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The lactic acid content of the saliva after carbohydrate ingestion.

II. source of the salivary lactic acid and inhibition of its formation.[1]

By

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In a preliminary report of this investigation (ERICSSON & HELLSTRÖM, 1950; in Swedish, with summary and diagram legends in English) an account was given of some earlier publications on the intra-oral production of acids, and of lactic acid in particular. Since this acid is present in greater quantities than the others and is comparatively easy to analyse, the investigations were concentrated on the lactic acid content of the saliva after carbohydrate ingestion and on factors that might influence this. It was demonstrated in a limited number of tests that the formation of lactic acid began immediately after carbohydrate ingestion, that the maximum concentration in the saliva was usually attained about 5 minutes after the ingestion and receded to the low fasting levels after about half an hour. The lactic acid curves of three subjects showed individually characteristic trends but in the separate subject they were about the same for each of the four test foods — cane sugar, white bread, boiled potato, and raw apple.

It was assumed that the salivary lactic acid either was the result of diffusion from the greater aggregates of oral bacteria,

[1] This investigation forms part of co-ordinated work on dental caries sponsored by the Swedish Medical Research Council through its Sub-committee for Caries Research.

possibly especially from the dental plaque, or had been formed in the saliva by bacteria derived from these aggregates. This view was in conformity with the demonstration by SREEBNY and collaborators (1950) that the oral glycolytic enzymes are intra-cellular. On the other hand, the hypothesis was directly contrary to MÖLLMANN's (1935) assertion that the salivary lactic acid originated exclusively in the glands and was secreted with the saliva. In some cases MÖLLMANN found lactic acid levels about as high as our concentration maxima, but since the samples were not taken at the same short, fixed intervals before and after carbohydrate ingestion they did not give as clear a picture of the connexion between the ingestion and the acid formation.

This report provides an account of (1) some tests indicating the source of the salivary lactic acid and (2) investigations of the effect of some antienzymatics and antibiotics on the intra-oral formation of lactic acid.

I. Procedure.

The following standard method of saliva collection was applied. Samples containing at least 0.2 ml of the mixed rest saliva were taken before the carbohydrate ingestion and 2, 5, 10, 20, and 30 minutes after its completion. The first sample, for determination of the fasting value, was taken at least 45 minutes after the last meal. The samples taken after the ingestion consisted of the saliva collected from the relaxed subjects $1^1/_2$—$2^1/_2$, $4^1/_2$—$5^1/_2$ minutes, and so on, after its termination. Of the samples, 0.2 ml aliquots were immediately pipetted into the copper sulphate solution used to stop further fermentation.

Unless otherwise stated a standard ingestion procedure was followed in which a lump of ordinary cane sugar (1.5 gm) was sucked for two minutes.

Lactic acid was analysed by the colorimetric method of BARKER & SUMMERSON (1941), with some small modifications relating mainly to the proportions of the reagents. Each series of analyses included a sufficient number of standard lithium lactate solutions to provide a calibration curve from 0 to 3 mC. (The designation mC — millimoles per liter — is used throughout this paper.) The error in analysis was calculated to be less than 5 per cent.

II. Sources of the lactic acid.

In theory there would seem to be three possible sources of salivary lactic acid

(1) secretion with the saliva in the glands;

(2) formation in the saliva by the salivary and bacterial enzymes; and

(3) formation within the greater oral aggregates of bacteria in the dental plaques and the retention sites of the mucous membranes. The following experiments were performed in an attempt to resolve this problem.

A. The pure parotid and mandibular secretions of a number of subjects were analysed separately both before and shortly after standard ingestion of sugar. The subjects were chosen from those enumerated in Section III, implying that their mixed resting saliva was repeatedly shown to contain appreciable amounts of lactic acid after standard ingestions. The secretions of the mandibular glands were collected at their orifices with a glass ejector, while the parotid saliva was shut off by means of parotid clips or cotton rolls. Parotid saliva was collected by a similar suction method at the parotid papilla on either side, while contact between cheek and adjacent teeth and gums was prevented and the mandibular saliva removed with an ordinary ejector. Immediately before the samplings following the sugar ingestion a small area round the glandular duct orifice was rapidly washed with water and dried.

