

# Turku sugar studies VIII

## Principal microbiological findings

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The possible qualitative and/or quantitative alterations in the proportions of cultivable groups of oral microorganisms were analysed during a clinical trial involving the consumption of fructose (F) or xylitol (X) in comparison to sucrose (S). Supragingival plaque samples and paraffin-stimulated saliva were collected from 115 subjects. The samples were dispersed by sonication, diluted stepwise, plated on blood agar, Mac Leod agar, MacConcey agar, Rogosa S.L. agar, and Sabouraud agar plates and incubated anaerobically and/or aerobically. The number of the total colony forming units (CFU) on blood agar plates in anaerobic incubation was about  $1-3 \times 10^9$ /ml saliva and  $1-4 \times 10^8$ /mg plaque and in aerobic respectively  $5-18 \times 10^8$ /ml saliva and  $10^8$ /mg plaque. The total CFU on Mac Leod agar was of a similar order of magnitude. The variation between subjects and consecutive determinations was relatively large. The arithmetic mean of the total CFU on MacConcey agar was about  $1-5 \times 10^6$ /ml saliva, on Rogosa S.L. agar  $6-130 \times 10^3$ /ml saliva and on Sabouraud about  $1-2 \times 10^8$ /ml saliva, all in aerobic incubations. Replacement of dietary sucrose with xylitol did not affect the proportion of major microbial categories in saliva or dental plaque. The percentage of typical streptococcal colonies on blood agar was of a similar order of magnitude (about 60—70 %) during the diets. The arithmetic and geometric means of the total CFU values on Rogosa and Sabouraud agar plates were significantly lower in the X-group than in the S- or F-groups after a diet period of some months. It was thought that the reason for the reduction of acidogenic and aciduric oral flora in the X-group was partly due to the fact that xylitol is generally not metabolized by these microorganisms.

*Key-words:* Microbiology; saliva; plaque; sucrose; fructose; xylitol

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Diet has been listed among the most prominent environmental factors which may affect the composition of the oral flora (*Hardie & Bowden, 1974; Geddes & Jenkins, 1974*). A dietary change may thus lead to an alteration of the ecological balance of the microbial flora and the development of a new infection in the alimentary system. The changes may be either harmful to or desirable for the health of the individual. In the present

study an attempt was made to analyse possible alterations in the oral flora during a clinical trial involving the consumption of fructose (F) or xylitol (X) in comparison to sucrose (S).

Various methods have been employed to determine the microbial composition of the oral flora. These include direct microscopic examination, the use of selective culture media for the isolation and enumeration of specific organisms,

and the use of non-selective media and identification of various colony types (Gibbons *et al.*, 1964). However, most of the culture methods reveal only a fraction of the total microflora (Strålfors, 1950; Socransky *et al.*, 1963; Gibbons *et al.*, 1964; Slack & Bowden, 1965; Handelman & Hess, 1969).

When studying the oral flora it is important to select methods suited for the aims of the study. The present trial faced two main problems: (1) possible qualitative and/or quantitative alterations in the proportions of the predominant cultivable groups of microorganisms on non-selective media in highly diluted samples, using blood agar and MacLeod chocolate agar plates as culture media and (2) the enumeration of specific organisms using selective MacConcey, Rogosa S.L. agar and Sabouraud agar culture media (Figs. 1, 2). The role of streptococci was thought to be so important in the etiology of dental caries and the formation of plaque that findings concerning them have been studied separately (Gehring *et al.*, 1975). It is well known that saliva contains large numbers of bacteria. These organisms have been thought to be dislodged from dental plaque or from gingival debris as well as from the dorsum of the tongue through the washing action of saliva (Gibbons *et al.*, 1964). Consequently, it was thought that saliva could be utilized as a substitute for the major ecosystems in the oral cavity and that major alterations would be reflected in it. In addition, changes in plaque microbiology were analysed, particularly in view of the primary role of dental plaque in the etiology of dental caries and gingivitis. Dental plaque, mainly from smooth tooth surfaces, was thought to be representative of the specific ecosystems in the mouth.

#### MATERIAL AND METHODS

*Subjects.* A detailed description of the subjects participating in the Turku sugar trial as well as the general arrangement of the 2-year diet period has been given separately (Mäkinen & Scheinin, 1975 a). The final material consisted of 33 subjects in the S-, 35 in the F- and 47 in the X-group. The caries incidence (Scheinin, Mäkinen & Ylitalo, 1975) as well as the development of the gingival conditions (Paunio *et al.*, 1975) has been given separately. Oral biochemical findings including quantitations of plaque (Mäkinen & Scheinin, 1975 b) as well as the results of the analyses of the parameters of general health have also been reported (Huttunen, Mäkinen & Scheinin, 1975; Mäkinen & Scheinin, 1975 c).

*Collection of samples.* In connection with the clinical examinations supra-gingival plaque samples from the buccal and lingual surfaces of all teeth as well as from the approximal surfaces of upper and lower incisors were obtained using sterile periodontal curettes. The material was collected from the proximity of the cervical margin, not, however, including fillings or crowns, and also excluding the intracrevicular part of the gingival pockets. Areas where the development of caries could occur were especially included.

The sample was collected on a sterile watch-glass. A constant amount about 1 mg in weight of each plaque sample was taken with a sterile instrument with a circular depression, 1 mm in diameter, 0.5 mm deep. The watch-glass with the plaque sample was weighed and reweighed after the removal of the sample and the exact weight of the sample was thus obtained. The collection of plaque was performed in five minutes and the weighing in one minute.

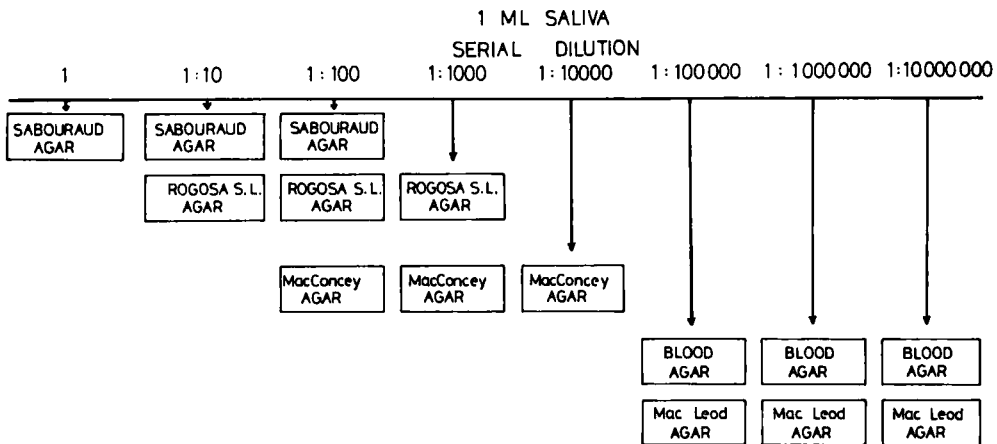


Fig. 1. Scheme for the serial dilution of saliva

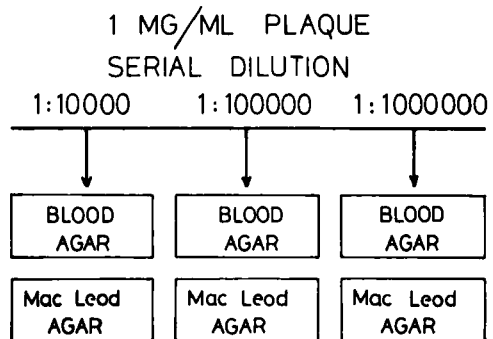


Fig. 2. Scheme for the serial dilution of dental plaque.

After the plaque collection, paraffin-stimulated saliva samples (about 5 ml) were collected. Samples with blood contamination were discarded and in these few cases collection was started from the very beginning after the bleeding had ceased.

The collections were performed between 8.30 a.m. and 3.00 p.m. In duplicate determinations performed at the onset of the experiment, after one year's diet, and after two years' diet, the second sample was taken on the following day between 8.00 and 8.30 a.m. The subjects came to the microbial sampling procedure without having brushed their teeth for 12—24 hours.

*Sample dispersions.* The plaque samples were dispersed by sonication with an ultrasonic disintegrator (100 W, Measuring & Scientific Equipment Ltd., London, England) using a microprobe with 3 mm end diameter. The sonication was performed for 30 sec with a amplitude of 7  $\mu$ m. Similar sonication was performed for 5 sec for 1 ml saliva samples. After these procedures, the sample suspensions (1 ml) were serially diluted stepwise to concentrations of 1:10, 1:100, 1:1000, 1:10000, 1:100000, and 1:1000000 and/or 1:10000000 (Figs. 1 and 2).

*Growth media.* The agar media tested were:

1. *Blood agar*, 7% hemolyzed horse

- blood (Orion Diagnostica, Helsinki, Finland) for non-selective total growth.
2. *MacLeod chocolate agar* (blood agar base) with 7% heated, defibrinated horse blood (Orion Diagnostica) for nonselective total growth.
  3. *MacConcey agar*, a bile salt lactose agar (Orion Diagnostica) for Gram negative bacteria, especially for *Enterobacteriaceae* population.
  4. *Rogosa S. L. agar* (Orion Diagnostica) for the *Lactobacillus* population.
  5. *Sabouraud dextrose agar* with 12 mg/ml penicillin, 53 mg/ml Streptomycin and 0.5 mg/ml actidione (Orion Diagnostica) selective for yeasts.

**Sample plating.** One-tenth ml of the dilutions were plated on various growth media using three different dilutions for each media as indicated in Figs. 1 and 2. The time from sample collection to completion of spreading plates was maintained at 2 h or less by the use of 3 operators. Between inoculations the samples were stored in ice.

**Incubation.** Blood agar and MacLeod agar plates were incubated aerobically at 37°C for three days, as well as anaerobically in an atmosphere of nitrogen. MacConcey agar plates were incubated for two days, Rogosa S.L. agar plates for four days and Sabouraud antibiotic agar plates for seven days at +37°C aerobically.

**Differential colony counts.** The counts of the colonies on each of the plates were performed both in daylight without magnification and under 2.5 × magnification. The colonies on the most highly diluted samples of each series of media were identified and counted. These counts were then made for the two other dilutions. Total colony forming unit (CFU) values exceeding 100, however, were not counted for the entire sample, but estimated by

counting the number of colonies on a constant area and then multiplying the result to represent the whole area of the medium. The principle used for obtaining the estimated mean value included the use of actual counts ranging from 20—100 in the dilution rate, if possible. For example, when the CFU-values on three different dilutions were,  $800 \times 10^6$  (estimate),  $78 \times 10^7$  and  $9 \times 10^8$ , the value  $78 \times 10^8$  was selected to represent the mean value.

**Identification principles.** The following criteria were used in the identification of various colony types on different media and thereafter in dividing the microbes roughly into the following categories:

Blood agar: (1) *Streptococcus*  $\alpha$ -hemolyticus, colonies 0.3—0.8 mm in diameter, dark, semi-transparent, convex colonies producing a zone of partial hemolysis with greenish discoloration surrounding the colonies. This group represented mainly *Streptococcus viridans*, but some *Diplococcus pneumoniae* strains were also included.

(2) *Streptococcus* non-hemolyticus, colonies of 0.4—1.5 mm in diameter, circular, dark, low convex, no alteration in the medium. This group represented *Streptococcus faecalis* and various facultative and anaerobic streptococci.

(3) *Streptococcus*  $\beta$ -hemolyticus, 0.6—1.0 mm in diameter, circular, dark, discrete, low-convex disks showing  $\beta$ -hemolysis. These were considered to represent *Streptococcus pyogenes* species.

(4) A group of greyish colonies, small 0.5—2 mm in diameter, grey, round, opaque and dry colonies as well as grey transparent disks of smooth and rough form, were thought to represent *Neisseria*, *Veillonella* and *Corynebacteria* genera, but small *Enterobacteria*, and in anaerobic

cultivation *Bacterioides* colonies could be included.

(5) A group of white colonies, colonies of 1—3 mm in diameter, white or cream coloured, round, circular, transparent or opaque as well as dry were included. These were mainly without hemolysis but  $\beta$ -hemolysis was allowed. This group was considered to include *Lactobacillus*, *Staphylococcus* and *Candida* genera in aerobic and also *Actinomyces* in anaerobic cultivation.

(6) A group of unidentified colonies. In this group colony types of irregular morphology and colour (mainly large, often  $\beta$ -hemolytic, grey, white or creamy as well as dark) were included. Genera which were morphologically classified and included in this group were, among others, *Escherichia coli*, *Aerobacter*, *Proteus*, *Klebsiella*, *Pseudomonas*, *Leptothrichia* and *Clostridia*.

Mac Leod agar: (1) Streptococcus hemolyticus colonies with green hemolysis, similar to those included on blood agar in group Streptococcus  $\alpha$ -hemolyticus. This group included, among others, *Streptococcus viridans*, *Streptococcus pyogenes* and *Diplococcus pneumoniae*.

(2) Streptococcus non-hemolyticus, criteria similar to those on blood agar.

(3) A group of greyish colonies, criteria as in group 4 on blood agar. This group also included *Haemophilus* strains.

(5) A group of unidentified colonies; see group 6 on blood agar.

(6) Total CFU-value on Mac Leod agar.

MacConcey agar: (1) *Escherichia coli*, moderately large, rose-pink, thick, circular discs and smooth colonies. This group may include *Klebsiella* and *Aerobacter* strains, as well.

(2) Neisseria and streptococci group,

minute, 0.5—1 mm in diameter, grey rough as well as dry colonies.

(3) Klebsiella group, large (over 3 mm in diameter), raised, very viscous and pink colonies, probably representing *Klebsiella* species.

(4) A group of white colonies, opaque, 1—2 mm in diameter, smooth and white colonies, probably representing *Lactobacillus* population, were occasionally observed.

(5) Total CFU-value on MacConcey agar.

Rogosa S. L. agar: (1) Lactobacillus, round 1—3 mm in diameter, colour ranging from opaque white to light yellow.

(2) Streptococci and diptheroid colonies, minute, 0.2—0.9 mm in diameter, circular semi-transparent discs.

(3) Candida, more than 3 mm in diameter, intensely white with a typical odour.

(4) Total CFU on Rogosa agar.

Sabouraud antibiotic agar: (1) Candida large, more than 2 mm in diameter, white and smooth colonies with a typical odour.

(2) Other yeast species.

(3) Total CFU on Sabouraud agar.

