be of any value in showing the effects of hygiene measures. For this reason a pilot study was first undertaken to determine the best quantity of whole saliva to use for the actual effectiveness study.

Two subjects known to show positive caries activity with the Snyder's test were used. Salivary samples were collected from each subject in the following manner: The mouth was first rinsed with water; the subject chewed paraffin for 25 chewing strokes (all chewing being done on the same side), and then he was requested to spit, repeat the process and spit again. The first two samples of saliva having thus been discarded, the third sample was then collected in a sterile vial. The paraffin was then removed, the mouth rinsed again, and the entire collection procedure was repeated until four samples had been collected.

Prepared Snyder's tubes containing 10 ml of Snyder test agar were inoculated. The saliva was inoculated in the following quantities: 0.2 ml, 0.1 ml, 0.07 ml, 0.04 ml, and 0.02 ml. In the case of subject B, the 0.2 sample was not used in view of his known strongly positive reaction. The medium was melted by placing the tubes in boiling water until melted throughout. The tubes were allowed to cool to  $45^{\circ}$ C and the saliva sample was added and mixed with the medium. The tubes were allowed to solidify at room temperature, and were then incubated at  $37^{\circ}$ C for 72 hours.

The saliva samples were collected before the hygienic procedure, after brushing, after using the Water Pik and after brushing and using the Water Pik. This entire program was repeated on three days for each of the two subjects. The results are summarized in Tables I and II. The reduction in Snyder's reaction with the hygienic measures used was significant in both subjects and was particularly dramatic in subject A (Table I).

Tab	le	I
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Initial Overall Snyder's Test Activity With Hygiene Procedures

Caries		Subject A		Teathburshise
Activity	Initial	Toothbrushing	Water Pik	Toothbrushing and Water Pik
ċ	o	0	2	21
Slight	0	4	22	18
Moderate	60	56	36	21
Marked	0	0	0	o
Marked	0	0	0	0

 $x^2 = 108.20$ 

The data from this pilot study were used in planning a more detailed study using six subjects and a slightly different, but more normal hygiene program.

Table II   Initial Overall Snyder's Test With Hygiene Procedures					
		Subject B			
Carles Activity	Initiai	Toothbrushing	Water Pik	Toothbrushing and Water Plk	
0					
S∣igh†					
Moderate	35	44	43	48	
Marked	13	Ļ	5	0	

x<sup>2</sup> ≈ 18.272

From each of the subjects an initial sample was collected as in the pilot study; he then performed a hygiene procedure and another saliva sample was collected. The sampling was done at the same time on three consecutive days for three weeks as depicted in Table III.

# Table III

Sampling Program With Hygiene Procedures

	Week !	Week 2	Week 3
First day	Brush	Brush and Water Pik	Water Pik
Second day	Brush <b>a</b> nd Water Pik	Water Pik	Brush
Third day	Water Pik	Brush	Brush and Water Pik

The quantities of saliva inoculated in the Snyder's media differed slightly from those of the pilot study: 0.2 ml, 0.07 ml, and 0.02 ml. In all other respects the techniques were identical to those of the pilot study.

### RESULTS

The Snyder's caries activity for all subjects and changes noted with the hygiene methods used are given in Tables IV, V, and VI. A cursory look at this data reveals but little change in the caries activity test with the hygiene methods. Only in those subjects with very high activity (Subjects D and F) was there any significant decrease in the activity and even this slight reduction was not seen when the Water Pik was used. The combined data for all subjects are given in Table VII. Chi square analysis reveals no significant differences in any of the distributions as related to oral hygiene methods.

Table	IV
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Snyder's Test Changes With Toothbrushing

	Carles Activity				
Su	b ject	Negative	Slight	Moderate	Marked
Α	before	25	2	O	0
	after	21	5	1	0
В	before	8	5	13	1
	after	4	8	15	0
С	before after	6 10	15 14	6 3	0
D	before	0	0	3	24
	after	0	0	10	17
E	before	19	5	3	0
	after	22	5	0	0
F	before	0	0	l	26
	after	0	0	L	23

### Table V

Snyder's Test Changes With the Use of the Water Pik

Su	b ject	Negative	Caries A Slight	ctivity Moderate	Marked
A	before after	23 21	3 4	1	0
В	before after	8 8	5 9	12 7	2 3
С	before after	12 12	3 9	8 6	4
D	before after	0 0	0	0 0	27 27
Ε	before after	17 21	6 3	4 3	0 0
F	before after	0 0	0 0	0 0	27 27

#### Table VI

Snyder's	Test	Changes	With	Toothbrushing	and
	the	Use of	the Wa	ater Pik	

			Carles Activity					
Şu	bject	Negative	Slight	Moderate	Marked			
A	before after	15 14	8 	14 2	0 0			
В	before after	12 5	5	9	 0			
с	before after	8 I0	6 9	13 8	0 0			
D	before after	0 0	0 0	6	26 21			
E	before after	19 20	5 6	3	0 0			
F	before after	0 0	0 0	0 4	27 23			

The overall data are expressed slightly differently in Table VIII. A numerical value was given to each caries activity level so that parametric analyses could be performed. The values assigned were 0 for negative activity, 1 for slight activity, 2 for moderate activity, and 3 for marked activity. As seen in Table VII no significant differences resulted from the hygiene procedures with this type of analysis. The parametric arrangement of the

#### Table VII

Combined Analysis of Carles Activity After Hygiene Methods							
	(BRUSHING)						
	0 Activity	Slight Activity	Moderate Activity	Marked Activity			
Before brushing	58	27	31	46			
After brushing	57	32	33	40			
(WATER PIK)							
Before Water Pik	60	17	25	60			
After Water Pik	62	62 25		57			
	(BRUSHING - WATER PIK)						
Before brushing and Water Pik	54	23	31	54			
After brushing and Water Pik	49	37	33	43			

data does, however, enable a factorial analysis. This analysis of variance table (Table IX) indicates that between subject variation was the significant factor. The type of hygiene practice was inconsequential even with subject interaction. The practice of a hygiene procedure did account for a significant portion of the variance observed (P < .05).