In several of these tests, collecting times considerably exceeding those of the standard technique were required in order to obtain sufficient volumes. All these long collecting periods fall, however, within the time interval where standard samplings in the same subjects yielded greatly increased quantities of lactate. Tables 1 and 2 give the test results for mandibular and parotid saliva, respectively.

Since the lactate content of these pure secretions has never exceeded the order of magnitude of the fasting values, it can be concluded that the lactate is not derived from the greater salivary glands. The minute secretions of the numerous small glands of the oral mucosa have not been analysed on account of technical difficulties, but it seems most improbable that they alone should yield the lactate. The increased salivary lac-

Table 1.

The lactic acid content of mandibular saliva, sampled at the gland orifices, compared with the average lactic acid content of the pooled oral secretions.

Subject	Saliva	Initial content	Lactic acid content after sucrose ingestion		
G. B.	Mandibular saliva	0.49	2—7 min. 0.77	14—16 min. 0.25	
	Comp.: mixed saliva	0.54	5 min. 5.96	10 min. 3.93	20 min. 2.47
B. J.	Mandibular saliva	0	2—4 min. 0	10—12 min. 0	
	Comp.: mixed saliva	0.15	2 min. 4.50	10 min. 4.19	
I. H.	Mandibular saliva	0	2—4 min. 0	10—12 min. 0	
	Comp.: mixed saliva	0.09	2 min. 1.90	10 min. 2.84	

Table 2.

The lactic acid content of parotid saliva, sampled at the gland orifice, compared with the average lactic acid content of the pooled oral secretions.

Subject	Saliva	Initial content	Lactic acid content after sucrose ingestion		
B. J.	Parotid saliva	0.30	1—11 min. 0.37		
	Comp.: mixed saliva	0.15	2 min. 4.50	5 min. 4.57	10 min. 4.19
E. S.	Parotid saliva	0.18	1—19 min. 0.28		
	Comp.: mixed saliva	0.25	2 min. 3.51	5 min. 3.78	10 min. 4.75
E. K.	Parotid saliva	0.38	1—9 min. 0.34		
	Comp.: mixed saliva	0.21	2 min. 2.10		5 min. 3.32

tate content after carbohydrate ingestion should accordingly be due to formation in the oral cavity.

B. Formation of acid within the saliva, sufficiently rapid to account for the observed lactate increase, seemed *a priori* unlikely, judging from previous data. Assuming about half of the acid formed to be lactic (SUMMERSON & NEUWIRTH, 1941; MUNTZ, 1943), the total amount of acid present in the 5 minute samples from the subjects reported in Section III may be calculated to be 6—12 milli-equivalents per liter. Knowing the average buffer capacity of the saliva (ERICSSON, 1949) one can predict that such amounts of acid will markedly decrease its pH value. If, however, the saliva had been incubated with fermentable carbohydrate *in vitro*, so that the salivary enzymes and bacteria could act under conditions similar to those in the oral cavity, no pH decrease was evident until a much greater period had elapsed (VOLKER & PINKERTON, 1947; WESTIN, 1948; HILL & WHITE, 1949).

A few consistent tests were therefore taken as sufficient indication that the lactic acid was not formed within the volume of saliva constituting the sample.

Samples of unstimulated mixed saliva from three of the subjects in Section III were first analysed for their individual initial lactate contents. Of each of these samples 2 ml were then incubated with 20 mg glucose at 37°, shaking continuously. Samples for analysis were taken after 5, 10 and, in one case, 30 minutes. The results appear in Table 3.

Table 3.

Lactic acid formation in saliva glucose mixtures in vitro.