*Statistical treatment.* In this study non-parametric tests for significance were performed. (1) The Wilcoxon test was used when the reproducibility of the microbial analyses on the basis of duplicate determinations carried out at the onset, at the 12-months phase, and at the end of the study was tested; these analyses were performed with all the subjects as a whole; (2) variations during the study within each sugar groups were tested with the Friedman-test; (3) the significance of the differences between the sugar groups as tested with the Kruskal-Wallis test, as well as with the Mann-Whitney U-test

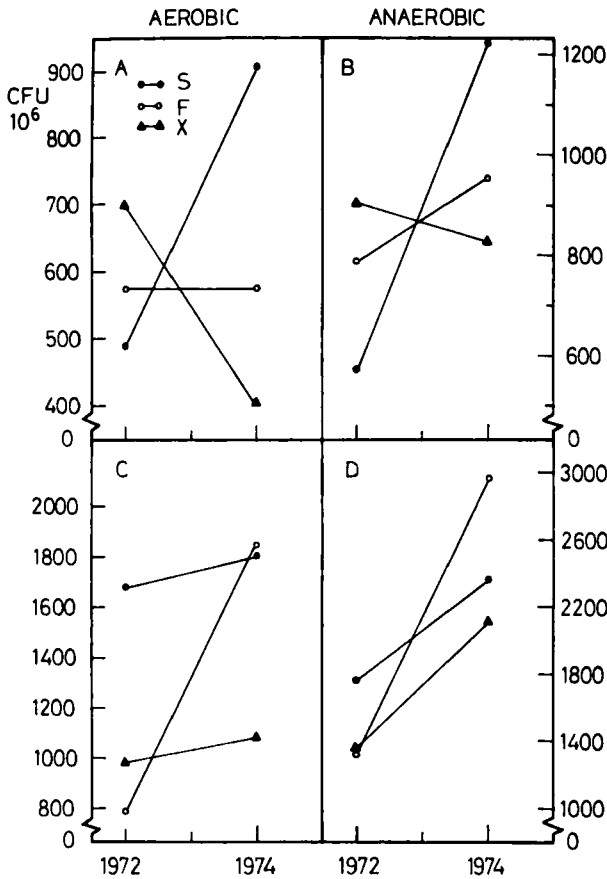


Fig. 3. The geometric means (A, B) and arithmetic means (C, D) of the total salivary CFU-values ( $n \times 10^6$ ) on aerobic (A, C) and anaerobic (B, D) incubations on blood agar plates at the onset (1972) and termination (1974) of the sugar diet. S = sucrose; F = fructose; X = xylitol.

between the sugar group pairs at each examination period (Bradley, 1968; Sachs, 1972).

#### RESULTS

*Salivary microbial population on blood and MacLeod agar plates.* The overall predominant viable microbial composition in the saliva of all the subjects at the baseline examination before the onset of the experiment revealed that the magnitude of the total CFU on blood agar plates was about  $1-2 \times 10^8$ /ml saliva in anaerobic and  $5-15 \times 10^8$  in aerobic cultivation (Figs. 3-5, Tables I, II). The range between subjects was, however,

relatively large; variations of the magnitude of  $10^8-10^4$  could be observed (Tables I, II). The mean counts between samples obtained on two consecutive days at the baseline examination differed in about 2/3 of the cases significantly (Tables I and II).

Similar results of the same magnitude were obtained with the MacLeod chocolate agar (about 1/2 of the cases differed significantly between the duplicate determinations).

At the termination of the experiment, the difference between the samples taken on two consecutive days were generally no longer significant. On blood agar plates, only a few cases revealed significant

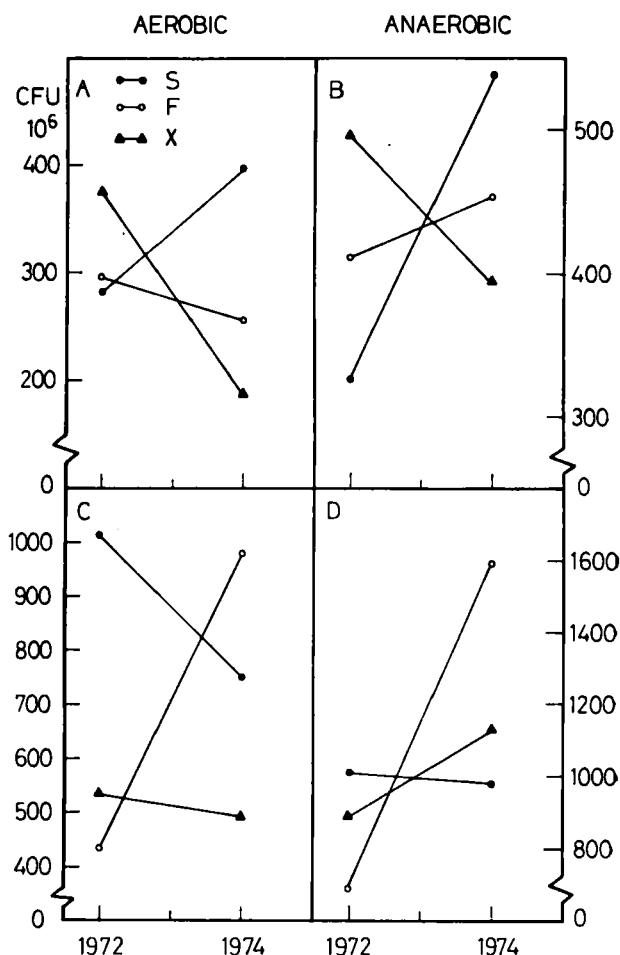


Fig. 4. The geometric means (A, B) and arithmetic means (C, D) of the salivary CFU-values of *Streptococcus α-hemolyticus* ( $n \times 10^6$ ) on aerobic (A, C) and anaerobic incubations on blood agar plates at the onset (1972) and termination (1974) of the sugar diet. S = sucrose; F = fructose; X = xylitol.

differences (Tables III & IV) and on MacLeod agar only one.

In the calculations of the possible changes in the colony counts during the two year diets, the geometric means of the first and second duplicate tests at the onset and at the termination of the diet period were used. The arithmetic mean values tended to increase in all sugar groups. Geometric means in anaerobic incubations increased in the S- and F-groups and decreased in the X-group (Figs. 3–5, Tables V–VIII). These changes within the different sugar groups were occasionally statistically significant as seen in Tables IX and X.

There were practically no significant differences between the sugar groups at the onset of the experiment. This situation remained practically unchanged during the experiment. Table XI shows an example of the statistical analyses performed and indicates that the differences between the sugar groups were significant in a few cases only. On blood agar, however, a statistically significant decrease was demonstrable in the CFU-values of *Streptococcus α-hemolyticus*, *Neisseria* and *Lactobacillus* groups as well as in total CFU in X-group in comparison to the S-group (Table XI).

The streptococci predominated numeri-

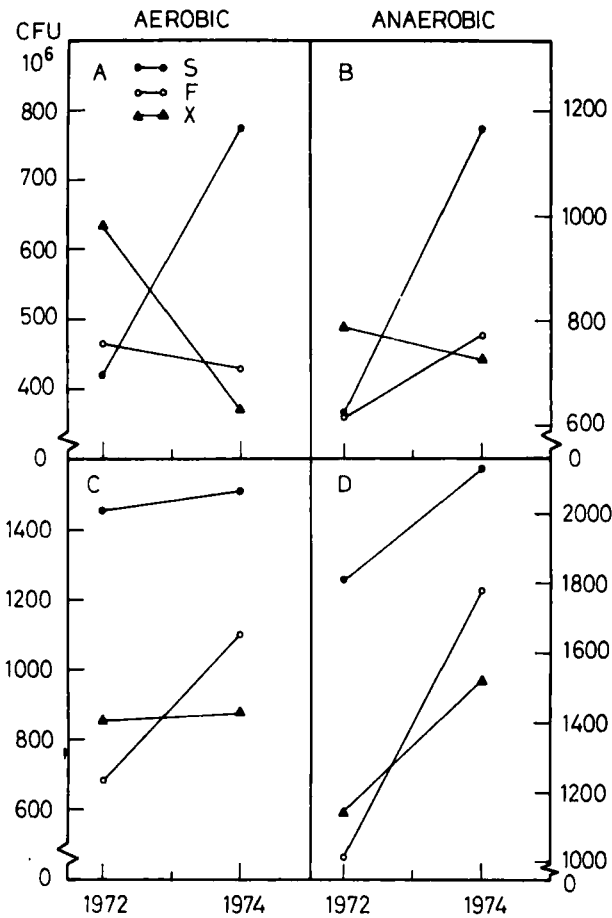


Fig. 5. The geometric means (A, B) and arithmetic means (C, D) of the total salivary CFU-values ( $n \times 10^6$ ) on aerobic (A, C) and anaerobic (B, D) incubations on Mac Leod agar at the onset (1972) and termination (1974) of the sugar diet. S = sucrose; F = fructose; X = xylitol.

cally (70–80 % of the total CFU) in arithmetic calculations both in aerobic and anaerobic cultivations on blood agar plates (Table XII).

*Microbial population of dental plaque on blood agar and Mac Leod agar plates.* The arithmetic mean of the viable bacterial population both on blood and Mac Leod agar plates was of the magnitude of  $1-4 \times 10^8$ /mg plaque in anaerobic and about  $10^8$  in aerobic incubation (Figs. 6–8, Tables XIII–XVI). Again the variation between individuals was relatively large; 1000 fold variations could be observed. The mean counts in the samples

taken on two consecutive days at the baseline examination did not differ significantly (Tables XIII & XIV) at the onset of the study, whereas at the termination of the experiment significant differences were demonstrable (Tables XV & XVI).

In the calculations of the possible changes in the colony counts during the two year diet period, the mean values of the first and second duplicate tests at the onset and at the termination of the diet period were used. Both the arithmetic and geometric mean values tended to increase within the S- and F-groups, whereas in

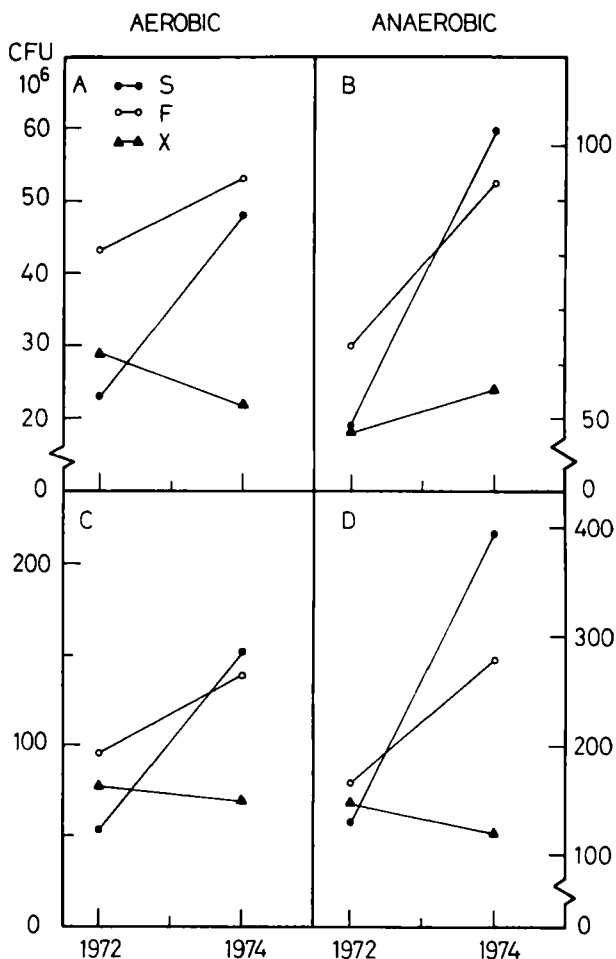


Fig. 6. The geometric means (A, B) and arithmetic means (C, D) of the total CFU-values of dental plaque on aerobic (A, C) and anaerobic (B, D) incubations ( $n \times 10^6$ ) on blood agar, at the onset (1972) and termination (1974) of the sugar diet. S = sucrose; F = fructose; X = xylitol.

the X-group they decreased or remained on the same level (Figs. 6–8, Tables XVII–XX). The results of the Wilcoxon-test for these differences of geometric means were as presented in Tables XXI–XXII.

No significant differences between the different sugar groups were demonstrable in anaerobic cultivation at the onset of the experiment or at the termination of the diet period. After the diet period, significant differences between the groups are shown in Table XXIII.

The proportion of streptococcal CFU of the total CFU was relatively constant

(70–80 %) during the diet period (Table XXIV).

*Microbial population on MacConcey plates.* The arithmetic mean value of the total salivary CFU on MacConcey agar was  $1-5 \times 10^6$  during the whole diet period (Fig. 9, Table XXV). The main portion of the CFU was observed to be typical *E. coli* colonies (Fig. 10, Table XXVI). However, neisseria and some unidentified gram negative as well as gram positive bacterial colonies could also be seen (Table XXVII). Comparison between the duplicate determinations at the baseline examination, after one year's,

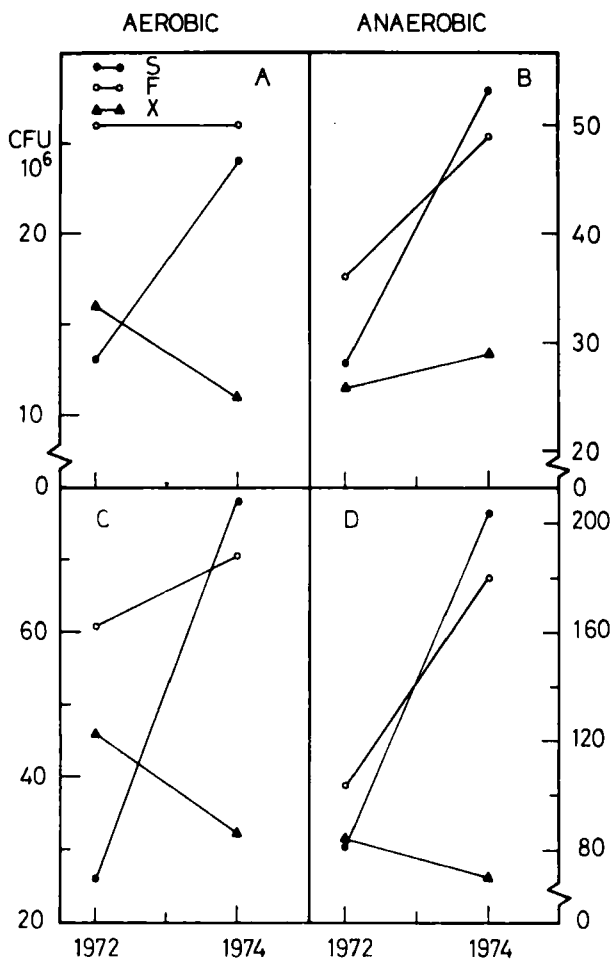


Fig. 7. The geometric means (A, B) and arithmetic means (C, D) of the CFU-values of *Streptococcus alpha-hemolyticus* ( $n \times 10^6$ ) of dental plaque on aerobic (A, C) and anaerobic incubations on blood agar, at the onset (1972) and termination (1974) of the sugar diet. S = sucrose; F = fructose; X = xylitol.

and two years' diet period revealed significant differences in *E. coli* and total CFU in the F- and X-groups after one year, and in the X-group after two years' diet period. The changes within the three sugar groups were generally statistically significant, as seen in Table XXVIII. No significant differences could be observed between the different sugar groups (the only exception being the total CFU on the second duplicate examination at the end of the diet period, Kruskal-Wallis  $\alpha = 0.0348$ ; U-tests between each sugar group, however, revealed no significant differences).

