THE EFFECTS OF A DIETARY SUPPLEMENT OF FRESH ORANGES ON THE ORAL HEALTH OF CHILDREN

By

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Submitted to the Faculty of the Graduate School in partial fulfillment of the requirements for the degree of Master of Science in Dentistry, Indiana University School of Dentistry, 1973.

ACKNOWLEDGEMENTS

The author would like to dedicate this thesis to his wife, Dr. Diane Carol Halvorson Dilley, whose patience and understanding were unending during the undertaking of this project.

Special thanks are extended to Dr. Grant Palmer and the Sunkist Corporation whose grant made this project possible and whose help in performing the diet analysis was a monumental effort.

The author owes much gratitude to Dr. James R. Roche, as chairman of Graduate Pedodontics, and as Chairman of his Graduate Committee. He has been of unmeasurable assistance to the author in guiding this undertaking as well as his graduate education.

The author is grateful to Dean Ralph E. McDonald for his assistance in making this project available and function smoothly and to Dr. Joseph C. Muhler for the initiation of this study. Professor Paul Barton, as a member of this Graduate Committee, was of great assistance in editing this work.

The author would like to thank Dr. Leonard G. Koerber and Dr. Rose H. Potter for their expertise and aid in the statistical analysis of this study. The author also acknowledges other members of this Graduate Committee: Dr. Paul E. Starkey and Dr. James E. House.

The author thanks the Hematology Department of Indiana University and Ms. Joyce Compton for their assistance in drawing and analysis of blood samples. The author also thanks Ms. Anita J. Weaver for her work as dental hygienist and Ms. Jeri Gruner for her help as dental assistant.

Finally the author must thank the children and staff of the Soldiers and Sailors Children's Home for their cooperation in this study.

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INTRODUCTION

Helping patients to maintain healthy oral soft tissues is one of the primary responsibilities of the dentist. Practically every child has some degree of gingival inflammation, which may vary with each individual from a localized marginal gingivitis to a chronic inflammation leading to tissue destruction.

Many theories, which go beyond providing good oral hygiene and proper diet, have been proposed to reduce gingival inflammation. One of these theories deals with supplementing the diet with additional vitamin C or citrus fruit. Additional synthetic ascorbic acid has been added to the diets of children by Kyhos[1] and Cohen,[2] with resulting decreases in inflammation of the periodontal tissues. Shannon and Gibson[3] and Pertlitsh et al.,[4] however, found no positive correlation between additional synthetic ascorbic acid and periodontal conditions. Nearly all the investigations using supplemental citrus fruit or juices have demonstrated a beneficial effect on the periodontium by decreasing inflammation. These studies include those by Mead,[5] Thomas,[6] Hanke,[7] and El-Ashiry et al.[8]

Very few studies have been conducted relating the effects of additional natural citrus fruit on the periodontium. The present investigation examined the effects on oral conditions of adding three oranges per day to the diet of children already receiving a balanced diet. It was theorized that over a prolonged period of time that the group receiving the oranges would demonstrate healthier gingival tissues than a control group.

REVIEW OF THE LITERATURE

I. Effects of Synthetic Ascorbic Acid and Citrus Fruits on the Periodontium

Over the past few decades, the effect of citrus fruit or the effect of ascorbic acid (vitamin C), which is an important component of citrus fruit, on the periodontium has been debated in the dental literature. The exact relationship between the ascorbic acid or citrus fruit and gingival health is not clear. Wolbach and Howe[9] postulated that in an ascorbic acid deficiency the supporting tissues cannot produce or maintain intercellular substances adequately and that ascorbic acid is therefore necessary.

The need for ascorbic acid and consumption of the substance vary with the individual. The recommended daily dietary allowance of ascorbic acid for ages one to 32 years is 40-60 mg.[10] Blockley and Baenziger,[11] on the basis of a controlled but not thoroughly documented study of 41 patients, proposed a correlation between lowered vitamin C content of the blood and certain periodontal disturbances. Where a vitamin C deficiency existed with a concurrent periodontal disturbance, the inflammation of the gingiva could not be reduced with localized treatment alone. In agreement with this view was Burrill's[12] report of 1396 patients over a two-year period, in which the blood plasma vitamin C levels tended to be lower in patients with gingivitis. Patients with periodontal disease exhibited a tendency toward lowered plasma ascorbic acid levels, but the total difference between these groups and their ascorbic acid levels was small and probably not significant.