Subject	Salivary lactic acid, mC			
	Initial	5 min.	10 min.	30 min.
B. J.	0.0	0.08	0.08	
E. S.	0.19	0.19	0.16	
I. H.	0.06	0.06	0.21	0.51

In similar experiments MÖLLMANN (*loc. cit.*) found no increase in the lactate content even after 1—3 days incubation. From this he drew the inference — which was clearly too wide — that the salivary lactic acid is not on the whole formed by carbohydrate decomposition. With such long incubation periods

and aerobic conditions particular regard should also be had to the further decomposition, to an unknown but probably considerable degree, of the lactic acid formed (NEUWIRTH & SUMMERSON, 1942; MUNTZ, 1943).

There seems to be greater justification, however, for the more restricted inference that the greater part of the lactic acid is not formed within the bulk of the saliva, probably on account of its too low concentration of bacteria.

C. Since the evidence pointed clearly to the fact that the acid formation took place essentially in the bacterial aggregates on the tooth surfaces and the mucous membranes, some tests were made in order to determine the part played by the dental plaque. The salivary lactate content was determined by the standard technique:

(1) in cases with pronounced plaques, before and after the removal of these plaques;

(2) in cases where full extraction was intended, before and after removal of the teeth;

(3) in full denture cases, with and without the dentures *in situ*.

(1) These tests were performed with two patients having very heavy coatings on the teeth. After the first series of standard samples the plaques were carefully removed, the mouth rinsed with water, and a fresh standard series run. The results appear in Table 4.

Table 4.

Plaque cases.

Subject		Salivary lactic acid, mC					
		Fasting	2 min.	5 min.	10 min.	20 min.	30 min.
P. K.	Before removal	0	2.85	6.15	4.32	2.20	0.49
	After removal	0.17	3.45	5.07	4.17	0.60	—
S. P.	Before removal	0.73	1.23	1.68	1.51	0.38	0.46
	After removal	0.14	1.38	0.93	1.13	0.93	0.10

Table 5.

Total extraction cases.

Subject		Salivary lactic acid, mC					
		Fasting	2 min.	5 min.	10 min.	20 min.	30 min.
H. M.	Before extraction[1]	0.44	4.53	4.87	2.45	0.48	0.21
	After extraction	0	6.63	6.72	5.13	0.75	0.48
P.-E. P.	Before extraction[2]	0.43	4.26	3.15	1.53	1.25	0.65
	After extraction	0.44	2.28	4.59	2.86	3.23	1.13

[1] (2) teeth)
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Table 6.

Full denture cases.

Subject		Salivary lactic acid, mC					
		Fasting	2 min.	5 min.	10 min.	20 min.	30 min.
K. S.	With denture	0	3.75	5.04	3.00	2.65	2.04
	Without denture	0	1.83	2.34	1.91	1.36	0.44
S. S.	With denture	0.22	3.87	3.69	2.60	0.38	0.21
	Without denture	0	1.56	1.44	1.18	0.06	0
R. E.	With denture	0.10	1.86	3.30	2.59	0.13	0.03
	Without denture	0.22	0.90	1.41	1.30	0.16	0.10

(2) These tests were made with two patients before extraction and after complete epithelial healing of the wounds; the conditions were otherwise identical. The results are given in Table 5.

(3) The results of the tests in the full denture cases appear in Table 6.

These experiments show that the lactic acid formation does not occur exclusively in the dental plaque. Even in edentulous mouths considerable quantities of acid are formed. The microscopic structure of the different parts of the oral mucosa justifies the supposition that the bacteria of the dorsum of the tongue may supply an appreciable part of the lactic acid diffusing into the saliva. A report by HILL & WHITE (*loc. cit.*) is in accord with this: the pH drop on the tongue surface after sugar ingestion followed the same course as the well-known Stephan curve of the dental plaque (STEPHAN 1938, 1940). The only consistent difference between the different sampling conditions applying to any one subject appeared in denture cases with and without the dentures *in situ*. This, however, tells us nothing about the ordinary dental plaque.