*Microbial population on Rogosa S. L. agar.* The arithmetic mean values of the total CFU on Rogosa S. L. agar varied from 6–130  $\times 10^8$  per ml saliva (Fig. 11, Table XXIX). In this case no significant differences could be observed when the Wilcoxon-Wilcoxon-test was used in the comparison between the duplicate determinations at the beginning, after 1 year and after 2 years. During the diet period, the total colony count and the number of lactobacilli and streptococci increased in both the S- and F-groups, whereas these values decreased or remained at the initial level in the X-group (Figs. 12–13, Tables

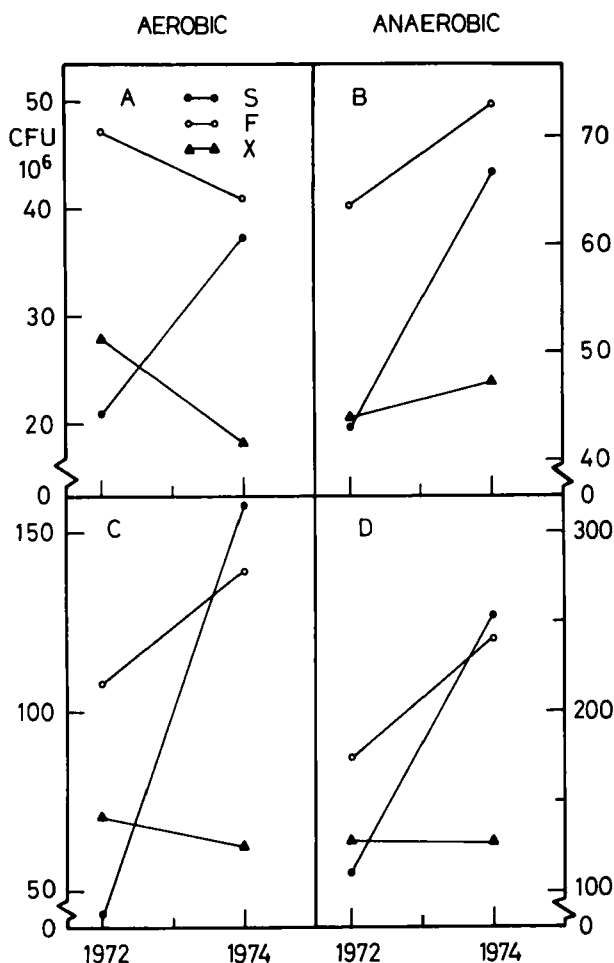


Fig. 8. The geometric means (A, B) and arithmetic means (C, D) of the total CFU-values ( $n \times 10^6$ ) of dental plaque on aerobic (A, C) and anaerobic (B, D) incubations on MacLeod agar at the onset (1972) and termination (1974) of the sugar diet. S = sucrose; F = fructose; X = xylitol.

XXIX—XXXII). These changes within the groups were statistically significant (Table XXXIII). The differences between the different sugar groups became statistically significant after 4 to 12 months' diet period (Table XXXIV).

The total CFU-value on Rogosa S. L. agar of one subject, (N:o 29) excluded from the study due to rampant caries and changed from fructose to xylitol consumption (not included in the X-group) decreased considerably at the point of diet change (Fig. 14). Clinically and radiographically detected dentinal lesions as well as new filled surfaces are presented in the same figures (Fig. 14).

*Microbial population on Sabouraud dextrose agar.* There were no statistically

significant differences in the numbers of salivary *Candida* (Table XXXV) in the S- and F-groups during the diet period, whereas the decrease (Fig. 15) observed in the X-group was statistically significant (Friedman test, 0.0045). Generally there was a statistically significant difference between the X-group on one hand and the F- and S-groups on the other (Table XXXVI).

*Variations of the CFU-values on different media.* Subjects with high counts on one medium generally had high counts on the others. This tendency was most marked for organisms cultured in large numbers.

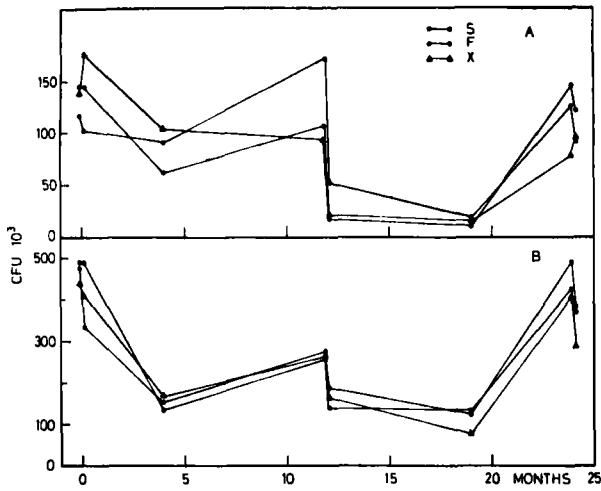


Fig. 9. The geometric means (A) and arithmetic means (B) of the total salivary CFU-values on MacConcey agar plates as a function of time and sugar group. S = sucrose; F = fructose; X = xylitol.

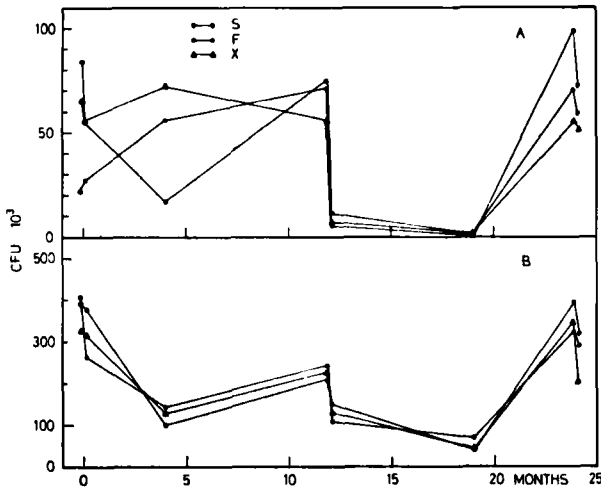


Fig. 10. The geometric means (A) and arithmetic means (B) of the salivary CFU-values of *E. Coli* on MacConcey agar as a function of time and sugar group. S = sucrose; F = fructose; X = xylitol.

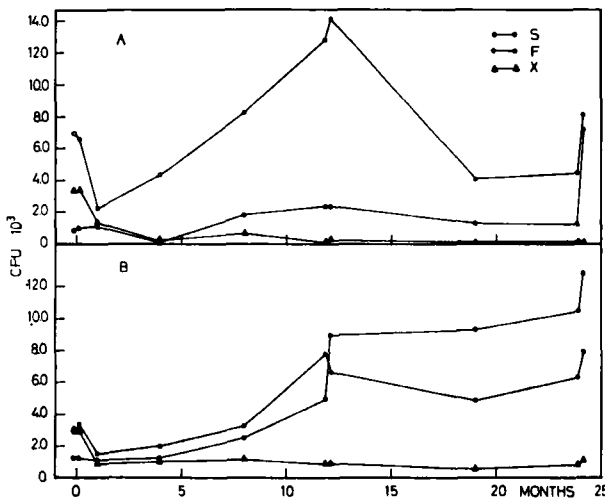


Fig. 11. The geometric means (A) and arithmetic means (B) of the total salivary CFU-values Rogosa S.L. agar as a function of time and sugar group. S = sucrose; F = fructose; X = xylitol.

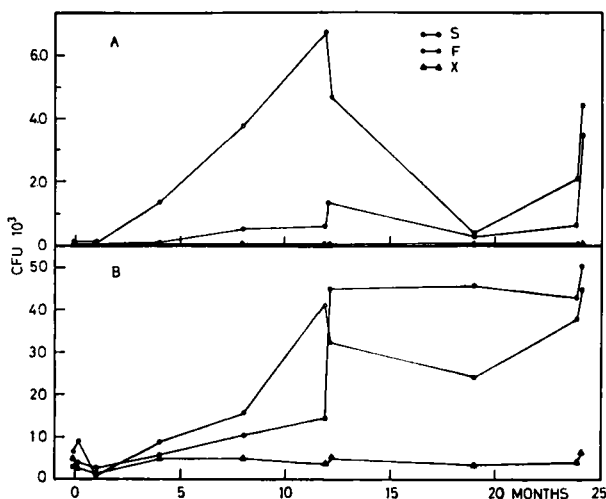


Fig. 12. The geometric means (A) and arithmetic means (B) of the salivary CFU-values of lactobacilli on Rogosa S.L. agar, as a function of time and sugar group. S = sucrose; F = fructose; X = xylitol.

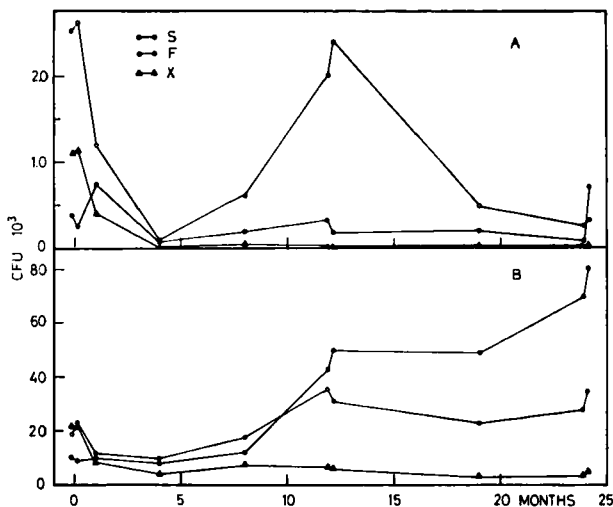


Fig. 13. The geometric means (A) and arithmetic means (B) of the salivary CFU-values of streptococci and diptherioids on Rogosa S.L. agar as a function of time and sugar group. S = sucrose; F = fructose; X = xylitol.

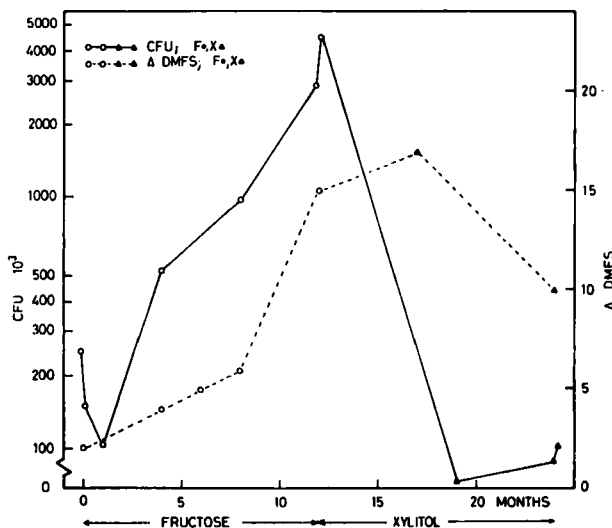


Fig. 14. The total salivary CFU-values on Rogosa S.L. agar in logarithmic scale (to the left) and clinically and radiographically detected dental lesions as well as new filled surfaces (to the right) of test person No. 29, excluded from the study due to rampant caries and changed from fructose to xylitol consumption.

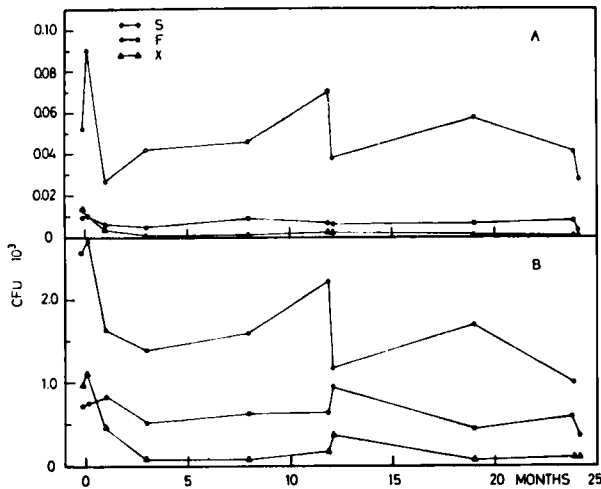


Fig. 15. The geometric means (A) and arithmetic means (B) of the total salivary CFU-values on Sabouraud antibiotic agar as a function of time and sugar group. S = sucrose; F = fructose; X = xylitol.

However, the CFU-values for streptococci, for example, on blood agar and Rogosa S. L. agar differed from each other at least 1000-fold.

#### DISCUSSION

The data obtained indicate that replacing dietary sucrose with xylitol or fructose did not affect the proportion of major microbial categories in highly diluted saliva or dental plaque samples. The percentage of typical streptococci colonies of the total CFU on blood agar, for example, was of a similar order of magnitude during the whole trial. On the other hand, the viable count-means on the highly selective Rogosa and Sabouraud media were significantly lower in the X-group than in the S- or F-groups. This decrease corresponds to the findings concerning the CFU-values of *Streptococcus mutans* in the same subjects (Gehring *et al.*, 1975). The present microbial findings are to be interpreted as relative: the samples from each person in the different sugar groups were collected, treated, cultivated and counted in an identical way, and are thus comparable at least to each other.

This laboratory was not equipped to culture large numbers of saliva and plaque samples under continuous anaerobic conditions. Thus, the conventional anaerobic method (aerobic collection and plating followed by anaerobic incubation) with its obvious disadvantages (Syed & Loesche, 1973) was the only possibility for carrying out these cultivations. Practical considerations did not allow the use of conventional anaerobic incubation for all of the media used, and thus the selective media were incubated aerobically only.

Originally an attempt was made to identify all the different colony types on both blood and MacLeod agar plates on the »genera» level, even on these unselective media. In practice, however, this goal was not accomplished because of the huge amount of subcultivations necessary. Thus different colony types were roughly put into morphological categories, and the taxonomic exactness of this study is not absolute. On the other hand, the total CFU-values as well as the streptococcal classification on these media can be considered reliable.

This study revealed that individual variation in the total CFU was unex-

pectedly large. Thus, the use of only three dilutions ( $10^n$ ,  $10^{n+1}$ ,  $10^{n+2}$ ) was observed not to be enough. The fact that streptococci were highly predominant on the nonselective media used evidently masked the formation of colonies of less common oral microorganisms on these media. The expression »not detected» used in this study means that certain colony types could not be seen in the smallest dilution used. It does not necessarily mean that these microorganisms are not present, but that their number is smaller than the detection limit, which in certain cases was as high as  $10^6$  per ml saliva. When using selective media, difficulties due to large variations between the predominant and minor microbial categories are not totally overcome. Evidently certain selective media have some undesirable selectiveness, as well. Thus the plaque streptococcal CFU-values on blood agar reported here differ considerably from those obtained with phenol red agar (Gehring *et al.*, 1974, 1975). This difference may be partly explained by the lyophilization procedure used in the latter study, but is partly due to the selectiveness of the phenol red agar.

The large variation in the total CFU-values on blood agar and MacLeod agar plates between the duplicate experiments revealed that the total number of oral microorganisms varies considerably. These findings are fully in agreement with those concerning the hour-to-hour variation during the day (Schjött *et al.*, 1970) which indicate that variations of over 10 fold magnitude occur daily in the arithmetic means of total CFU's. This variation was probably one reason for the fact that the relative decrease in the total CFU-values in the X-group during the diet period was not always statistically significant. Because the absolute amount of dental

plaque also decreased in the X-group (Mäkinen & Scheinin, 1975 b) the total number of plaque microorganisms was reduced even more than reported here in the group. This decrease did not affect the *inter-genera* variation of the dental plaque.

Streptococci represent a large part of the normal oral flora and the actual magnitude of the number of streptococci per mg plaque corresponds to that reported by Gibbons (1964). When the proportion of different groups of microorganisms from the total CFU-values are reported, the present findings are not, however, identical to those reported earlier (Howell *et al.*, 1965; Loesche, Hocket & Syed, 1972; Loesche & Syed, 1973; Bowden & Hardie, 1971). The proportion of streptococcal CFU of the total CFU is higher in the present study. It is known that the proportion of different microorganisms differs in dental plaque taken from different sites on the teeth, and the present figures correspond relatively well to those obtained when experimental plaque development is studied (Slack & Bowden, 1965), as well as to counts of fissure plaque (Theilade *et al.*, 1974; Thott *et al.*, 1974). These findings suggest that the plaque analysed in this trial represents a »young» plaque type.

The total salivary CFU-values on blood agar plates are similar to those recently reported by Schjött *et al.* (1970), but are higher than those reported by Richardson & Jones (1958). The present findings revealed a somewhat higher percentage of streptococcal CFU than reported by Gordon & Jong (1968), Slack & Bowden (1965) and Richardson & Jones (1958). The present figures are of a similar order of magnitude as those reported in infants and children (McCarthy *et al.*, 1965).