Hard et al.,[13] found that in 248 children receiving what he considered marginal amounts of ascorbic acid (69-82 mg mean daily), the subclinical signs assessed by the biomicroscopic method suggested that increased chronic gingival lesions are more frequent than the acute form. Cohen[2] treated twelve mentally retarded patients who had poor oral hygiene and extensive gingival problems of inflammation, enlargement and even necrotizing ulcerative gingivitis with 500 mg of ascorbic acid daily. No attempt was made to improve oral hygiene. After four weeks no changes in the soft tissues were noted, but during the fifth week the inflammation and enlargement began to subside, and after ninety days the gingival condition of all patients had improved markedly and there was a cessation of the superimposed acute necrotizing gingivitis. In addition, there was a reduction in the amount of materia alba, calculus, and stain.

Kyhos[1] conducted a study of 71 adult males for 17 months to test the effects of 25 mg, 50 mg, and 75 mg of ascorbic acid supplementation on periodontal tissues. The conditions of the soft tissues of all subjects improved, but those receiving 50 mg and 75 mg supplements demonstrated the greatest improvement. When placebos were then substituted, the periodontium returned to its puffiness and abnormal redness after four to five weeks. There was no mention of controls in this study.

In a study by Linghorne and associates[14] of 150 Royal Canadian Air Force personnel under 30 years of age, both males and females, supplements of vitamins (including ascorbic acid) over an eight-month period did not affect gingival inflammation when used alone. However, once inflammation was eliminated by cleaning the teeth, removing calculus, and giving oral hygiene instructions, a supplement of 75 mg of ascorbic acid per day delayed the recurrence of signs of inflammation beyond what had occurred with a supplement of 10 or 25 mg of ascorbic acid.

Weisberger, Young, and Morse[15] in a study of 65 patients concluded that vitamin C therapy could not produce a complete cure in certain long-standing cases of periodontal disease which included bleeding, hyperemia, and loss of tissue firmness. They found more periodontal problems in subjects with less than 0.7 mg % plasma ascorbic acid. These same patients showed improvement with additional ascorbic acid therapy.

Shannon and Gibson[3] in a study of 341 adult males found no correlation (as determined by correlation coefficients) between ascorbic acid in the blood, and caries or periodontal index as evaluated by a method devised by O'Leary.[16] Perlitsh and associates[4] reported the same lack of correlation between ascorbic acid levels and gingival health in 26 men stationed in Antarctica during a 10-month period. The Papillary, Marginal, and Attached Gingival (P.M.A.) Index was used to evaluate the gingival condition. King,[17] Ungley and Horton[18] showed that vitamins in large doses failed to exert any beneficial effect on gingivitis. Retarski and co-workers,[19] in a controlled study of 66 subjects over a 6-month period, stated that the assumption of a scorbutic basis for gingivitis is unwarranted unless there is antecedent or present clinical evidence of scurvy. Parfitt and Speirs[20] reviewed the literature and concluded that a nutritional deficiency is not an etiologic factor in periodontal diseases encountered on the North American continent. Therefore, treatment with supplements of vitamin C is unwarranted unless a definite deficiency can be shown.

Grandison, Scott, and Cruickshank[21] investigated the influence of synthetic vitamin C in 20 children and 20 controls during a two-year period. They concluded that 200 mg per day of synthetic ascorbic acid had no discernable effect on soft tissue structures.

In an effort to compare natural vitamin C and the synthetic form, Todhunter and co-workers[22] fed animals comparable amounts of synthetic ascorbic acid in aqueous solution and lemon juice. The results revealed that blood plasmas and adrenal content of ascorbic acid were the same, as were weight gains; but the animals which were fed the lemon juice showed fewer hemorrhages when scored for scurvy. These results indicate that lemon juice may contain an additional factor which aids in the prevention of hemorrhages characteristic of scurvy.

In some human studies there appears to be an additional factor or a better utilization of ascorbic acid in its natural form than in the synthetic form. Linghorne and associates[14] in a study fed equal amounts of natural and synthetic forms of ascorbic acid to 150 Royal Canadian Air Force personnel. The group receiving the natural form (in foods) had less recurrence of inflammation after the teeth had been cleaned, all calculus removed, and oral hygiene instructions given. The inflammation was assigned to one of four classifications as judged by color photographs of the patients. The difference between the groups was not statistically significant.