III. The effect of some inhibitors on the intraoral formation of lactic acid.

The subjects chosen for these investigations were ten healthy persons with good dental status, and few carious cavities. No dental treatment was received during the experimental period of about one year. A further condition was that the subject's normal uninfluenced oral development of lactic acid after carbohydrate supply should exceed a certain minimum level. This was necessary because the principle of these tests required a comparison of each subject's normal lactate curve (average of at least three experimental series) with the curve obtained after applying the inhibiting substance — other conditions remaining unchanged.

Preliminary experiments with some antibacterial substances (such as hydrogen peroxide, urea peroxide, sodium benzoate, methyl salicylate, cetyl pyridinium chloride) suggested the choice of sodium fluoride, penicillin, and aureomycin for subsequent tests.

Methods.

A. The inhibitory effect of sodium fluoride was studied in the following way. After giving a sample of the mixed resting saliva the subject rinsed his mouth for one minute with 15 ml of a 0.5 % solution of sodium fluoride. This was the highest concentration having no unpleasant taste. Immediately after

this rinse the subject was required to ingest cane sugar, following the standard procedure, after which further samples were collected at the specified intervals.

B. The inhibitory effects of penicillin and aureomycin were tested with trochi designed for oral application. These trochi consisting essentially of sucrose, in addition to the active agents, (see Table 7), no additional sugar ingestion was required. The

Table 7.

Composition of penicillin and aureomycin trochi.

Penicillin	Milligrams
Sodium Penicillin G	0.66
Lactose	20
Sucrose	970
Magnesium stearate	10
Aureomycin	
Aureomycin hydrochloride	15
Gum arabic	40
Sucrose	1,430
Magnesium stearate	7.5
Oil of peppermint	6
Colour q. s.	

trochi were allowed to melt completely in the mouth, this requiring an average time of about 6 minutes for the penicillin trochi and 10 minutes for the aureomycin trochi. The intervals for the standardized collection of saliva samples were reckoned from the completion of this ingestion. Tests were never performed less than three days after any previous ingestion of antibiotics.

In order to test the prolonged effect of penicillin and aureomycin, standard series of analyses were run also in connection with standard cane sugar ingestion 5 hours after the administration of the antibiotics.

The penicillin tests were performed with two batches of the same composition — Preparations I and II —, these being used 4—18 and 4—9 months, respectively, after date of manufacture.

Since the aureomycin trochi contained some peppermint oil which might have exerted some inhibitory effect *per se*, control tests were made with two of the subjects using trochi of the same composition except for the aureomycin. Normal lactate curves were obtained.

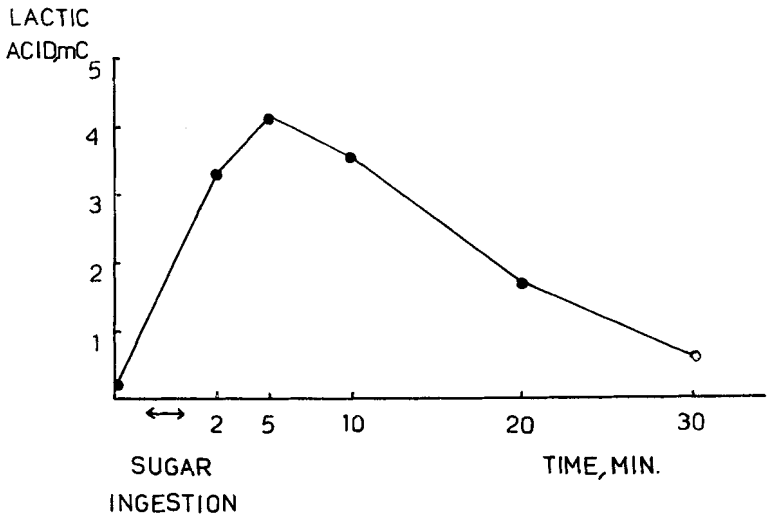

Fig. 1. Average salivary lactate curve of 10 subjects.