The total CFU value in the beginning of the trial on Rogosa agar is somewhat lower than generally reported (Richardson & Jones, 1958; Handelman & Mills, 1965; Frostell & Nord, 1972; Klein, Guinard & Frank, 1973). The main cause of this difference is thought to be connected with dietary habits, most of the subjects being dental students and dental school personnel. During the trial, however, the total CFU of Rogosa S. L. agar in the S- and F-groups significantly increased. This is thought to be connected with diet change in all sugar groups: although the subjects were advised not to change their normal dietary habits during the diet period, the possibility of getting various products free of charge for two years led to an increase in consumption. This was observed to be the cause of the increase in total CFU on Rogosa agar from zero to millions in at least some subjects, although this kind of diet change was not so common that it affected the mean value of the amount or frequency of sugar consumption (Mäkinen & Scheinin, 1975 b). It should be noted that this kind of rise in the total CFU values did not occur in the X-group, and the CFU values of the person who changed from F- to X-consumption during the trial decreased rapidly after the change.

In conclusion, neither fructose nor xylitol affected the ecological balance of microbial flora in highly diluted samples; on the other hand, xylitol reduced the number of most microorganisms, especially the acidogenic and aciduric flora representing a minor ecosystem in the oral cavity.

The effect seems to be due to the irrelevancy of xylitol, and other five carbon polyols in general, for microbial metabolism. It also appears that the reduction of oral *Candida* is explained

through findings (Mäkinen *et al.*, 1975) showing that certain yeast cells do not use xylitol for growth. Thus the reduction of the CFU of oral yeasts reported here has one natural explanation.

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# TABLES

Table I. Colony forming unit values of microbial categories per ml saliva on blood agar plates ( $n \times 10^6$ ) on aerobic cultivation at the baseline examination before the onset of the experiment in samples taken on two consecutive days as well as the comparison between the two examinations (Wilcoxon-test)

Microbial categories	Sample	N	Min.	Max.	Med.	$\bar{x}$ arith	S.D.	$\alpha$
Streptococcus $\alpha$ -hemolyticus	day 1	122	4	4000	350	439	469	0.0065
	day 2	122	4	20000	450	851	2169	
Streptococcus non-hemolyticus	day 1	122	2	1400	100	190	249	n.s.
	day 2	122	n.d.	5000	100	305	649	
Streptococcus $\beta$ -hemolyticus	day 1	122	n.d.	70	0	5	15	n.s.
	day 2	122	n.d.	70	0	4	10	
Veillonella-, Neisseria-, Corynebacteria-group	day 1	122	0.5	1000	48	93	132	0.0016
	day 2	122	n.d.	8000	70	203	733	
Actinomyces-, Lactobacillus-, Staphylococcus-, Candida-group	day 1	122	n.d.	450	11	39	66	0.0001
	day 2	122	n.d.	2000	30	85	210	
Not identified	day 1	122	n.d.	110	6	16	24	n.s.
	day 2	122	n.d.	400	10	23	45	
Total cultivable flora	day 1	122	7	4096	610	783	709	0.0038
	day 2	122	8	34400	807	1470	3565	

n.d. indicates values less than  $10^6$

Table II. Colony forming unit values of microbial categories per ml saliva on blood agar plates ( $n \times 10^6$ ) on anaerobic cultivation at the baseline examination before the onset of the experiment in samples taken on two consecutive days, as well as the comparison between the two examinations (Wilcoxon-test)

Microbial categories	Sample	N	Min.	Max.	Med.	$\bar{x}$ arith	S.D.	$\alpha$
Streptococcus $\alpha$ -hemolyticus	day 1	122	4	8000	325	677	1049	0.0041
	day 2	121	10	20000	600	1055	2123	
Streptococcus non-hemolyticus	day 1	122	1	2000	120	259	380	0.0077
	day 2	121	n.d.	6000	200	403	732	
Streptococcus $\beta$ -hemolyticus	day 1	122	n.d.	300	0	7	29	n.s.
	day 2	121	n.d.	200	0	8	25	
Veillonella, Neisseria-, Corynebacteria-group	day 1	122	n.d.	1400	40	100	195	0.0000
	day 2	121	n.d.	13000	100	272	1195	
Actinomyces-, Lactobacillus-, Staphylococcus-, Candida-group	day 1	122	n.d.	370	20	41	59	0.0048
	day 2	121	n.d.	3000	30	81	276	
Not identified	day 1	122	n.d.	600	4	22	59	n.s.
	day 2	121	n.d.	400	10	29	52	
Total cultivable flora	day 1	122	9	10110	640	1106	1515	0.0009
	day 2	121	13	40400	1166	1847	3941	

n.d. indicates values less than  $10^6$

Table III. Colony forming unit values of microbial categories per ml saliva on blood agar plates ( $n \times 10^6$ ) on aerobic cultivation at the termination of the experiment in samples taken on two consecutive days, as well as the comparison between the two examinations (Wilcoxon-test)

Microbial categories	Sample	N	Min.	Max.	Med.	$\bar{x}$ arith.	S.D.	$\alpha$
Streptococcus $\alpha$ -hemolyticus	1	115	10	10000	200	715	1114	n.s.
	2	101	20	20000	200	691	2017	
Streptococcus non-hemolyticus	1	115	5	5000	100	400	708	n.s.
	2	101	5	10000	100	424	1084	
Streptococcus $\beta$ -hemolyticus	1	115	n.d.	300	0	12	38	n.s.
	2	101	n.d.	100	0	7	20	
Veillonella-, Neisseria-, Corynebacteria-group	1	115	1	1100	60	219	288	n.s.
	2	101	n.d.	2000	70	216	317	
Actinomyces-, Lactobacillus-, Staphylococcus-, Candida-group	1	115	n.d.	800	30	97	139	0.0422
	2	101	n.d.	800	30	79	117	
Not identified	1	115	n.d.	220	10	30	48	n.s.
	2	100	n.d.	600	12	50	99	
Total cultivable flora	1	115	53	16254	580	1473	2061	n.s.
	2	100	27	30984	484	1450	3273	

n.d. indicates values less than  $10^6$

Table IV. Colony forming unit values of microbial categories per ml saliva on blood agar plates ( $n \times 10^6$ ) on anaerobic cultivation at the termination of the experiment in samples taken on two consecutive days, as well as the comparison between the two examinations (Wilcoxon-test)

Microbial categories	Sample	N	Min.	Max.	Med.	$\bar{x}$ arith.	S.D.	$\alpha$
Streptococcus $\alpha$ -hemolyticus	day 1	114	10	20000	800	1474	2925	0.0176
	day 2	100	10	20000	400	1074	2800	
Streptococcus non-hemolyticus	day 1	114	5	20000	200	907	2704	n.s.
	day 2	100	5	10000	200	571	1250	
Streptococcus $\beta$ -hemolyticus	day 1	114	n.d.	300	0	17	44	n.s.
	day 2	100	n.d.	300	0	12	39	
Veillonella, Neisseria-, Corynebacteria-group	day 1	114	n.d.	3000	100	271	402	n.s.
	day 2	100	n.d.	2000	100	245	325	
Actinomyces-, Lactobacillus-, Staphylococcus-, Candida-group	day 1	114	n.d.	2000	50	151	235	n.s.
	day 2	100	1	1000	40	121	172	
Not identified	day 1	114	n.d.	600	20	45	91	n.s.
	day 2	100	n.d.	900	18	74	148	
Total cultivable flora	day 1	114	44	41380	1550	2865	5774	0.0480
	day 2	100	31	32020	985	2096	4263	

n.d. indicates values less than  $10^6$

Table V. Arithmetic and geometric means as well as standard deviations and 68 % intervals of the colony forming unit values of microbial categories on blood agar-plates per ml saliva ( $n \times 10^6$ ) as a function of sugar diet in aerobic incubation at the onset (year 1972) and at the termination (1974) of the experiment. The figures represent calculations of the geometric means of the samples taken on two consecutive days

Microbial categories	Examination year	N	Sucrose				Fructose				Xylitol					
			$\bar{x}_{arith.}$	S.D. <sub>arith.</sub>	Logarithmic scale $x_{geom.}$	68 % interval	$\bar{x}_{arith.}$	S.D. <sub>arith.</sub>	Logarithmic scale $x_{geom.}$	68 % interval	$\bar{x}_{arith.}$	S.D. <sub>arith.</sub>	Logarithmic scale $x_{geom.}$	68 % interval		
Streptococcus	1972	35	1030	2022	277	65—1186	38	433	257	296	132—662	49	534	388	372	159—872
$\alpha$ -hemolyticus	1974	27	753	587	399	113—1416	29	975	2737	259	61—1100	45	482	599	189	49—728
Streptococcus	1972	35	338	595	54	6—457	38	179	152	79	18—343	49	235	211	121	41—352
non-hemolyticus	1974	27	523	679	191	40—912	29	540	1386	124	25—610	45	298	400	92	20—421
Streptococcus	1972	35	4.6	11.4	0	0—0	38	5.7	13.2	0	0—0	49	3.3	8.0	0	0—0
$\beta$ -hemolyticus	1974	27	18.4	38.8	0	0—0	29	8.6	19.4	0	0—2	45	6.1	17.0	0	0—0
Veillonella,																
Neisseria-	1972	35	230	687	34	3—350	38	89.4	61.3	38	4—329	49	136	144	71	25—197
Corynebacteria-	1974	27	342	353	148	33—663	29	184	242	72	20—259	45	172	240	46	4—492
group																
Actinomyces,																
Lactobacillus,	1972	35	57.5	170	7	0—126	38	66.5	74.1	13	0—636	49	61.5	95.7	4	0—836
Staphylococcus,	1974	27	102	93.8	50	14—174	29	103	132	39	10—145	45	76.5	105	9	0—336
Candida-group																
Not identified	1972	35	21.4	37.3	0	0—157	38	14.4	19.7	0	0—109	49	22.3	25.3	0	0—465
	1974	27	64.4	97.6	1	0—796	29	41.9	56.8	3	0—365	44	25.9	36.5	5	0—146
Total cultivable	1972	35	1681	3314	490	124—1937	38	789	411	576	277—1196	49	992	651	701	308—1595
flora	1974	27	1804	1588	905	232—3531	29	1854	4329	578	145—2301	34	1004	1218	406	108—1518

Table VI. Arithmetic and geometric means as well as standard deviations and 68 % intervals of the colony forming unit values of microbial categories on blood agar plates per ml saliva ( $n \times 10^6$ ) as a function of sugar diet in anaerobic incubation at the onset (year 1972) and at the termination (1974) of the experiment. The figures represent calculations of the geometric means of the samples taken on two consecutive days

Microbial categories	Exam- nation year	SUCROSE					FRUCTOSE					XYLITOL				
		N	$\bar{x}_{arith.}$	S.D. <sub>arith.</sub>	Logarithmic scale 68 % interval	Arithmetic scale 68 % interval	N	$\bar{x}_{arith.}$	S.D. <sub>arith.</sub>	Logarithmic scale 68 % interval	Arithmetic scale 68 % interval	N	$\bar{x}_{arith.}$	S.D. <sub>arith.</sub>	Logarithmic scale 68 % interval	Arithmetic scale 68 % interval
Streptococcus $\alpha$ -hemolyticus	1972	35	1029	2021	329	83—1312	37	749	696	415	144—1199	49	844	890	494	194—1259
	1974	26	986	762	539	161—1806	29	1597	3272	456	109—1906	45	1129	2948	388	98—1542
Streptococcus non-hemolyticus	1972	35	282	406	98	23—417	37	399	588	129	32—530	49	317	289	131	18—973
	1974	26	708	870	262	56—1228	29	984	2278	190	42—865	45	664	1921	173	37—820
Streptococcus $\beta$ -hemolyticus	1972	35	635	17.5	0	0—0	37	9.0	19.3	0	0—2	49	7.2	25.1	0	0—0
	1974	26	23.5	53.0	0	0—1	29	14.1	31.6	0	0—3	45	9.5	26.0	0	0—0
Veillonella,																
Neisseria,	1972	35	343	1113	44	9—220	37	90.2	81.5	36	4—364	49	147	200	52	7—370
Corynebacteria- group	1974	26	402	466	167	41—688	29	203	242	92	29—297	45	204	245	56	2—1767
Actinomyces-, Lactobacillus-, Staphylococcus-, Candida-group	1972	35	80.6	255	8	0—149	37	62.3	61.0	13	0—679	49	46.5	49.6	3	0—847
	1974	26	164	160	78	25—244	29	139	170	62	20—195	45	99.6	109	26	2—325
Not identified	1972	35	30.8	62.8	0	0—205	37	19.4	29.7	0	0—128	49	25.6	29.6	1	0—269
	1974	26	88.3	138	5	0—720	29	37.4	44.5	8	1—138	45	53.6	91.3	7	0—328
Total cultivable flora	1972	35	1773	3728	573	147—2238	37	1328	1243	786	293—2107	49	1388	1223	905	371—2207
	1974	26	2371	2138	1230	339—4457	29	2975	5705	955	256—3353	45	2161	5026	826	220—3096

Table VII. Arithmetic and geometric means as well as standard deviations and 68% intervals of the colony forming unit values of microbial categories on MacLeod agar plates per ml saliva ( $n \times 10^4$ ) as a function of sugar diet in aerobic incubation at the onset (year 1972) and at the termination (1974) of the experiment. The figures represent calculations of the geometric means of the samples taken on two consecutive days

Microbial categories	Examination year	Sucrose				Fructose				Xylitol						
		N	$\bar{x}_{arith.}$	S.D. <sub>arith.</sub>	Logarithmic scale 68% interval	N	$\bar{x}_{arith.}$	S.D. <sub>arith.</sub>	Logarithmic scale 68% interval	N	$\bar{x}_{arith.}$	S.D. <sub>arith.</sub>	Logarithmic scale 68% interval			
Streptococcus hemolyticus	1972	35	767	1776	212	55—820	38	356	251	224	91—549	49	450	307	305	138—672
	1974	27	645	610	334	101—1097	29	538	1113	190	53—685	45	369	468	154	42—558
Streptococcus non-hemolyticus	1972	35	262	424	80	19—341	38	187	193	65	7—630	49	243	183	151	58—392
	1974	27	423	540	168	43—658	29	301	581	92	22—376	45	266	392	84	19—366
Veillonella,																
Neisseria,	1972	35	309	1107	38	7—203	38	92.9	108	21	1—404	49	116	134	42	7—257
Corynebacteria-group	1974	27	314	319	139	36—533	29	208	328	91	27—301	45	187	231	76	19—304
Actinomyces,																
Lactobacillus,	1972	35	70.3	252	5	0—136	38	34.5	37.0	4	0—414	49	34.6	45.4	1	0—571
Staphylococcus,	1974	27	63.9	93.5	3	0—256	29	34.5	55.2	0	0—379	45	35.1	56.8	2	0—320
Candida-group																
Not identified	1972	35	53.1	201	1	0—218	38	13.8	14.1	0	0—58	49	20.0	21.2	1	0—270
	1974	27	66.3	98.0	1	0—271	29	21.8	23.0	2	0—105	45	25.0	41.2	0	0—240
Total cultivable flora	1972	35	1461	3640	420	117—1510	38	685	471	464	207—1041	49	864	526	636	302—1336
	1974	27	1512	1433	776	224—2680	29	1104	2040	431	125—1492	45	883	1138	370	103—1327

Table VIII. Arithmetic and geometric means as well as standard deviations and 68 % intervals of the colony forming unit values of microbial categories on MacLeod agar plates per ml saliva ( $n \times 10^6$ ) as a function of sugar diet in anaerobic incubation at the onset (year 1972) and at the termination (1974) of the experiment. The figures represent calculations of the geometric means of the samples taken on two consecutive days