Using the natural form of ascorbic acid, several researchers have noted an improvement in gingival conditions without concurrent damage to hard tissues. A study by Thomas[6] involving the ingestion of 8 oz. of reconstituted orange juice daily for over one year by 27 young adults (sophomore and junior dental students), demonstrated less gingival pathology than a control group of the same size. This consumption of juice showed no increase in susceptibility to dental caries, no effect on the number of lactobacilli, no erosion of dental hard tissues, and no hypersensitivity of cervical cementum, as measured by a detailed clinical examination, complete mouth radiographs, Kodachrome photographs, and bacteriologic examination of saliva. Mead[5] conducted a study of 131 male prisoners, ages 19-55 years, 67 of whom ate one-half a grapefruit three times a day for five months; oral examination revealed an improved tone of oral tissues in the study group,, with less soreness and tenderness, and a decrease in gingival bleeding. The methods of examination were not given. The improvement in the health of the oral tissues was experienced by 99% of the study group while only 2% of the control group showed any improvement. No decalcification or other deleterious effects on tooth enamel were found.

Hanke[7] reported on a study of children aged 10-17 years. The experimental group was composed of 341 children with the following conditions: 242 who had varying gingival pathology ranging from a mild to a spongy form of gingivitis and 99 who were free of any gingival pathology. These children consumed 16 oz. of fresh orange juice plus the juice of one lemon per day for one year. A control group of 99 children with varying gingival conditions did not receive the fresh citrus juices. No oral hygiene instructions were given and no plaque or calculus was removed for either the experimental or control group.

The results were reported as follows for the experimental group. Of the 99 previously free from gingivitis, none developed a gingival condition during the test period. Eighty children with mild gingivitis, obvious thickening of gingiva, possible hyperemia, and response to pressure as if edematous, all showed improvement after the one-year period. Eighty children with severe gingivitis, decidedly thickened, quite hyperemic gingiva, and responded to pressure as if edematous, demonstrated the following after treatment: 76 (95%) had healthy oral tissues, 1 (1%) showed improvement of oral tissues, and 2 (3%) did not show improvement; one child was lost from this group. The remaining 80 children with the spongy form of gingivitis, which in addition to the previously mentioned conditions would hemorrhage under mild mechanical irritation, responded to treatment in the following manner: 47 (55%) became healthy, 19 (24%) showed improvement, and 13 (16%) did not improve at the end of the juice consumption period; one child was lost from this group.

In summary, of 242 children with gingivitis of varying degrees, 86% became healthy, 8% showed improvement, and 6% did not improve. The 99 children free of any gingivitis remained in this category. Over a similar period of time which was to serve as a recheck period and with the elimination of the additional citrus juice, 144 of 264 of the remaining subjects' periodontal health began to deteriorate to the inflammatory conditions that pre-existed before the study began.

In a more recent study at the University of Alabama, El-Ashiry and associates[8] demonstrated varying improvement of gingival conditions over a three-week period in 102 male and female patients ages 20-59 years who were treated by different methods, including prophylaxis, supplements of bioflavonoids, or with natural or synthetic vitamin C. The rates of improvement of the different groups are as follows:

1. Prophylaxis, additional natural vitamin C concentrate and bioflavonoids treatment (71% improvement).
2. Prophylaxis, additional synthetic vitamin C and bioflavonoids treatment (69% improvement).
3. Prophylaxis and additional synthetic vitamin C treatment (58% improvement).
4. No prophylaxis, but additional natural vitamin C concentrate and bioflavonoids treatment (57% improvement).
5. No prophylaxis, but additional synthetic vitamin C and bioflavonoids treatment (57% improvement).
6. No prophylaxis, but additional synthetic vitamin C treatment (45% improvement).
7. Prophylaxis and placebo (30% improvement).
8. No prophylaxis, but a placebo (10% improvement).

These results, however, must be tempered with the authors' comment that additional vitamin C above the tissue saturation level may not affect periodontal conditions. Both plasma ascorbic acid and tissue ascorbic acid tests were used for evaluation. This study indicated that vitamin C therapy was effective only in clinical or subclinical vitamin C deficiencies.