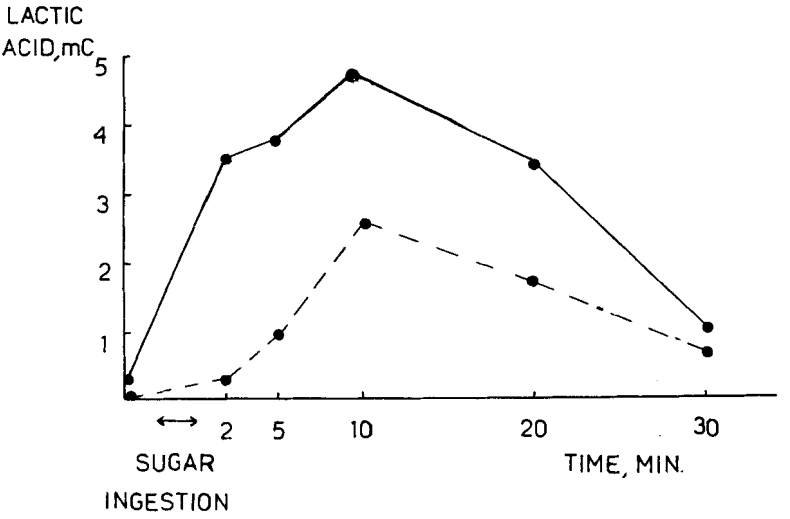

Fig. 2. Typical curves showing effect of fluoride.
———— The subject's average normal curve
— — — » » curve after fluoride mouthwash.

Results.

The average normal lactate curve for the 10 subjects is given in Fig. 1. The maximum values ranged between 2.84 and 5.96 mC and occurred after 5 minutes in 7 cases, after 10 minutes

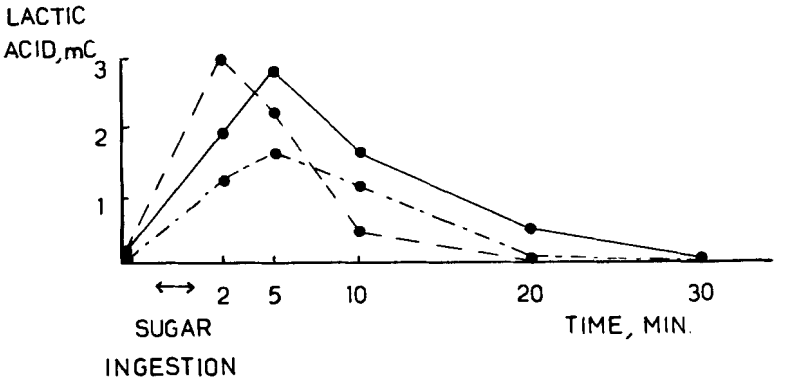

Fig. 3. Typical curves showing effect of penicillin.
———— The subject's average normal curve
— — — — » » curve immediately after penicillin II
—·—·—·— » » curve 5 hours after penicillin II.

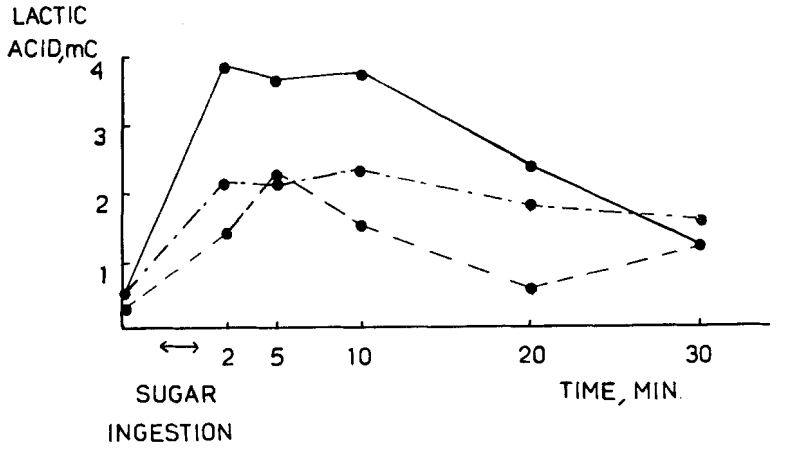

Fig. 4. Typical curves showing effect of aureomycin.
———— The subject's average normal curve
— — — — » » curve immediately after aureomycin
—·—·—·— » » curve 5 hours after aureomycin.