Microbial categories	Examination year	N	Sucrose				Fructose				Xylitol					
			Arithmetic scale $\bar{x}_{arith.}$	S.D. $S.D._{arith.}$	Logarithmic scale $\bar{x}_{geom.}$	68 % interval	Arithmetic scale $\bar{x}_{arith.}$	S.D. $S.D._{arith.}$	Logarithmic scale $\bar{x}_{geom.}$	68 % interval	Arithmetic scale $\bar{x}_{arith.}$	S.D. $S.D._{arith.}$	Logarithmic scale $\bar{x}_{geom.}$	68 % interval		
Streptococcus hemolyticus	1972	35	997	1722	339	87-1329	38	543	470	329	132-826	49	622	643	365	145-923
	1974	27	929	753	558	186-1672	29	799	1216	355	105-1196	45	852	1943	322	-1111
Streptococcus non-hemolyticus	1972	35	424	697	124	26-583	38	324	472	43	1-1812	49	317	322	172	62-475
	1974	27	670	803	278	68-1128	29	473	676	173	45-667	45	508	961	158	37-677
Veillonella,																
Neisseria,	1972	35	303	929	51	10-273	38	88.0	65.9	29	1-1003	49	134	188	58	16-209
Corynebacteria-group	1974	27	429	326	235	73-763	29	304	398	146	47-455	45	263	265	113	14-887
Actinomycetes,																
Lactobacillus,	1972	35	58.2	112	6	0-197	38	45.2	54.4	9	0-259	49	41.0	51.7	5	0-602
Staphylococcus-, Candida-group	1974	27	79.4	74.2	22	2-236	29	64.8	114	4	0-539	45	53.1	62.4	5	0-467
Not identified	1972	35	26.6	45.7	0	0-351	38	20.3	27.3	0	0-391	49	29.6	35.5	2	0-405
	1974	27	84.3	130	3	0-336	29	48.2	95.3	4	0-140	45	48.4	69.0	1	0-617
Total cultivable flora	1972	35	1809	3069	634	160-2511	28	1020	895	633	265-1510	49	1144	1076	750	321-1751
	1974	27	2141	1925	1171	354-3870	29	1689	2408	774	235-2546	45	1724	3128	736	225-2410

Table IX. Significance level of differences in the salivary colony forming unit values on aerobic incubation on blood agar plates (Wilcoxon-test)

Microbial categories	Blood agar			Mac Leod agar		
	S	F	X	S	F	X
Streptococcus $\alpha$ -hemolyticus	$\Phi$	$\Phi$	++	$\Phi$	$\Phi$	++
Streptococcus non-hemolyticus	+	$\Phi$	$\Phi$	+	$\Phi$	++
Streptococcus $\beta$ -hemolyticus	$\Phi$	$\Phi$	$\Phi$			
Veillonella-, Neisseria-, Corynebacteria-group	++	$\Phi$	$\Phi$	++	+	$\Phi$
Actinomyces-, Lactobacillus-, Staphylococcus-, Candida-group	+++	$\Phi$	$\Phi$	$\Phi$	$\Phi$	$\Phi$
Not identified	+	+	$\Phi$	$\Phi$	$\Phi$	$\Phi$
Total cultivable flora	+	$\Phi$	++	+	$\Phi$	++

$\Phi$  = not significant,  $p \geq 0.05$  +,  $p \geq 0.01$  ++,  $p \geq 0.005$  +++

Table X. Significance level of differences in the salivary colony forming unit values on anaerobic incubation on blood agar plates (Wilcoxon-test)

Microbial categories	Blood agar			Mac Leod agar		
	S	F	X	S	F	X
Streptococcus $\alpha$ -hemolyticus	$\Phi$	$\Phi$	$\Phi$	$\Phi$	$\Phi$	$\Phi$
Streptococcus non-hemolyticus	++	$\Phi$	$\Phi$	+	$\Phi$	$\Phi$
Streptococcus $\beta$ -hemolyticus	$\Phi$	$\Phi$	$\Phi$			
Veillonella-, Neisseria-, Corynebacteria-group	+++	$\Phi$	$\Phi$	+++	++	++
Actinomyces-, Lactobacillus-, Staphylococcus-, Candida-group	+++	+	++	+	$\Phi$	$\Phi$
Not identified	++	++	++	++	$\Phi$	$\Phi$
Total cultivable flora	+	$\Phi$	$\Phi$	++	$\Phi$	$\Phi$

$\Phi$  = not significant,  $p \geq 0.05$  +,  $p \geq 0.01$  ++,  $p \geq 0.005$  +++

Table XI. Significance level of differences in the salivary colony forming unit values on blood agar between the sugar groups at the onset (1972) and termination (1974) of the diet period (Kruskal-Wallis and Mann-Whitney-U-tests)

Microbial categories	1972				1974			
	Kruskal-Wallis	S/F	U-test S/X	F/X	Kruskal-Wallis	S/F	U-test S/X	F/X
Streptococcus $\alpha$ -hemolytica	n.s.	n.s.	n.s.	n.s.	0.1027	n.s.	0.0429	n.s.
Streptococcus non-hemolyticus	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
Streptococcus $\beta$ -hemolyticus	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
Veillonella-, Neisseria-, Corynebacteria-group	n.s.	n.s.	n.s.	n.s.	0.0393	0.0687	0.0163	n.s.
Actinomyces-, Lactobacillus-, Staphylococcus-, Candida-group	0.0647	0.0161	n.s.	n.s.	0.0487	n.s.	0.0300	0.0773
Not identified	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
Total cultivable flora	n.s.	n.s.	n.s.	n.s.	0.0857	n.s.	0.0366	n.s.

Table XII. The colony forming unit values of salivary *Streptococcus  $\alpha$ -hemolyticus*, *Streptococcus non-hemolyticus*, *Streptococcus  $\beta$ -hemolyticus* and total streptococcal counts expressed as percentage of total viable counts on aerobic and anaerobic blood agar plates

Microorganism	Sugar group	Aerobic		Anaerobic	
		1972	1974	1972	1974
<i>Streptococcus <math>\alpha</math>-hemolyticus</i>	S	61.3	41.0	58.0	41.6
	F	55.0	52.6	56.4	53.8
	X	53.7	47.2	60.8	52.2
<i>Streptococcus non-hemolyticus</i>	S	20.0	28.4	15.9	29.9
	F	22.7	29.1	30.0	33.0
	X	23.7	29.2	22.8	30.7
<i>Streptococcus <math>\beta</math>-hemolyticus</i>	S	0.27	1.02	0.37	0.99
	F	0.72	0.46	0.68	0.57
	X	0.33	0.60	0.52	0.44
Total streptococci	S	81.7	71.8	74.3	72.4
	F	78.3	82.2	87.1	87.2
	X	77.9	78.3	84.1	83.4

Table XIII. Colony forming unit values of microbial categories per mg plaque on blood agar plates ( $n \times 10^6$ ) on aerobic cultivation at the baseline examination before the onset of the experiment in samples taken on two consecutive days, as well as the comparison between the two examinations (Wilcoxon-test)

Microbial categories	Sample	N	Min.	Max.	Med.	$\bar{x}$ arith	S.D.	$\alpha$
Streptococcus $\alpha$ -hemolyticus	day 1	122	n.d.	1430	17	155	1293	n.s.
	day 2	122	n.d.	620	21	51	87	
Streptococcus non-hemolyticus	day 1	122	n.d.	120	4	11	21	n.s.
	day 2	122	n.d.	230	3	11	26	
Streptococcus $\beta$ -hemolyticus	day 1	122	n.d.	60	n.d.	1	6	n.s.
	day 2	122	n.d.	7	n.d.	0.3	1	
Veillonella-, Neisseria-, Coryne- bacteria-group	day 1	122	n.d.	60	2	6	11	n.s.
	day 2	122	n.d.	90	3	6	11	
Actinomyces-, Lactobacillus-, Staphylococcus-, Candida-group	day 1	122	n.d.	130	1	6	18	n.s.
	day 2	122	n.d.	270	1	9	28	
Not identified	day 1	122	n.d.	10	n.d.	0.5	1	n.s.
	day 2	122	n.d.	30	n.d.	1	3	
Total cultivable flora	day 1	122	n.d.	1437	30	180	1300	n.s.
	day 2	122	n.d.	866	36	77	124	

n.d. indicates values less than  $10^6$

Table XIV. Colony forming unit values of microbial categories per mg plaque on blood agar plates ( $n \times 10^6$ ) on anaerobic cultivation at the baseline examination before the onset of the experiment in samples taken on two consecutive days, as well as the comparison between the two examinations (Wilcoxon-test)

Microbial categories	Sample	N	Min.	Max.	Med.	$\bar{x}$ arith	S.D.	$\alpha$
Streptococcus $\alpha$ -hemolyticus	day 1	122	n.d.	1140	24	86	157	n.s.
	day 2	122	n.d.	1430	38	93	168	
Streptococcus non-hemolyticus	day 1	122	n.d.	400	5	17	45	n.s.
	day 2	121	n.d.	860	6	27	85	
Streptococcus $\beta$ -hemolyticus	day 1	122	n.d.	70	0	1	6	n.s.
	day 2	121	n.d.	9	0	0.5	1	
Veillonella, Neisseria-, Coryne- bacteria-group	day 1	122	n.d.	500	3	25	68	n.s.
	day 2	121	n.d.	430	5	22	57	
Actinomyces-, Lactobacillus-, Staphylococcus-, Candida-group	day 1	122	n.d.	240	1	9	26	n.s.
	day 2	121	n.d.	330	1	14	41	
Not identified	day 1	122	n.d.	30	0	2	10	n.s.
	day 2	121	n.d.	90	0	2	9	
Total cultivable flora	day 1	122	n.d.	1467	45	141	249	n.s.
	day 2	121	n.d.	2800	57	158	307	

n.d. indicates values less than  $10^6$

Table XV. Colony forming unit values of microbial categories per mg plaque on blood agar plates ( $n \times 10^6$ ) on aerobic cultivation at the termination of the experiment in samples taken on two consecutive days, as well as the comparison between the two examinations (Wilcoxon-test)

Microbial categories	Sample	N	Min.	Max.	Med.	$\bar{x}$ arith.	S.D.	$\alpha$
Streptococcus $\alpha$ -hemolyticus	day 1	113	n.d.	570	12	52	100	0.0024
	day 2	101	n.d.	1110	20	65	136	
Streptococcus non-hemolyticus	day 1	113	n.d.	200	4	19	38	0.0021
	day 2	101	n.d.	250	7	27	51	
Streptococcus $\beta$ -hemolyticus	day 1	113	n.d.	40	0	1	5	n.s.
	day 2	101	n.d.	120	0	2	12	
Veillonella-, Neisseria-, Corynebacteria-group	day 1	113	n.d.	140	1	7	18	0.0025
	day 2	101	n.d.	220	4	13	28	
Actinomyces-, Lactobacillus-, Staphylococcus-, Candida-group	day 1	113	n.d.	190	3	9	22	0.0002
	day 2	101	n.d.	440	5	19	55	
Not identified	day 1	113	n.d.	30	0	1	4	0.0001
	day 2	101	n.d.	50	0	3	7	
Total cultivable flora	day 1	113	n.d.	1006	24	90	170	0.0001
	day 2	101	n.d.	2001	35	130	259	

n.d. indicates values less than  $10^6$

Table XVI. Colony forming unit values of microbial categories per mg plaque on blood agar plates ( $n \times 10^6$ ) on anaerobic cultivation at the termination of the experiment in samples taken on two consecutive days, as well as the comparison between the two examinations (Wilcoxon-test)

Microbial categories	Sample	N	Min.	Max.	Med.	$\bar{x}$ arith.	S.D.	$\alpha$
Streptococcus $\alpha$ -hemolyticus	day 1	115	1	1670	31	114	254	0.0000
	day 2	100	n.d.	3330	50	183	449	
Streptococcus non-hemolyticus	day 1	115	n.d.	380	9	34	66	0.0014
	day 2	100	n.d.	3330	19	80	344	
Streptococcus $\beta$ -hemolyticus	day 1	115	n.d.	80	n.d.	2	9	0.0146
	day 2	100	n.d.	60	n.d.	2	7	
Veillonella, Neisseria-, Corynebacteria-group	day 1	115	n.d.	270	2	14	34	0.0002
	day 2	100	n.d.	440	8	24	55	
Actinomyces-, Lactobacillus-, Staphylococcus-, Candida-group	day 1	115	n.d.	500	3	18	53	0.0001
	day 2	100	n.d.	440	10	29	56	
Not identified	day 1	115	n.d.	30	n.d.	2	4	0.0001
	day 2	100	n.d.	110	1	6	16	
Total cultivable flora	day 1	115	1	1790	56	184	332	0.0000
	day 2	100	1	4700	109	324	732	

n.d. indicates values less than  $10^6$

Table XVII. Arithmetic and geometric means as well as standard deviations and 68 % intervals of the colony forming unit values of microbial categories on blood agar plates per mg plaque ( $n \times 10^6$ ) as a function of sugar diet in aerobic incubation at the onset (year 1972) and at the termination (1974) of the experiment. The figures represent calculations of the arithmetic means of the samples taken on two consecutive days

Microbial categories	Examination year	Sucrose				Fructose				Xylitol						
		N	$\bar{x}_{arith.}$	S.D.	Logarithmic scale 68 % interval	N	$\bar{x}_{arith.}$	S.D.	Logarithmic scale 68 % interval	N	$\bar{x}_{arith.}$	S.D.	Logarithmic scale 68 % interval			
Streptococcus $\alpha$ -hemolyticus	1972	35	26.3	23.2	13	5-33	38	61.2	83.0	26	7-106	49	45.5	74.4	16	5-56
	1974	27	78.3	155	24	6-98	29	71.1	115.8	26	8-91	45	32.6	55.2	11	3-41
Streptococcus non-hemolyticus	1972	35	5.0	6.0	0	0-32	38	16.4	26.2	1	0-83	49	11.2	16.9	1	0-59
	1974	27	31.7	51.9	5	0-92	29	30.2	41.5	10	3-39	45	14.5	33.1	4	1-26
Streptococcus $\beta$ -hemolyticus	1972	35	0.4	1.0	0	0-0	38	0.5	1.0	0	0-0	49	0.7	4.3	0	0-0
	1974	27	3.5	14.5	0	0-0	29	2.1	6.7	0	0-0	45	0.3	0.6	0	0-0
Veillonella-																
Neisseria-, Corynebacteria- group	1972	35	5.0	6.5	2	0-9	38	5.5	6.7	0	0-23	49	7.0	10.2	1	0-24
	1974	27	15.9	30.3	1	0-43	29	12.4	23.8	2	0-29	45	5.4	7.9	0	0-40
Actinomyces-, Lactobacillus-, Staphylococcus-, Candida-group	1972	35	5.3	11.7	0	0-32	38	9.1	15.4	0	0-106	49	7.8	27.3	0	0-23
	1974	27	22.4	49.2	5	0-48	29	19.4	44.8	4	0-36	45	6.1	10.7	1	0-29
Not identified	1972	35	0.5	0.8	0	0-0	38	0.6	1.4	0	0-0	49	0.8	2.4	0	0-0
	1974	27	1.6	2.3	0	0-5	29	3.5	8.7	0	0-6	45	1.3	2.4	0	0-1
Total cultivable flora	1972	35	42.5	40.0	23	9-58	38	93.3	85.0	43	12-158	49	73.0	108	29	8-98
	1974	27	153	291	48	11-199	29	139	225	53	15-185	45	60.1	103	22	6-78