In contradiction to the above studies, Hicks[23] made several subjective observations on excessive citrus juice consumption, with two to three oranges or grapefruit being considered excessive. He expressed the view that consumption of citrus fruit or juices in large quantities (such as 6 to 8 oz. of juice per day) is harmful to the deep and superficial tissues of the oral cavity. These harmful effects include hyperemic gingival tissues, easy and excessive bleeding, marked mobility of teeth, loss of alveolar process, and hypersensitivity of oral tissues. Hicks stated that these effects occur over a prolonged period of one to fifteen years. His observation were based on clinical examinations and radiographs over a 15-year period. The methods of evaluation were not given.

The evidence for lessening gingival inflammation by using a supplement of synthetic or natural vitamin C is therefore very confusing. Findings appear to show that an ascorbic acid supplement in a synthetic or natural form may be either effective or ineffective in influencing gingival conditions. Very few studies are helpful in determining the value of natural ascorbic acid supplements by themselves. Also, the comparative worth between the natural and synthetic forms of ascorbic acid for gingival health remains unclear.

II. Epidemiology of Gingivitis

The high prevalence of gingivitis in children is well documented. Massler, Cohen, and Schour[24] using the PMA Index found that in 17,079 children, 6-16 years old, almost all had some degree of gingival inflammation. Benjamin, Russell, and Smiley[25] in 25 Indiana counties reported that of 3,880 rural white children ages 9-20 years, the average per county periodontal score as evaluated by the Russell Index was above the score necessary for all children to have some type of periodontal condition. Moore[26] reported that in 1,123 children ages 7-13 years that gingivitis was present in 93% of the children as determined by the PMA Index.

Recently, in unpublished data from the Dental Health Task Force Project 1970-1972, Bell[27] reported that nearly all of the 11,228 children ages 6-18 years residing in Indiana, who were examined using the Russell Index, had gingivitis to some degree.

James and associates[28] found that the prevalence of gingivitis in 11 and 12 year olds varied between 8% and 40%. Carter and Wells[29] observed that 50% of 29,500 children ages 6 to 12 years had gingivitis in 88% to 97% of 183 children ages 12-14 years. Jameson and coworkers[31] determined that gingivitis was present to some degree in 99% of 159 children ages 5 to 14 years. These studies indicate that gingivitis has a wide prevalence in children.

III. Epidemiology of Dental Caries

In children ages 6 to 18 years dental caries has almost as high an incidence as gingivitis. Miller and Chrietzburg[32] reported that in a group of 1,578 white children 6-15 years old in a fluoridated Georgia area, 60.5% had dental caries in permanent teeth; in another group of 983 white children ages 6-11 years in the same area, 76.6% had dental caries in primary teeth. The decayed, missing or filled (DMF) teeth rate of the first group averaged 2.38 per child, while the decayed or filled rate of the second group averaged 3.44 per child. The ADA Type III Dental Examination (mouth mirrors, explorers, and artificial light) was used and no radiographs were taken.

Wachowski[33] demonstrated in 11 counties of Arizona that the averages of all age groups ages 5-13 years needing dental restorations ranged from 66.5% to 86.5%. Brucker[34] using mouth mirrors, explorers, college pliers and scalers, demonstrated that of 1,864 white children ages 4-14 years, 41.8% of the first primary molars were carious and 51% of the second primary molars were carious. Filled teeth were not included in this study and no radiographs were used.

In an unpublished Dental Health Task Force Project, Bell[27] reported that in nine-year olds, a dmft average of 3.59 existed in primary teeth. Of the permanent teeth for 16-year olds in the same study, a DMFT of 5.67 was reported. No radiographs were used in these determinations. These studies indicate that a high percentage of children have a significant amount of dental caries.

IV. Relationship of Plaque to Dental Caries and Gingivitis

Plaque has been described as a nitrogenous network of mucin, desquamated cells and microorganisms. The relationship of plaque and dental caries formation has been hypothesized by Easlick[35] in the following formula:

Refined carbohydrate + Bacteria = acid plaque

Acid plaque + susceptible tooth surface = dental caries

Shafer, Hine, and Levy[36] in their text propose that plaque is a definite contributing factor to at least the initiation of carious lesions. They also state that enamel caries begins beneath the dental plaque. The presence of dental plaque, however, does not mean that a carious lesion will ensue. Graf[37] has stated that low pH values, as measured by glycolytic activity in plaque at the enamel surface, are thought to represent the initial cariogenic attack. His conclusions were based on a five-year study with over 400 recordings on five patients.

Thus it appears that a relationship exists between plaque formation and the possibility of dental caries occurring on a susceptible tooth surface.