in 2, and after 2 minutes in 1 case. Typical individual curves for the different inhibitors are given in Figs. 2—4.

The percentage reduction of the salivary lactic acid was calculated for each subject and each interval after ingestion. Tables 8—14 give the maximum, minimum and mean (M) reductions. The P value, as calculated according to Student's t-test, and its estimated significance are also given except for the values

Table 8.

Effect of 0.5 % sodium fluoride solution.

	Percentage reduction in lactic acid				
	2 min.	5 min.	10 min.	20 min.	30 min.
Max., min.	97.8 — 91.0	93.4 — 15.6	78.5 — 17.1	80.6 — 16.5	100 — 26.7
Med.	57.7	65.6	51.9	32.1	50.8
P	< 0.01	< 0.001	< 0.001	> 0.05	
Sign.	XX	XXX	XXX	0	

Table 9.

Immediate effect of penicillin, Preparation I.

	Percentage reduction in lactic acid				
	2 min.	5 min.	10 min.	20 min.	30 min.
Max., min.	75.4 — 47.9	100 — 30.4	100 — 6.7	100 — 55.6	100 — 48.9
Med.	13.0	43.7	64.0	59.5	— 5.7
P	> 0.05	< 0.01	< 0.001	< 0.01	
Sign.	0	XX	XXX	XX	

Table 10.

Effect of penicillin, Preparation I, after 5 hours.

	Percentage reduction in lactic acid				
	2 min.	5 min.	10 min.	20 min.	30 min.
Max., min.	68.0 — 66.4	94.7 — 65.9	71.3 — 7.6	100 — 6.0	100 — 227.0
Med.	26.5	32.7	47.6	52.1	5.2
P	> 0.05	< 0.05	< 0.001	< 0.001	
Sign.	0	X	XXX	XXX	

Table 11.

Immediate effect of penicillin, Preparation II.

	Percentage reduction in lactic acid				
	2 min.	5 min.	10 min.	20 min.	30 min.
Max., min.	66.9 — 57.8	77.3 — 6.9	96.7 — 13.8	100 — 46.7	100 — 212
Med.	0.7	27.3	51.9	82.2	61.7
P	> 0.05	< 0.01	< 0.001	< 0.001	
Sign.	0	XX	XXX	XXX	

Table 12.

Effect of penicillin, Preparation II, after 5 hours.

	Percentage reduction in lactic acid				
	2 min.	5 min.	10 min.	20 min.	30 min.
Max., min.	97.2 — 11.1	76.8 — 12.2	73.3 — 3[illegible].0	100 — 89.6	100 — 300.0
Med.	31.4	27.0	26.8	34.8	7.4
P	< 0.01	< 0.05	< 0.05	> 0.05	
Sign.	XX	X	X	0	

Table 13.

Immediate effect of aureomycin.

	Percentage reduction in lactic acid				
	2 min.	5 min.	10 min.	20 min.	30 min.
Max., min.	96.0 — 42.2	88.0 — 37.6	83.9 — 47.9	87.3 — 127.0	88.4 — 911.0
Med.	47.0	43.0	49.5	35.6	— 86.6
P	< 0.01	< 0.01	< 0.01	> 0.05	
Sign.	XX	XX	XX	0	

Table 14.

Effect of aureomycin after 5 hours.