Table XVIII. Arithmetic and geometric means as well as standard deviations and 68 % intervals of the colony forming unit values of microbial categories on blood agar plates per mg plaque ( $n \times 10^6$ ) as a function of sugar diet in anaerobic incubation at the onset (year 1972) and at the termination (1974) of the experiment. The figures represent calculation of the arithmetic means of the samples taken on two consecutive days

Microbial categories	Exam-ination year	Sucrose				Fructose				Xylitol						
		N	$\bar{x}_{arith.}$	S.D. <sub>arith.</sub>	Logarithmic scale 68 % interval	N	$\bar{x}_{arith.}$	S.D. <sub>arith.</sub>	Logarithmic scale 68 % interval	N	$\bar{x}_{arith.}$	S.D. <sub>arith.</sub>	Logarithmic scale 68 % interval			
Streptococcus $\alpha$ -hemolyticus	1972	35	82.7	118	28	7-106	37	103	211	36	11-123	49	83.7	111	26	6-115
	1974	27	204	376	53	14-206	28	180	420	49	11-209	45	68.0	79.2	29	8-106
Streptococcus non-hemolyticus	1972	35	11.4	14.2	2	0-61	37	31.9	87.3	2	0-82	49	22.9	38.6	2	0-86
	1974	27	118	332	12	1-135	28	43.9	65.3	16	4-56	45	26.1	39.7	8	1-59
Streptococcus $\beta$ -hemolyticus	1972	35	0.5	1.2	0	0-0	37	1.5	6.5	0	0-0	49	0.4	0.8	0	0-0
	1974	27	3.4	13.4	0	0-0	28	3.0	7.7	0	0-0	45	0.5	1.3	0	0-0
Veillonella-																
Neisseria-, Corynebacteria- group	1972	35	22.6	51.8	1	0-198	37	22.1	59.3	0	0-50	49	25.3	42.9	1	0-60
	1974	27	31.2	53.6	3	0-60	28	19.5	34.1	2	0-60	45	11.2	12.8	2	0-81
Actinomyces-, Lactobacillus-, Staphylococcus-, Candida-group	1972	35	10.3	21.0	0	0-87	37	10.9	16.5	0	0-187	49	12.5	40.2	0	0-33
	1974	27	30.9	50.1	2	0-134	28	26.1	41.0	6	1-61	45	12.0	22.6	2	0-24
Not identified	1972	35	2.1	3.7	0	0-1	27	3.6	12.9	0	0-0	49	1.8	3.5	0	0-0
	1974	27	7.6	15.1	0	0-10	28	3.6	7.1	0	0-5	45	2.5	4.4	0	0-2
Total cultivable flora	1972	35	130	165	49	14-175	37	174	363	63	19-212	49	147	202	48	12-200
	1974	27	395	680	102	26-407	28	277	522	93	23-376	45	120	143	54	15-187

Table XIX. Arithmetic and geometric means as well as standard deviations and 68 % intervals of the colony forming unit values of microbial categories on MacLeod agar plates per mg plaque ( $n \times 10^6$ ) as a function of sugar diet in aerobic incubation at the onset (year 1972) and at the termination (1974) of the experiment. The figures represent calculations of the arithmetic means of the samples taken on two consecutive days

Microbial categories	Exam-ination year	N	Sucrose				Fructose				Xylitol													
			$\bar{x}_{arith.}$	S.D. <sub>arith.</sub>	$\bar{x}_{geom.}$	68 % interval	Arithmetic scale	Logarithmic scale	68 % interval	N	$\bar{x}_{arith.}$	S.D. <sub>arith.</sub>	$\bar{x}_{geom.}$	68 % interval	Arithmetic scale	Logarithmic scale	68 % interval	N	$\bar{x}_{arith.}$	S.D. <sub>arith.</sub>	$\bar{x}_{geom.}$	68 % interval		
Streptococcus hemolyticus	1972	35	24.9	35.7	12	4-37	38	63.0	84.0	29	6-129	49	44.6	72.1	16	4-59								
	1974	27	95.2	213	22	5-92	29	95.2	213	23	5-93	45	37.3	81.5	10	3-37								
Streptococcus non-hemolyticus	1972	35	4.9	5.0	2	0-22	38	22.1	33.9	2	0-22	49	12.2	20.1	2	0-108								
	1974	27	24.1	37.9	6	1-25	29	77.3	134	3	0-147	45	15.1	43.8	3	0-27								
Veillonella,																								
Neisseria,	1972	35	9.5	24.5	3	0-10	38	9.7	12.2	2	0-30	49	9.1	13.5	2	0-28								
	1974	27	28.8	69.0	2	0-50	29	31.7	72.1	3	0-41	45	8.8	10.9	1	0-54								
Corynebacteria-																								
group																								
Actinomyces,																								
Lactobacillus,	1972	35	1.6	3.0	0	0-2	38	10.4	28.5	0	0-66	49	3.9	11.8	0	0-6								
Staphylococcus,	1974	27	7.3	21.8	0	0-10	29	4.2	6.2	0	0-6	45	1.1	1.3	0	0-3								
Candida-group																								
Not identified	1972	35	0.9	1.6	0	0-0	38	3.3	7.8	0	0-2	49	1.7	4.5	0	0-1								
	1974	27	3.7	9.2	0	0-0	29	1.9	3.5	0	0-1	45	0.9	2.1	0	0-1								
Total cultivable flora	1972	35	41.8	61.8	21	7-64	38	108.5	166.4	47	11-198	49	71.5	102	28	8-103								
	1974	27	159	316	37	8-161	29	139	242	41	10-166	45	63.2	129	18	5-70								



Table XXI. *Significance level of differences in the colony forming unit values on aerobic incubation of dental plaque on blood agar plates (Wilcoxon-test)*

Microbial categories	Blood agar			Mac Leod agar		
	S	F	X	S	F	X
Streptococcus $\alpha$ -hemolyticus	+	$\emptyset$	$\emptyset$	$\emptyset$	$\emptyset$	+
Streptococcus non-hemolyticus	++	+	$\emptyset$	+	$\emptyset$	$\emptyset$
Streptococcus $\beta$ -hemolyticus	$\emptyset$	$\emptyset$	$\emptyset$	$\emptyset$	$\emptyset$	$\emptyset$
Veillonella-, Neisseria-, Corynebacteria-group	$\emptyset$	$\emptyset$	$\emptyset$	$\emptyset$	$\emptyset$	$\emptyset$
Actinomyces-, Lactobacillus-, Staphylococcus-Candida-group	+++	+	++	+	$\emptyset$	$\emptyset$
Not identified	+	++	+	$\emptyset$	$\emptyset$	$\emptyset$
Total cultivable flora	+	$\emptyset$	$\emptyset$	$\emptyset$	$\emptyset$	$\emptyset$

Table XXII. *Significance level of differences in the colony forming unit values on anaerobic incubation of dental plaques on blood agar plates (Wilcoxon-test)*

Microbial categories	Blood agar			Mac Leod agar		
	S	F	X	S	F	X
Streptococcus $\alpha$ -hemolyticus	+++	+++	+++	$\emptyset$	$\emptyset$	$\emptyset$
Streptococcus non-hemolyticus	++	+++	+++	$\emptyset$	$\emptyset$	$\emptyset$
Streptococcus $\beta$ -hemolyticus	+++	+++	+++	$\emptyset$	$\emptyset$	$\emptyset$
Veillonella-, Neisseria-, Corynebacteria-group	+++	+++	+++	$\emptyset$	$\emptyset$	$\emptyset$
Actinomyces-, Lactobacillus-, Staphylococcus-, Candida-group	+	+++	+++	+	$\emptyset$	$\emptyset$
Not identified	++	+++	+++	$\emptyset$	$\emptyset$	$\emptyset$
Total cultivable flora	+++	+++	+++	$\emptyset$	$\emptyset$	$\emptyset$

Table XXIII. Significance level of differences in the colony forming unit values of dental plaque between the sugar groups at the onset (1972) and termination (1974) of the diet period on blood agar (Kruskal-Wallis and Mann-Whitney U-tests)

Microbial categories	1972				1974			
	Kruskal-Wallis	S/F	U-test S/X	F/X	Kruskal-Wallis	S/F	U-test S/X	F/X
Streptococcus $\alpha$ -hemolyticus	n.s.	n.s.	n.s.	n.s.	0.0090	n.s.	0.0251	0.0053
Streptococcus non-hemolyticus	n.s.	n.s.	n.s.	n.s.	0.0340	n.s.	n.s.	0.0107
Streptococcus $\beta$ -hemolyticus	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
Veillonella-, Neisseria-, Corynebacteria-group	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
Actinomyces-, Lactobacillus-, Staphylococcus-, Candida-group	n.s.	n.s.	n.s.	n.s.	0.0004	n.s.	0.0007	0.0014
Not identified	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
Total cultivable flora	0.114	0.0389	n.s.	n.s.	0.0096	n.s.	0.0219	0.0065

Table XXIV. The colony forming unit values of *Streptococcus  $\alpha$ -hemolyticus*, *Streptococcus non-hemolyticus*, *Streptococcus  $\beta$ -hemolyticus* and total streptococci in dental plaque expressed as percentage of total viable counts on aerobic and anaerobic blood agar plates

Microorganism	Sugar groups	Aerobic		Anaerobic	
		1972	1974	1972	1974
Streptococcus $\alpha$ -hemolyticus	S	62.8	51.2	63.6	51.7
	F	66.0	51.2	59.2	65.0
	X	62.4	55.3	56.9	56.6
Streptococcus non-hemolyticus	S	11.8	20.7	8.8	29.9
	F	17.58	23.4	18.3	15.8
	X	15.3	20.4	15.6	21.8
Streptococcus $\beta$ -hemolyticus	S	0.94	2.29	0.38	0.86
	F	0.54	1.51	0.86	1.08
	X	0.96	0.50	0.27	0.42
Total streptococci	S	74.6	74.1	72.8	82.38
	F	83.7	74.4	78.4	81.9
	X	78.6	78.9	72.8	78.8

Table XXV. Frequency of occurrence and total colony forming unit values on MacConcey agar per ml saliva ( $n \times 10^3$ ) as a function of time and sugar diet

	N	N—	N+	Logarithmic scale				Arithmetic scale				
				$\bar{x}_{\log}$	S.D. <sub>log</sub>	Min.	Max.	Med.	$\bar{x}_{\text{geom.}}$	68 % interval	$\bar{x}_{\text{arith.}}$	S.D. <sub>arith.</sub>
<b>Sucrose</b>												
— 1 day	33	0	33	11.89	2.04	6.5	14.5	12.6	146.4	19.0—1131.6	492.3	557.7
0 »	32	0	32	11.88	2.09	6.0	15.1	12.0	144.6	17.9—1164.9	493.2	741.3
4 month	33	0	33	11.03	1.58	7.6	13.1	11.4	62.0	12.7—301.3	136.4	130.4
12 » (1)	33	1	32	11.58	2.60	—0.98	13.8	12.2	107.4	8.0—1441.7	257.4	217.7
12 » (2)	33	4	29	9.76	4.48	—0.98	13.3	11.3	17.4	0.2—1535.9	187.8	206.5
19 »	33	2	31	9.70	3.35	—0.98	13.6	10.3	16.3	0.6—463.6	125.9	214.0
24 » (1)	32	1	31	11.89	2.78	—0.98	14.7	12.5	145.8	9.0—2354.3	492.9	625.2
24 » (2)	26	0	26	11.72	1.64	8.8	14.9	11.8	123.9	24.0—639.4	384.3	647.1
<b>Fructose</b>												
— 1 day	35	0	35	11.68	2.03	6.9	14.7	12.1	118.5	15.5—902.3	487.6	709.5
0 »	35	0	35	11.55	1.61	8.5	14.9	11.5	104.1	20.8—521.5	338.1	692.5
4 month	35	0	35	11.42	1.10	9.6	13.1	11.3	91.6	30.4—276.4	156.3	150.3
12 » (1)	35	0	35	11.98	1.41	8.0	13.1	12.4	160.2	39.0—658.4	276.9	193.7
12 » (1)	35	1	34	10.86	2.40	—0.98	13.3	11.2	52.2	4.7—578.4	140.0	165.5
19 »	35	3	32	9.87	3.67	—0.98	14.0	10.6	19.3	0.5—759.7	129.0	227.4
24 » (1)	33	1	32	11.73	2.74	—0.98	14.7	11.9	124.2	8.1—1914.6	427.6	589.3
24 » (2)	26	1	25	11.42	2.94	—0.98	14.5	12.0	90.7	4.8—1720.1	369.4	544.3
<b>Xylitol</b>												
— 1 day	47	0	47	11.84	1.74	8.0	14.7	12.2	138.7	24.4—789.5	440.7	645.4
0 »	47	0	47	12.06	1.66	5.7	14.5	12.3	174.0	32.9—919.6	414.4	478.7
4 month	47	0	47	11.53	1.17	8.7	13.1	11.5	102.2	31.6—329.8	169.2	138.0
12 » (1)	47	1	46	11.45	2.51	—0.98	13.8	12.2	94.3	7.6—1167.9	261.7	231.0
12 » (2)	47	5	42	9.90	4.10	—0.98	13.6	11.0	20.0	0.3—1208.6	165.6	208.9
19 »	47	5	42	9.37	3.84	—0.98	13.3	10.3	11.7	0.3—546.3	77.4	113.7
24 » (1)	47	3	44	11.27	3.61	—0.98	14.5	12.4	78.7	2.1—2922.8	404.4	466.2
24 » (2)	44	1	43	11.36	2.55	—0.98	14.2	11.9	86.5	6.8—1106.0	287.6	324.6

Table XXVI. Frequency of occurrence and total colony forming unit values of *E. coli* per ml saliva ( $n \times 10^3$ ) on Mac Concey agar as a function of time and sugar diet

	N	N—	N+	Logarithmic scale					Arithmetic scale				
				$\bar{x}_{\log}$	S.D. <sub>log</sub>	Min.	Max.	Med.	$\bar{x}_{\text{geom.}}$	68 % interval	$\bar{x}_{\text{arith.}}$	S.D. <sub>arith.</sub>	
<b>Sucrose</b>													
— 1 day	33	0	33	11.34	2.39	5.7	14.5	12.2	84.1	7.6	— 921.7	389.4	499.4
0 »	33	0	33	10.94	2.74	4.6	14.9	11.5	56.6	3.6	— 381.0	377.8	649.8
4 month	33	3	30	9.74	3.76	—0.98	12.9	10.8	17.1	0.4	— 730.2	100.8	107.1
12 » (1)	33	1	32	11.35	2.56	—0.98	13.7	12.2	85.0	6.6	—1095.6	208.7	193.2
12 » (2)	33	7	26	8.67	5.32	—0.98	13.1	10.8	5.9	0.03	—1201.6	147.6	173.4
19 »	33	10	23	6.57	5.30	—0.98	12.9	8.5	0.7	0.0	— 143.1	45.3	83.6
24 » (1)	32	1	31	11.51	2.80	—0.98	14.5	11.8	99.8	6.1	—1636.6	390.0	537.9
24 » (2)	26	0	26	11.20	1.87	6.9	14.8	10.9	73.4	11.3	— 476.6	320.4	617.7
<b>Fructose</b>													
— 1 day	35	2	33	10.02	3.90	—0.98	14.7	10.6	22.5	0.5	—1116.8	405.0	700.1
0 »	35	1	34	10.22	3.07	—0.98	14.9	10.6	27.5	1.3	— 593.9	262.4	638.3
4 month	35	1	34	10.94	2.42	—0.98	12.9	11.2	56.6	5.0	— 633.8	141.3	132.1
12 » (1)	35	2	33	11.19	3.37	—0.98	13.1	12.2	72.2	2.5	—2104.3	242.7	179.1
12 » (2)	35	4	31	9.43	4.06	—0.98	13.1	10.6	12.5	0.2	— 725.3	107.9	159.7
19 »	35	9	26	7.30	5.15	—0.98	13.8	9.2	1.5	0.0	— 254.1	67.3	173.0
24 » (1)	33	1	32	11.16	2.90	—0.98	14.5	11.2	70.1	3.8	—1280.8	323.7	457.1
24 » (2)	26	1	25	10.99	3.04	—0.98	14.5	11.8	59.1	2.8	—1237.6	288.5	440.1
<b>Xylitol</b>													
— 1 day	47	1	46	11.10	2.66	—0.98	14.5	11.5	65.9	4.6	— 942.3	332.5	513.4
0 »	47	1	46	10.94	3.01	—0.98	14.4	11.3	56.4	2.8	—1141.8	324.4	464.6
4 month	47	0	47	11.20	1.38	8.0	12.8	11.6	73.0	18.3	— 290.7	132.6	106.1
12 » (1)	47	2	45	10.94	3.13	—0.98	13.7	12.2	56.3	2.5	—1292.4	225.0	215.6
12 » (2)	47	10	37	8.62	5.23	—0.98	13.6	10.8	5.6	0.03	—1036.0	132.7	178.4
19 »	47	11	36	7.42	4.86	—0.98	13.1	9.2	1.7	0.01	— 215.5	43.4	85.8
24 » (1)	47	3	44	10.93	3.62	—0.98	14.5	12.0	55.6	1.5	—2079.3	345.4	448.9
24 » (2)	44	1	43	10.85	2.55	—0.98	14.2	11.4	51.3	4.0	— 656.9	201.6	289.9