Another positive relationship appears to exist between plaque formation and the initiation of gingival inflammation. Shafer, Hine and Levy[38] have stated that microorganisms must be listed as one of the causes of gingivitis. These microorganisms reside in plaque on the teeth. Ash and Giltin[39] demonstrated a high degree of correlation (0.66 with a probability of 0.01) between plaque and gingivitis.

Loe[40] concluded from his study that if plaque is allowed to form and remain for ten to twenty-one days, the adjacent gingiva will show inflammatory changes. This was demonstrated in a study using an analysis of Kodachrome slides.

In a study by Eichel,[41] a significant correlation was found between the amount of plaque formation in 48 hours and the periodontal condition of the anterior teeth as scored with the PMA Index and Periodontal Index for 56 children ages 8-12 years. Therefore, it may be assumed that deposits of plaque on the teeth contribute to gingivitis and marginal periodontal disease.

METHODS AND MATERIALS

Three hundred children, full-time boarding students at the Soldiers and Sailors Children's Home of Knightstown, Indiana, were chosen for this study. Both sexes were included and the age range was six through twenty years. The children were evaluated for gingival condition, plaque formation, calculus deposits, and decayed, missing or filled surfaces of the teeth. They were subsequently divided into experimental and control groups with 150 children in each group after the initial data had been collected.

To assess gingival status, the PMA Index criteria as originated by Massler, Cohen and Schour[24] were used on the gingiva of each of the four vertical sides of each tooth. These criteria and their corresponding numerical values are:

0. No gingivitis, pale pink color, firm texture, close adherence to tooth, tissue should fill interdental space.
1. Mild inflammation with slight change in color and little loss of contour or change in texture.
2. Moderate inflammation with swelling and redness. Papilla or margins become blunted or rounded.
3. Severe inflammation, marked swelling and redness, pocket formation and spontaneous bleeding. There is involvement of adjacent tissues and slight degeneration.
4. Very severe inflammation, including ulceration and sloughing.

The total numerical PMA score for each mouth was divided by the total available tooth surfaces to give an average score for each child.

To assess plaque formation, each mouth was first disclosed with an erythrosin dye (Trace disclosing solution)[a]. The amount of

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[a]Trace disclosing solution concentrate, Lorvic Corp., St. Louis, Mo. 63134.

plaque was then measured by the Quigley and Hein Plaque Scoring System.[42] The plaque on the labial and lingual surfaces of each tooth was scored on a scale from zero to five, as follows:

0 no plaque

1 flecks of plaque at gingival margin

2 definite line of plaque at gingival margin

3 plaque on gingival 1/3 of tooth surface

4 plaque on gingival 2/3 of tooth surface

5 plaque on more than 2/3 of tooth surface.

The total numerical score of each mouth was then divided by the total available labial and lingual tooth surfaces to give an average score for each child.

The numbers of decayed, missing and filled tooth surfaces was calculated for each mouth. Each tooth was dried with air and examined clinically using a Kerr #4 front surface mirror and sharp SS White #23 explorer under adequate artificial light. A tooth was scored carious if the explorer point would stick and hold in an area. In addition, posterior bitewing radiographs were taken using Kodak Ultra Speed Type 2 bitewing film. These films were taken by the same person, a certified dental hygienist.

From a critical examination of the radiographs, a tooth surface was declared carious if the caries appeared to penetrate through the enamel layer. If a tooth was declared missing due to unnatural causes, a score of four was given to a posterior tooth and a score of three to an anterior tooth. No score was given to a naturally exfoliated tooth. This arbitrary system was used because a tooth may be lost due to caries without carious involvement of all surfaces. This total numerical score was compared to the total available tooth surfaces for each mouth, scoring five for a posterior tooth and four for an anterior tooth.

The four vertical surfaces of each tooth were also examined for the presence or absence of calculus. No attempt was made to quantitate the amount on each tooth surface. This total was then registered for each child.

All tooth surfaces were examined for the presence of white spots in the enamel. The number of white spots were totaled for each child.

To test the reliability of the single examiner, 21 retests on the gingival status were performed three weeks after the original examination.

All 300 children were then given a thorough prophylaxis, which included disclosing, scaling and polishing with rubber cup, Zircate Treatment Paste and dental flossing with unwaxed floss, by one of two registered dental hygienists. This procedure provided a baseline from which to identify any changes that would occur. The study group was then randomly divided into two groups, based upon the findings prior to the prophylaxis, according to age, sex, gingival condition, and plaque score so that the two groups of 150 children were as close to identical as possible.