### Table VIII

Average Carles Activity With Various Hygiene Methods

	Before Test	After Test	Difference
Brushing	1.40 ± 0.10	1.35 ± 0.09	0.05
Water Pik	1.52 ± 0.10	1.43 ± 0.10	0.09
Brush & Pik	1.52 ± 0.10	1.43 ± 0.10	0.09

The basic aim of this study was to evaluate the usefulness of the Snyder's test by using varying quantities of saliva in the inoculation. Table X gives the distribution of caries activity states when only what appeared to be the optimum salivary dilution was used for each subject. In other wordsthe lowest salivary quantity was used for those subjects with high caries activity. No significant differences were noted between any of the distribution patterns in Table X.

### Table IX

Analysis of Variance: Factors Related to Snyder's Test Reaction

,		'		
Source of Variation	Sum of Squares	d f	Mean Squares	F
Subjects	1135.72	5	227.14	582
Type hyglene method	1.73	2	0.87	
Hygiene procedure	.8	I	1.81	4.64
Interactions:				
Subject × type method	8.16	10	0.82	
Subjects x procedure	4.84	5	0.97	
Type method x procedure	0.01	2	00	
Second order interaction	2.15	10	0.22	
Between group	1154.42	35	32.98	
Within group (error)	365.46	936	0.39	
Total	1519.88	971		

### DISCUSSION AND CONCLUSIONS

In view of the positive results reported by Toto<sup>6</sup> and the promising results of our pilot study, it was somewhat discouraging to note the lack of effect of hygiene practices on the Snyder's caries activity test.

### Table X

Combined Analysis of Carles Activity With Selected Dilutions

	(BRUSHING)				
	0 Activity	Slight Activity	Moderate Activity	Marked Activity	
Before brushing	13	13	15	13	
After brushing	7	19	19	9	
		(WATER PIK)			
Before Water Pik	8	124	11	21	
After Water Pik	10	12	ıЦ	18	
	(BRUS	SHING - WATER	PIK)		
Before brushing and Water Pik	8	11	17	18	
After brushing and Water Pik	8	16	15	15	

Parenthetically, the borderline significances found with hygiene procedures when using parametric analyses should be discounted as of no practical importance. These type of data render parametric analyses invalid except for pointing to factors of importance. The number one factor in explaining the variances was, of course, the between subject differences. It must of course be remembered that there are two possible explanations for these negative findings: A lack of sufficient precision of the test or an insufficient cleansing effect of the hygiene procedures.

It should be noted that a high degree of uniformity of test results was present within the subjects indicating that the Snyder's test should be useful for longitudinal studies. In other words, this study should not lead one to the conclusion that the test is worthless for any purpose. In fact, the next logical step would seem to be the testing of more intensive oral hygiene programs to see if the Snyder's caries activity could be reduced.

When one considers the basis of the sampling technique for the Snyder's test, it would seem that any procedure which might increase the level of dislodged plaque organisms should increase the Snyder's test reaction. In other words, it could be conjectured that toothbrushing alone might increase the reaction and a flushing device like the Water Pik might decrease it without actually reducing the plaque to any appreciable extent. The results of this study certainly do not bear this out and actually indicate the opposite to be true. The slight decreases noted in the test reaction were associated with toothbrushing and not with the use of the Water Pik.

It is concluded that the Snyder's caries activity test provides a reproducible test which should be useful in longitudinal studies; however, the use of this test in routine hygiene procedures at best seem equivocal.

#### REFERENCES

- Snyder, M. A simple method for the estimation of relative numbers of lactobacilli in the saliva. J. D. Res., 19:349, 1940.
- Snyder, J. L., Porter, D. R., Claycomb, C. K. and Sims, W. Evaluation of laboratory tests for the estimation of carics activity. J.A.D.A. 65:30-45, 1962.
- Green, G. E., Weisenstein, P. R. and Permar, D. Studies on salivary lactobacilli and dental caries in children during a dentifrice test program. J. D. Res. 36:828-838, 1957.
- Shklair, I. L., Englander, H. R., Stein, L. M. and Kesel, R. G. Preliminary report on the effect of complete mouth rehabilitation on oral lactobacilli counts. J.A.D.A. 53:155-158, 1956.
- Elliot, R. P. Full mouth rehabilitation retards oral lactobacilli. J. Tenn. S. Dent. Assn. 44:13-20, 1964.
- Toto, P. D., Evans, C. L. and Sawinski, V. J. Reduction of acidogenic microorganisms by toothbrushing. J. D. Child. 34:38-40, Jan. 1967.

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