Table XXVII. Frequency of occurrence and total colony forming unit values of neisseria per ml saliva ( $n \times 10^6$ ) on Mac Concey agar as a function of time and sugar diet

	N	N—	N+	Logarithmic scale				Arithmetic scale				
				$\bar{x}_{\log}$	S.D. <sub>log</sub>	Min.	Max.	Med.	$\bar{x}_{\text{geom.}}$	68 % interval	$\bar{x}_{\text{arith.}}$	S.D. <sub>arith.</sub>
<b>Sucrose</b>												
— 1 day	33	10	23	6.83	5.44	—0.98	13.3	3.2	0.923	0.004—212.8	56.2	114.9
0 »	33	6	27	8.50	4.89	—0.98	13.1	10.8	4.900	0.037—654.1	92.0	127.0
4 month	32	10	22	6.68	5.33	—0.98	12.2	9.3	0.792	0.004—162.9	32.1	51.1
12 » (1)	33	11	22	6.55	5.53	—0.98	11.8	9.9	0.699	0.003—176.7	35.8	42.8
12 » (2)	33	9	24	7.00	5.12	—0.98	11.5	9.2	1.092	0.007—183.0	30.1	35.4
19 »	33	5	28	8.26	4.33	—0.98	13.1	9.2	3.876	0.051—292.9	69.8	125.4
24 » (1)	30	8	22	7.79	5.55	—0.98	13.1	11.0	2.415	0.009—621.8	89.6	120.4
24 » (2)	24	6	18	7.45	5.10	—0.98	12.4	9.9	1.719	0.010—281.7	41.6	58.3
<b>Fructose</b>												
— 1 day	35	11	24	6.53	5.38	—0.98	13.8	9.2	0.685	0.003—148.5	58.2	164.8
0 »	35	11	24	6.89	5.55	—0.98	13.3	9.2	0.987	0.004—254.8	68.1	135.3
4 month	35	6	29	7.99	4.29	—0.98	11.5	9.2	2.996	0.040—215.3	28.2	33.9
12 » (1)	35	14	21	5.79	5.67	—0.98	11.5	8.9	0.326	0.001— 95.0	27.3	34.2
12 » (2)	35	11	24	6.44	5.20	—0.98	12.1	9.2	0.625	0.003—113.9	23.8	37.0
19 »	35	6	29	8.40	4.50	—0.98	12.9	9.9	4.450	0.049—401.6	58.6	94.4
24 » (1)	32	7	25	8.24	5.16	—0.98	13.8	10.8	3.778	0.021—656.5	100.9	192.9
24 » (2)	25	9	16	6.23	5.71	—0.98	13.8	8.5	0.507	0.001—152.7	76.8	207.2
<b>Xylitol</b>												
— 1 day	46	10	36	8.15	5.10	—0.98	13.8	9.9	3.460	0.021—567.3	110.1	195.6
0 »	46	8	38	8.88	4.71	—0.98	13.6	10.6	7.218	0.065—800.6	98.3	162.6
4 month	46	9	37	7.98	4.61	—0.98	11.5	9.9	2.944	0.029—295.5	35.9	37.5
12 » (1)	47	20	27	5.33	5.59	—0.98	12.2	8.0	0.205	0.001— 55.2	26.1	42.7
12 » (2)	47	13	34	6.69	4.95	—0.98	12.2	9.0	0.804	0.006—113.7	24.6	39.5
19 »	47	10	37	7.60	4.67	—0.98	12.0	9.3	1.999	0.019—212.4	30.3	36.9
24 » (1)	42	23	19	4.43	6.09	—0.98	13.3	— 0.98	0.984	0.000— 37.2	51.0	109.7
24 » (2)	40	16	24	6.25	6.06	—0.98	13.3	9.5	0.518	0.001—222.0	78.2	136.0

Table XXVIII. Significance level of differences in the salivary colony forming unit values on Mac Concey agar within the sugar groups during the diet period (Friedman test)

E. Coli	S	<0.0001
	F	0.0003
	X	<0.0001
Neisseria	S	n.s.
	F	n.s.
	X	<0.0001
Total	S	<0.0001
	F	0.0084
	X	<0.0001

Table XXIX. Frequency of occurrence and total colony forming unit values on Rogosa S. L. agar per ml saliva ( $n \times 10^6$ ) as a function of time and sugar diet

	N	N—	N+	Logarithmic scale					Arithmetic scale				
				$\bar{x}_{\log}$	S.D. <sub>log</sub>	Min.	Max.	Med.	$\bar{x}_{\text{geom.}}$	68 % interval	$\bar{x}_{\text{arith.}}$	S.D. <sub>arith.</sub>	
<b>Sucrose</b>													
— 1 day	32	6	26	6.78	4.16	—0.98	11.6	8.0	0.884	0.013—56.84	14.54	24.55	
0 »	32	6	26	6.86	4.17	—0.98	11.3	8.3	0.957	0.014—62.07	14.26	20.85	
1 month	32	6	26	7.12	4.17	—0.98	11.4	9.0	1.232	0.19—79.66	13.36	18.21	
4 »	32	10	22	5.67	4.82	—0.98	11.5	7.3	0.289	0.002—35.93	14.48	27.54	
8 »	32	4	28	7.57	3.87	—0.98	12.2	8.8	1.945	0.040—93.39	25.11	43.01	
12 » (1)	33	4	29	7.67	3.95	—0.98	13.8	8.3	2.355	0.045—122.07	56.30	175.55	
12 » (2)	33	3	30	7.75	3.85	—0.98	14.5	7.7	2.318	0.049—108.94	93.90	348.65	
19 »	33	6	27	7.22	4.56	—0.98	14.5	7.8	1.362	0.015—126.39	35.13	349.55	
24 » (1)	32	7	25	7.24	5.02	—0.98	14.4	9.0	1.399	0.009—213.06	107.76	321.66	
24 » (2)	26	1	25			—0.98			8.636	0.327—228.08	131.88	309.08	
<b>Fructose</b>													
— 1 day	34	1	33	8.85	2.42	—0.98	12.1	9.2	7.009	0.623—78.86	28.91	45.85	
0 »	34	1	33	8.83	2.56	—0.98	12.4	9.0	6.867	0.532—88.57	35.67	61.83	
1 month	33	4	29	7.70	3.58	—0.98	11.5	9.0	2.212	0.061—79.62	16.97	26.44	
4 »	34	1	33	8.30	2.46	—0.98	11.8	8.4	4.022	0.345—46.85	21.43	36.85	
8 »	34	1	33	9.00	2.48	—0.98	12.2	9.3	8.137	0.679—97.54	34.81	52.55	
12 » (1)	35	2	33	9.52	3.10	—0.98	13.6	9.8	13.652	0.614—303.15	78.40	144.97	
12 » (2)	35	0	35	9.56	2.07	5.7	13.5	9.9	14.161	1.792—111.92	66.37	128.10	
19 »	35	3	32	8.30	3.55	—0.98	12.9	9.2	4.043	0.116—140.59	49.05	92.58	
24 » (1)	33	2	31	8.43	3.29	—0.98	13.6	8.8	4.583	0.171—123.16	64.14	151.10	
24 » (2)	26	1	25	9.01	3.05	—0.98	13.2	9.1	8.162	0.387—172.11	79.82	150.48	
<b>Xylitol</b>													
— 1 day	41	5	36	8.20	3.80	—0.98	12.3	9.3	3.629	0.082—161.43	31.69	55.79	
0 »	41	4	37	8.18	3.48	—0.98	12.6	9.3	3.591	0.111—116.56	29.68	57.58	
1 month	40	7	33	7.07	3.95	—0.98	11.2	8.6	1.172	0.023—60.79	11.61	17.70	
4 »	40	13	27	5.52	4.77	—0.98	11.3	7.1	0.249	0.002—29.41	11.27	20.20	
8 »	39	8	31	6.69	4.26	—0.98	11.9	8.3	0.802	0.011—56.95	14.77	28.93	
12 » (1)	47	15	32	5.42	4.62	—0.98	11.5	7.6	0.225	0.002—22.92	9.68	20.68	
12 » (2)	47	11	36	6.11	4.22	—0.98	11.5	7.4	0.450	0.006—30.65	9.83	19.49	
19 »	47	17	30	4.41	4.33	—0.98	12.0	5.7	0.082	0.001—6.24	6.31	23.96	
24 » (1)	47	15	32	4.95	4.34	—0.98	12.0	7.0	0.141	0.001—10.89	7.27	24.42	
24 » (2)	44	14	30	5.02	4.44	—0.98	12.1	6.4	0.151	0.001—12.82	10.33	31.96	

Table XXX. Frequency of occurrence and total colony forming unit values of lactobacilli per ml saliva ( $n \times 10^8$ ) on Rogosa S. L. agar as a function of time and sugar diet

	N	N—	N+	Logarithmic scale					Arithmetic scale				
				$\bar{x}_{\log}$	S.D. <sub>log</sub>	Min.	Max.	Med.	$\bar{x}_{\text{geom.}}$	68 % interval	$\bar{x}_{\text{arith.}}$	S.D. <sub>arith.</sub>	
<b>Sucrose</b>													
— 1 day	31	12	19	4.10	4.33	—0.98	10.6	5.3	0.060	0.001—	4.61	3.26	7.93
0 »	30	12	18	4.00	4.39	—0.98	10.6	5.5	0.054	0.001—	4.40	4.02	8.85
1 month	33	11	22	4.55	4.20	—0.98	10.8	6.0	0.094	0.001—	6.31	3.53	9.11
4 »	33	13	20	4.34	4.60	—0.98	10.8	6.0	0.076	0.001—	7.68	6.20	13.27
8 »	33	7	26	6.15	4.16	—0.98	11.5	7.1	0.470	0.007—	30.26	10.85	20.93
12 » (1)	33	6	27	6.62	4.13	—0.98	11.5	7.3	0.756	0.012—	46.79	15.69	27.10
12 » (2)	33	4	29	7.06	3.83	—0.98	13.8	7.7	1.169	0.025—	53.70	45.20	174.22
19 »	33	10	23	5.67	4.91	—0.98	13.8	6.7	0.292	0.002—	39.86	46.42	174.69
24 » (1)	32	8	24	6.55	4.89	—0.98	12.9	8.3	0.702	0.005—	93.89	38.31	79.62
24 » (2)	26	2	24	8.15	3.52	—0.98	12.9	8.4	3.466	0.102—	117.23	50.03	107.53
<b>Fructose</b>													
— 1 day	35	13	22	4.42	4.56	—0.98	11.0	4.6	0.083	0.001—	7.97	6.96	15.34
0 »	33	9	24	5.43	4.33	—0.98	11.5	6.9	0.227	0.003—	17.32	9.22	20.91
1 month	34	15	19	3.10	3.95	—0.98	10.6	4.6	0.022	0.000—	1.16	2.40	7.58
4 »	35	3	32	7.21	3.07	—0.98	10.6	7.6	1.346	0.062—	29.09	8.95	12.99
8 »	35	1	34	8.25	2.33	—0.98	11.5	8.3	3.812	0.870—	39.32	15.76	25.05
12 » (1)	35	2	33	8.81	2.97	—0.98	12.9	9.2	6.751	0.347—	131.28	41.06	76.50
12 » (2)	35	1	34	8.47	2.70	—0.98	12.6	8.3	4.791	0.321—	71.49	33.69	62.35
19 »	37	8	27	6.52	4.52	—0.98	12.2	7.8	0.676	0.007—	62.07	24.99	50.63
24 » (1)	33	4	29	7.65	3.85	—0.98	12.9	8.7	2.109	0.044—	99.28	38.07	81.54
24 » (2)	26	2	24	8.46	3.41	—0.98	12.6	8.8	4.734	0.155—	144.70	45.30	76.93
<b>Xylitol</b>													
— 1 day	47	23	24	3.39	4.51	—0.98	11.5	4.6	0.030	0.000—	2.71	4.57	15.37
0 »	46	19	27	3.80	4.25	—0.98	10.8	5.5	0.045	0.000—	3.14	3.12	9.13
1 month	47	19	28	3.46	3.85	—0.98	10.1	4.6	0.031	0.000—	1.48	1.24	3.98
4 »	47	20	27	4.28	4.74	—0.98	10.6	6.2	0.072	0.000—	8.28	5.57	10.39
8 »	46	16	30	4.79	4.46	—0.98	10.8	6.6	0.120	0.001—	10.45	5.23	11.25
12 » (1)	47	16	31	4.86	4.42	—0.98	10.8	6.9	0.129	0.001—	10.72	4.55	9.15
12 » (2)	47	13	34	5.51	4.23	—0.98	10.8	7.3	0.246	0.003—	17.02	5.48	10.91
19 »	47	22	25	3.43	4.36	—0.98	11.3	5.3	0.030	0.000—	2.42	3.68	12.31
24 » (1)	47	15	32	4.69	4.15	—0.98	11.3	6.4	0.109	0.001—	6.96	4.39	12.75
24 » (2)	44	14	30	4.73	4.26	—0.98	11.4	6.0	0.113	0.001—	8.03	6.49	18.50

Table XXXI. Frequency of occurrence and total colony forming unit values of streptococci per ml saliva ( $n \times 10^8$ ) on Rogosa S. L. agar as a function of time and sugar diet