A random sample of forty children, twenty from each of the two groups, with an equal number of males and females in each group, had blood samples drawn to examine for plasma calcium,[43] hemoglobin concentration,[44] and plasma ascorbic acid.[45] Results of these tests were to give a representation of the adequacy of the diet for three of the basic food groups: dairy, meats, and fruits or vegetables. Sample diets from the dietitian's office at the Soldiers and Sailors Children's Home were analyzed using the Home and Garden Bulletin No. 72, USDA, Aug., 1970, for all the children over a 46-week period. The menus were evaluated to ascertain whether the diet supplied the necessary nutrients and in what amounts.

Group I continued on the regularly scheduled diet to be served at the Soldiers and Sailors Children's Home with the exception of a substitution for any oranges in the diet with another non-citrus fruit or vegetable. Group II would, in addition to the same diet, consume one fresh sliced #113 orange at the end of each meal for a total of three oranges per day for 23 weeks.

The oranges were regularly distributed to the children in Group II and the eating of the oranges was supervised by two responsible adults associated with the Soldiers and Sailors Home. The physical arrangements of the dining hall provided two separate areas for food distribution and eating which maintained the dispensing of oranges and supervision of their consumption in an organized and consistent manner.

Arrangements were made with the Supervisor of the Soldiers and Sailors Children's Home to have oranges supplied to children in Group II during any periods away from their regular dining facilities. During trips provided by the Soldiers and Sailors Children's Home, the children of Group II were provided with oranges and their eating supervised. There were 7 to 12 days during the experimental period of 23 weeks which were vacation time. During these periods, oranges were sent with the children of Group II, but their eating was not supervised.

After 23 weeks (July 5, 1972 to December 11-16, 1972) from the beginning of the eating of three oranges per day by Group II, the entire remaining study sample of 246 were re-examined for gingival status, plaque formation, calculus formation, presence of white spots, and decayed, missing or filled surfaces of each tooth, as had been done initially. The decrease in the study sample from 300 to 246 over the 23 weeks was due to the removal of children from the Soldiers and Sailors Children's Home by the State Government to their natural parents. Also, 34 children of the original 40 who provided blood specimens were resampled for the same hematologic components.

Statistical analysis was performed on the following five variables:

1. Gingival score
2. Plaque score
3. Calculus score
4. D.M.F.S. score
5. White spots score

RESULTS

The data for analysis on gingival and plaque scores are averages per subject over all tooth surfaces scored. For calculus; decayed, missing and filled surfaces; and white spot scores, the descriptive statistics represent the number of affected surfaces. Since these variables are frequency variables, statistical analyses were performed on the transformations of the proportion of the number of affected surfaces to the number of total available surfaces in each subject, based on the Freeman-Tukey[46] angular transformation for frequency variables.

For each variable studied, the factorial analysis of variance with repeat measures design[47] was performed. The factors are the two groups (123 subjects per group) and the two repeated examinations which are the "before" and "after" examinations on the subjects of each group. When a statistical difference was found by the analysis of variance, further t tests were performed after the analysis of variance. With this method, the following questions may be answered simultaneously:

1. Do the two groups differ significantly on--
   a. the first examination?
   b. the second examination?
2. Is there a significant difference between the two examinations for each group?

Gingival scores: Table I shows a highly significant difference ($F=38.80$ when $F_{1,244} = 6.82$ at the .01 level of confidence) between the two examinations for both groups, examination two having the higher numerical value (see Table of means). The source of variation between groups and group-by-examination interaction was not significant.

Plaque scores: Table II shows significant differences between the two examinations and between the two groups ($F=19.53$ when $F_{1,}$

$_{244}$ = 6.82 at the .01 level of confidence and F_1 = 3.90 when $F_{1,\ 244}$ = 3.90 at the .05 level of confidence, respectively).

The significant interaction term ($F = 6.55$ when $F_{1,\ 244} = 3.90$ at the .05 level of confidence) indicates that the two groups and the two examinations may not exhibit the same pattern of difference. Therefore t tests were necessary and are shown in Table III. In Group I, the second examination showed a significant rise ($t = 4.76$ when $t_{1,\ 244} = 2.58$ at the .01 level of confidence) but not in Group II. At the first examination, the two groups did not differ on plaque scores, but on the second examination, Group I showed a significantly higher plaque score ($t = 2.92$ when $t_{1,\ 244} = 1.97$ at the .05 level of confidence) than did Group II. The F test for group-by-examination interaction in Table II also tests the differences between the two examinations for the two groups, which are shown to be significantly higher ($F = 6.55$ when $F_{1,\ 244} = 3.90$ at the .05 level of confidence) in Group I than in Group II.