	Percentage reduction in lactic acid				
	2 min.	5 min.	10 min.	20 min.	30 min.
Max., min.	100 — 1.73	92.8 — 27.3	99.0 3.7	56.0 — 115.0	100 — 1,560
Med.	56.4	48.6	57.7	4.4	— 230.2
P	< 0.001	< 0.01	< 0.001	> 0.05	
Sign.	XXX	XX	XXX	0	

obtained at 30 min., which were excluded on account of their skew distribution. The symbol P denotes the probability of getting the observed lactate reductions solely by chance. The significance has been conventionally estimated and marked as follows:

XXX = highly significant difference ..... ($P \leqq 0.001$),
XX = significant difference ..... ($0.001 < P < 0.01$),
X = probable difference ..... ($0.01 < P < 0.05$),
0 = non-significant difference ..... ($0.05 < P$).

It appears from the tables, that all the tested substances have given reductions of the lactic acid formation which at most intervals are of more or less pronounced significance. The effect of penicillin and aureomycin is of the same order after 5 hours as immediately on the application.

Discussion.

The experiments have demonstrated the partial inhibitory effect of the test substances on intra-oral acid fermentation. Although the relative part played by the plaque bacteria is not clear it seems probable that the inhibition applies to the same extent to these as to other oral bacteria, implying a direct influence on the caries-producing mechanism. The results presented are thus in agreement with several results obtained with other methods: inhibition by penicillin of acid fermentation in saliva-carbohydrate mixtures *in vitro* (ZANDER and BIBBY, 1947); reduction of the caries activity in animals by the application of penicillin (MCCLURE and HEWITT, 1946; WEBMAN and collab., 1949; ZANDER and collab., 1951); the same effect with penicillin, aureomycin and other antibiotics (STEPHAN and collab., 1952), caries reduction in man by the application of penicillin (ZANDER, 1950); reduced lactobacillus counts in rat by the application of penicillin (MCCLURE and HEWITT, 1946; WEBMAN and collab., 1949) and in man (LUDWICK and FOSDICK, 1950). Some of the quoted authors have, however, had negative results with aureomycin, although this antibiotic strongly reduces the oral flora (KRASSE and collab., 1950). The results obtained with sodium fluoride are evidently due to its enzyme-inhibiting effect and are not directly comparable with the main part of the fluorin-caries studies where the solubility-decreasing effect may have played some part.

The results reported here do not of course warrant any extensive practical application of these inhibitors for caries prophylaxis. Risks of hypersensitiveness and harmful disturbances of the balance between bacterial species are familiar limiting factors in the use of penicillin and aureomycin. It is, however, quite conceivable that the rapid development in this field may lead to antibiotics having local caries-inhibiting effects without these restrictive disadvantages. The more universal employment of fluoride mouth-washes is precluded by inadequate knowledge of the resorption through the oral mucosa and the local and general

toxicity, even if no damage or discomfort was observed in our experiments.

Summary.

Experimental evidence indicates that the increased lactate content of the saliva after carbohydrate ingestion is not derived from the glands nor is it produced within the saliva, but originates in the greater bacterial aggregates of the oral cavity. Of these, the dental plaque is neither the sole nor the most important source.

The lactic acid formation is in part inhibited by local application of penicillin, aureomycin, and a 0.5 % solution of sodium fluoride. The inhibition is also demonstrable five hours after the supply of penicillin or aureomycin.

Zusammenfassung.

Die experimentellen Ergebnisse zeigen, dass der gesteigerte Lactatgehalt des Speichels nach Zufuhr von Kohlehydraten nicht von den Drüsen herrührt und nicht im eigentlichen Speichel gebildet wird, sondern ihren Ursprung in den grösseren Bakterienansammlungen der Mundhöhle hat. Unter diesen Ansammlungen ist der dentale Belag nicht die einzige oder vorherrschende Quelle.

Die Milchsäureproduktion wird teilweise gehemmt durch lokale Applikation von Penicillin, Aureomycin und halbprozentiger Lösung von Natriumfluorid. Die Hemmung ist auch erweisbar fünf Stunden nach der Zufuhr von Penicillin oder Aureomycin.