	N	N-	N+	Logarithmic scale					Arithmetic scale				
				$\bar{x}_{\log}$	S.D. <sub>log</sub>	Min.	Max.	Med.	$\bar{x}_{\text{geom.}}$	68 % interval	$\bar{x}_{\text{arith.}}$	S.D. <sub>arith.</sub>	
<b>Sucrose</b>													
— 1 day	33	9	24	5.98	4.53	—0.98	11.5	7.6	0.395	0.004—	36.72	10.81	19.88
0 »	31	10	21	5.55	4.73	—0.98	10.7	7.6	0.256	0.002—	29.34	8.53	12.64
1 month	33	7	26	6.62	4.22	—0.98	10.6	8.3	0.752	0.011—	51.15	9.20	10.77
4 »	33	16	17	4.07	5.09	—0.98	10.8	5.7	0.059	0.000—	9.49	7.76	14.68
8 »	33	12	21	5.34	5.04	—0.98	11.5	7.6	0.208	0.001—	32.34	13.06	22.53
12 » (1)	33	12	21	5.57	5.22	—0.98	13.8	7.6	0.263	0.001—	48.71	42.81	173.25
12 » (2)	32	13	19	5.15	5.43	—0.98	13.8	6.7	0.172	0.001—	39.15	49.26	176.66
19 »	33	12	21	5.35	5.20	—0.98	13.8	6.9	0.211	0.001—	38.46	48.34	174.90
24 » (1)	32	15	17	4.71	5.68	—0.98	14.2	6.1	0.111	0.000—	32.66	69.01	243.69
24 » (2)	26	7	19	6.58	5.14	—0.98	13.8	7.6	0.719	0.004—	122.9	81.73	208.91
<b>Fructose</b>													
— 1 day	35	4	31	7.86	3.56	—0.98	11.5	9.2	2.582	0.073—	91.01	19.56	30.06
0 »	34	3	31	7.89	3.38	—0.98	12.2	8.8	2.687	0.091—	79.29	24.40	45.09
1 month	34	6	28	7.10	3.99	—0.98	11.5	8.4	1.215	0.022—	65.88	12.75	20.84
4 »	35	14	21	4.60	4.84	—0.98	11.5	6.9	0.099	0.001—	12.49	9.43	20.99
8 »	35	9	26	6.47	4.65	—0.98	11.5	8.0	0.644	0.006—	67.13	17.69	30.39
12 » (1)	35	7	28	7.61	4.55	—0.98	12.9	9.2	2.026	0.021—	190.95	36.02	72.24
12 » (2)	35	5	30	7.78	4.00	—0.98	12.9	9.2	2.384	0.044—	129.86	31.67	69.55
19 »	35	10	25	6.21	4.84	—0.98	12.2	8.0	0.498	0.003—	63.12	22.47	44.30
24 » (1)	33	9	24	5.61	4.56	—0.98	12.9	6.6	0.276	0.003—	26.28	25.78	73.84
24 » (2)	26	9	17	5.63	5.17	—0.98	12.9	7.8	0.280	0.002—	48.99	34.38	87.37
<b>Xylitol</b>													
— 1 day	47	10	37	7.02	4.48	—0.98	12.3	8.6	1.118	0.013—	98.75	22.55	44.24
0 »	46	9	37	7.04	4.30	—0.98	12.6	8.7	1.137	0.015—	84.20	22.53	50.23
1 month	47	12	35	6.02	4.34	—0.98	10.8	7.3	0.410	0.005—	31.45	8.49	13.82
4 »	47	33	14	1.89	4.52	—0.98	10.8	—0.98	0.006	0.000—	0.61	3.95	10.44
8 »	46	21	25	4.23	4.95	—0.98	11.5	6.4	0.069	0.000—	9.73	7.26	16.75
12 » (1)	47	26	21	3.17	4.79	—0.98	10.8	—0.98	0.024	0.000—	2.87	5.02	11.97
12 » (2)	47	26	21	3.09	4.69	—0.98	10.8	—0.98	0.022	0.000—	2.40	4.28	10.83
19 »	47	26	21	2.57	4.13	—0.98	11.3	—0.98	0.013	0.000—	0.81	2.59	11.91
24 » (1)	47	28	19	2.39	4.26	—0.98	11.3	—0.98	0.010	0.000—	0.77	2.88	12.00
24 » (2)	44	23	21	2.92	4.31	—0.98	11.5	—0.98	0.019	0.000—	1.39	3.24	18.31

Table XXXII. Frequency of occurrence and total colony forming unit values of candida per ml saliva ( $n \times 10^6$ ) on Rogosa S. L. agar as a function of time and sugar diet

	N	N—	N+	Logarithmic scale					Arithmetic scale			
				$\bar{x}_{\log}$	S.D. <sub>log</sub>	Min.	Max.	Med.	$\bar{x}_{\text{geom.}}$	68 % interval	$\bar{x}_{\text{arith.}}$	S.D. <sub>arith.</sub>
<b>Sucrose</b>												
— 1 day	33	18	15	2.32	3.82	—0.98	8.9	—0.98	0.010	0.000—0.463	0.71	1.61
0 »	33	18	15	2.49	3.98	—0.98	8.9	—0.98	0.012	0.000—0.643	0.78	1.57
1 month	33	22	11	1.77	4.00	—0.98	9.0	—0.98	0.006	0.000—0.320	0.84	1.88
4 »	33	22	11	1.55	3.71	—0.98	9.0	—0.98	0.005	0.000—0.192	0.56	1.56
8 »	33	20	13	2.15	4.01	—0.98	8.7	—0.98	0.009	0.000—0.474	0.78	1.61
12 » (1)	33	21	12	1.96	4.03	—0.98	8.7	—0.98	0.007	0.000—0.399	0.78	1.55
12 » (2)	33	21	12	1.78	3.84	—0.98	9.7	—0.98	0.006	0.000—0.275	0.95	3.10
19 »	33	20	13	1.97	3.78	—0.98	8.0	—0.98	0.007	0.000—0.311	0.44	0.86
24 » (1)	32	19	13	2.20	3.97	—0.98	8.6	—0.98	0.009	0.000—0.480	0.62	1.16
24 » (2)	26	18	8	1.29	3.53	—0.98	8.5	—0.98	0.003	0.000—0.124	0.33	0.94
<b>Fructose</b>												
— 1 day	35	13	22	3.85	4.00	—0.98	10.8	5.3	0.047	0.001—2.556	2.67	8.84
0 »	35	10	25	4.49	3.81	—0.98	10.8	5.3	0.090	0.002—4.023	2.75	8.70
1 month	34	16	18	3.29	4.22	—0.98	9.9	5.0	0.027	0.000—1.829	1.66	4.01
4 »	34	13	21	3.73	3.96	—0.98	9.4	4.6	0.042	0.001—2.202	1.37	2.78
8 »	35	14	21	3.82	4.14	—0.98	9.6	5.3	0.046	0.001—2.878	1.61	3.04
12 » (1)	35	12	23	4.24	4.05	—0.98	9.9	5.7	0.070	0.001—4.002	2.19	4.52
12 » (2)	35	14	21	3.63	3.99	—0.98	8.9	4.6	0.038	0.001—2.048	1.18	2.06
19 »	35	13	22	4.05	4.10	—0.98	9.4	5.7	0.057	0.001—3.444	1.69	2.98
24 » (1)	33	12	21	3.71	3.79	—0.98	9.0	4.6	0.041	0.001—1.822	0.99	1.82
24 » (2)	26	10	16	3.32	3.65	—0.98	9.6	4.6	0.028	0.001—1.066	0.93	2.94
<b>Xylitol</b>												
— 1 day	47	24	23	2.65	3.89	—0.98	9.5	—0.98	0.014	0.000—0.690	0.95	2.55
0 »	47	26	21	2.33	3.89	—0.98	9.8	—0.98	0.010	0.000—0.502	1.11	3.35
1 month	47	32	15	1.32	3.47	—0.98	9.5	—0.98	0.004	0.000—0.120	0.46	1.92
4 »	47	40	7	0.03	2.47	—0.98	7.8	—0.98	0.001	0.000—0.012	0.09	0.38
8 »	44	36	8	0.21	2.58	—0.98	7.2	—0.98	0.001	0.000—0.016	0.07	0.23
12 » (1)	47	37	10	0.55	3.02	—0.98	8.3	—0.98	0.002	0.000—0.036	0.19	0.64
12 » (2)	47	34	13	0.94	3.23	—0.98	9.0	—0.98	0.003	0.000—0.065	0.37	1.35
19 »	47	40	7	0.04	2.49	—0.98	7.1	—0.98	0.001	0.000—0.013	0.07	0.21
24 » (1)	47	38	9	0.27	2.64	—0.98	7.5	—0.98	0.001	0.000—0.017	0.09	0.30
24 » (2)	44	36	8	0.23	2.63	—0.98	6.9	—0.98	0.001	0.000—0.017	0.08	0.21

Table XXXIII. Significance level of differences in the salivary colony forming unit values on Rogosa S. L. agar within the sugar groups during the diet period (Friedman test)

Lactobacilli	S	0.0004
	F	<0.0001
	X	<0.0001
Streptococci	S	n.s.
	F	0.0004
	X	<0.0001
Total CFU	S	0.0258
	F	<0.0001
	X	<0.0001

Table XXXIV. Significance level on differences in the salivary colony forming unit values on Rogosa S. L. agar between the sugar groups during the diet period (Kruskal-Wallis and Mann-Whitney U-tests)

		-1	0	1	4	8	(1)	(2)	(1)	(2)	
		day	day	month	months	months	12	12	19	24	
		months									
Lacto- bacilli	Kruskal- Wallis	n.s.	n.s.	n.s.	0.0263	0.0000	0.0000	0.0081	0.0056	0.0041	0.0001
	S/F	n.s.	n.s.	n.s.	0.0174	0.0513	0.0143	n.s.	n.s.	n.s.	n.s.
S/X	U-test	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	0.0321	0.0297	0.0008
F/X		n.s.	n.s.	n.s.	0.0179	0.0006	0.0000	0.0021	0.0013	0.0010	0.0001
Strepto- cocci	Kruskal- Wallis	n.s.	n.s.	n.s.	n.s.	n.s.	0.0002	0.0002	0.0011	0.0158	0.0043
	S/F	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
S/X	U-test	n.s.	n.s.	n.s.	n.s.	n.s.	0.0284	n.s. (0.0638)	0.0101	0.0353	0.0019
F/X		n.s.	n.s.	n.s.	n.s.	n.s.	0.0000	0.0002	0.0002	0.0033	0.0127
Total CFU	Kruskal- Wallis	n.s.	n.s.	n.s.	n.s.	n.s.	0.0000	0.0005	0.0001	0.0015	0.0000
	S/F	n.s.	n.s.	n.s.	n.s.	n.s.	0.0190	0.0454	n.s.	n.s.	n.s.
S/X	U-test	n.s.	n.s.	n.s.	n.s.	n.s.	0.0282	n.s.	0.0042	0.0132	0.0002
F/X		n.s.	n.s.	n.s.	n.s.	n.s.	0.0000	0.0001	0.0000	0.0005	0.0002

Table XXXV. Frequency of occurrence and total colony forming unit values on Sabouraud antibiotic agar per ml saliva ( $n \times 10^6$ ) as a function of time and sugar diet

	N	N—	N+	Logarithmic scale					Arithmetic scale			
				$\bar{x}_{\log}$	S.D. $_{\log}$	Min.	Max.	Med.	$\bar{x}_{\text{geom.}}$	68 % interval	$\bar{x}_{\text{arith.}}$	S.D. $_{\text{arith.}}$
<b>Sucrose</b>												
— 1 day	33	18	15	2.32	3.82	—0.98	8.9	—0.98	0.010	0.000—0.468	0.71	1.61
0 »	33	18	15	2.49	3.98	—0.98	8.9	—0.98	0.012	0.000—0.643	0.78	1.57
1 month	33	22	11	1.77	4.00	—0.98	9.0	—0.98	0.006	0.000—0.320	0.84	1.88
4 »	33	22	11	1.55	3.71	—0.98	9.0	—0.98	0.005	0.000—0.192	0.56	1.56
8 »	33	20	13	2.15	4.01	—0.98	8.7	—0.98	0.009	0.000—0.474	0.78	1.61
12 » (1)	33	21	12	1.96	4.03	—0.98	8.7	—0.98	0.007	0.000—0.399	0.78	1.55
12 » (2)	33	21	12	1.78	3.84	—0.98	9.7	—0.98	0.006	0.000—0.275	0.95	3.10
19 »	33	20	13	1.97	3.78	—0.98	8.0	—0.98	0.007	0.000—0.311	0.44	0.86
24 » (1)	32	19	13	2.20	3.97	—0.98	8.6	—0.98	0.009	0.000—0.480	0.62	1.16
24 » (2)	26	18	8	1.29	3.53	—0.98	8.5	—0.98	0.003	0.000—0.124	0.33	0.94
<b>Fructose</b>												
— 1 day	35	13	22	3.85	4.00	—0.98	10.8	5.3	0.047	0.001—2.556	2.67	8.84
0 »	35	10	25	4.49	3.81	—0.98	10.8	5.3	0.090	0.002—4.023	2.75	8.70
1 month	34	16	18	3.29	4.22	—0.98	9.9	5.0	0.027	0.000—1.829	1.66	4.01
4 »	34	13	21	3.73	3.96	—0.98	9.4	4.6	0.042	0.001—2.202	1.37	2.78
8 »	35	14	21	3.82	4.14	—0.98	9.6	5.3	0.046	0.001—2.878	1.61	3.04
12 » (1)	35	12	23	4.24	4.05	—0.98	9.9	5.7	0.070	0.001—4.002	2.19	4.51
12 » (2)	35	14	21	3.63	3.99	—0.98	8.9	4.6	0.038	0.001—2.048	1.18	2.06
19 »	35	13	22	4.05	4.10	—0.98	9.4	5.7	0.057	0.001—3.444	1.69	2.98
24 » (1)	33	12	21	3.71	3.79	—0.98	9.0	4.6	0.041	0.001—1.822	0.99	1.82
24 » (2)	26	10	16	3.32	3.65	—0.98	9.6	4.6	0.028	0.001—1.066	0.93	2.94
<b>Xylitol</b>												
— 1 day	47	24	23	2.65	3.89	—0.98	9.5	—0.98	0.014	0.000—0.690	0.95	2.55
0 »	47	26	21	2.33	3.89	—0.98	9.8	—0.98	0.010	0.000—0.502	1.11	3.35
1 month	47	32	15	1.32	3.47	—0.98	9.5	—0.98	0.004	0.000—0.120	0.46	1.92
4 »	47	40	7	0.03	2.47	—0.98	7.8	—0.98	0.001	0.000—0.012	0.09	0.33
8 »	44	36	8	0.21	2.58	—0.98	7.2	—0.98	0.001	0.000—0.016	0.07	0.23
12 » (1)	47	37	10	0.55	3.02	—0.98	8.3	—0.98	0.002	0.000—0.036	0.19	0.64
12 » (2)	47	34	13	0.94	3.23	—0.98	9.0	—0.98	0.003	0.000—0.065	0.37	1.35
19 »	47	40	7	0.04	2.49	—0.98	7.1	—0.98	0.001	0.000—0.013	0.07	0.21
24 » (1)	47	38	9	0.27	2.64	—0.98	7.5	—0.98	0.001	0.000—0.018	0.09	0.30
24 » (2)	44	36	8	0.23	2.63	—0.98	6.9	—0.98	0.001	0.000—0.017	0.08	0.21

Table XXXVI. Significance level of differences in the salivary colony forming unit values on Sabouraud antibiotic agar between the sugar groups during the diet period (Kruskal-Wallis and Mann-Whitney U-tests)

	—1	0	1	4	8	(1) 12	(2) 12	19	(1) 24	(2) 24
Kruskal-Wallis	n.s.	n.s.	n.s.	0.0006	0.0011	0.0009	0.0163	0.0002	0.0012	0.0096
Mann-Whitney U-test										
S/F	n.s.	n.s.	n.s.	0.0272	n.s.	0.0419	n.s.	0.0319	n.s.	n.s.
S/X	n.s.	n.s.	n.s.	0.0335	0.0149	n.s.	n.s.	0.0092	0.0128	n.s.
F/X	n.s.	n.s.	n.s.	0.0000	0.0000	0.0000	0.0014	0.0000	0.0000	0.0003