Résumé.

Les résultats expérimentaux indiquent que le lactate provenant de la salive après l'ingestion d'hydrate de carbone ne se forme pas dans les glandes, ni dans la salive elle-même, mais il provient des grandes concentrations de bactéries de la bouche.

La plaque dentaire n'est pas l'unique source du lactate, ni la plus dominante.

La formation d'acide lactique est partiellement atténuée par l'application locale de pénicilline, d'auréomycine ou d'une solution de 0.5 % de fluorure de sodium. Cette atténuation est même démontrable cinq heures après l'application de pénicilline ou d'auréomycine.

References.

BARKER, S. B., and SUMMERSON, W. H.: The colorimetric determination of lactic acid in biological material. J. Biol. Chem. 138: 535, 1941.

ERICSSON, Y.: Enamel-apatite solubility. Acta Odont. Scand. 8: Suppl. 3, 1949.

ERICSSON, Y. and HELLSTRÖM, I.: Salivens mjölksyrehalt efter kolhydrattillförsel. (The lactic acid content of the saliva after carbohydrate ingestion.) Sv. Tandl. Sällsk. Festskr. 1950, p. 115.

HILL, T. J., and WHITE, B. J.: Acid production in saliva in in-vitro experiments. J. Dent. Res. 28: 391, 1949.

KRASSE, B., LUNDQVIST, C., and WINBLAD, S.: Aureomycinets lokala effekt på den aeroba munfloran hos friska. Odont. Revy 1: 28, 1950.

MCCLURE, F. J., and HEWITT, W. L.: The relation of penicillin to induced rat dental caries and oral L. acidophilus. J. Dent. Res. 25: 441, 1946.

MUNTZ, J. A.: Production of acids from glucose by dental plaque material. J. Biol. Chem. 148: 225, 1943.

MÖLLMANN, F.: Die Milchsäure im menschlichen Speichel und ihre Herkunft. Dissertation. Frankfurt am Main 1935.

NEUWIRTH, I., and SUMMERSON, W. H.: The utilization of lactic and pyruvic acids by oral microorganisms. J. Dent. Res. 21: 241, 1942.

SREEBNY, L. M., KIRCH, E. R., and KESEL, R. G.: The location of the glycolytic enzymes in saliva. J. Dent. Res. 29: 506, 1950.

STEPHAN, R. M.: Hydrogen ion concentration of the dental plaque. J. Dent. Res. 17: 251, 1938.

— Changes in hydrogen-ion concentration on tooth surfaces and in carious lesions. J. A. D. A. 27: 718, 1940.

STEPHAN, R. M., and collaborators: The comparative effects of penicillin, bacitracin, chloromycetin, aureomycin, and streptomycin on experimental dental caries and on certain oral bacteria in the rat. J. Dent. Res. 31, 1952 (in press).

SUMMERSON, W. H., and NEUWIRTH, I.: The production of acids from glucose by oral microorganisms. J. Dent. Res. 20: 157, 1941.

ZANDER, H. A.: Effect of penicillin dentifrice on caries incidence in school children. J. A. D. A. 40: 569, 1950.

ZANDER, H. A., and BIBBY, B. G.: Penicillin and caries activity. J. Dent. Res. 26: 353, 1947.

ZANDER, H. A., LISANTI, V. F., and SHIERE, F. R.: The validity of hamster studies in caries control. J. Dent. Res. 30: 139, 1951.

VOLKER, J. F., and PINKERTON, D. M.: Acid production in saliva-carbohydrate mixtures. J. Dent. Res. 26: 229, 1947.

WEBMAN, H., HILL, T. J., and KNIESNER, A. H.: The effect of penicillin on dental caries in rats fed on a coarse corn diet. J. Dent. Res. 28: 258, 1949.

WESTIN, G.: Personal communication.

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