<u>Transformed calculus scores:</u> Table IV shows significantly lower calculus scores in examination two for both groups. Sources of variation between groups and group-by-examination interaction were not significant.

<u>Transformed decayed, missing and filled surfaces and white spots scores:</u> Tables V and VI show that there are no significant differences either between the two groups, between the two examinations or group-by-examination interaction.

In trying to estimate the reliability of the examiner, two measurements of gingival condition were made for 21 subjects. The one way analysis of variance[47] was used to calculate for reliability estimates of the total score per subject. The reliability was defined as the variance attributable to the difference between subjects ($\sigma^2 s$) divided by the sum of the variances attributable to differences between subjects ($\sigma^2 s$) and scoring error ($\sigma^2 e$). This reliability was calculated as 74% or a scoring error of 26% as shown by Table VII.

BLOOD ANALYSIS RESULTS

The results of the blood analyses are in Table VIII. The normal average values for the three components measured are as follows:

plasma ascorbic acid	- 0.5 - 1.5 mg/100 ml[48]
plasma calcium	- 8.8 - 10.4 mg/100 ml[49]
hemoglobin	
children to 16 years	- 12.0 - 16.0 gm[50]
women	- 12.0 - 16.0 gm
men	- 14.0 - 18.0 gm

The addition of the oranges to the diet of Group II showed only differential changes in the ascorbic acid levels. Group I decreased slightly (.4 mg/100 ml) and Group II increased slightly (.2 mg/100 ml) over the 23-week test period. Plasma calcium and hemoglobin showed corresponding changes in the two groups. Plasma calcium decreased slightly (0.6 mg/100 ml) in both groups over the test period while hemoglobin increased slightly in both groups (0.1 gm).

All the averages at both testing times were within the normal ranges. However, slight fluctuations in the individual blood values were recorded. These results must be qualified as the blood analysis was performed for a representative sample from the two groups and cannot be conclusive for the entire study sample.

The results of the diet analysis showed that on a weekly basis both broups were served foods that totaled considerably more nutrients than the suggested average amounts for children. The only differences in the nutrient values were the oranges. The oranges supplied the following additional nutrients per week:

Nutrient	Weekly Average
Weight (gr)	3780.0
Water (gr)	3250.8
Food energy (cal)	1365.0
Protein (gr)	21.0
Fat	none
Carbohydrate (gr)	336.0
Calcium (mg)	1134.0
Iron (mg)	10.5
Vitamin A (I. U.)	5460.0
Thiamin (mg)	2.73
Riboflavin (mg)	1.05
Niacin (mg)	10.5
Ascorbic Acid (mg)	1386.0

The diet for both groups (not including the additional oranges of Group II) had the following weekly averages for the 23-week test period and for a 23-week control period, preceding the study:

Nutrient	23-Week Study Period	23-Week Control Period
Weight (gr)	15,777	15,744
Water (gr)	11,677	11,881
Food energy (cal)	20,007	18,517
Protein (gr)	749	702
Fat (gr)	1,669	1,601
Carbohydrate (gr)	2,235	2,148
Calcium (mg)	10,368	10,373
Iron (mg)	118.3	109.6
Vitamin A (I. U.)	75,479	69,001
Thiamin (mg)	10.58	10.14

Nutrient	23-Week Study Period	23-Week Control Period
Riboflavin (mg)	19.37	18.30
Niacin (mg)	122.91	110.02
Ascorbic Acid (mg)	806.5	855.0

TABLES

Table I

Analysis of Variance for Gingival Scores of Group I (Non-Orange Eaters) and Group II (Orange Eaters) and Between Examination 1 (Before the Oranges Were Added to the Diet) and Examination 2 (After the Oranges were Added)

Source of Variation	Degrees of Freedom	Mean Square	F	
Between groups	1	0.005	0.02	N.S.
Subjects in groups	244	0.255		
Between examinations	1	4.346	38.80	p>.001
Group X examination interaction	1	0.103	0.92	N.S.
Subjects X examination in groups	244	0.112		

Means	Group I	Group II
Examination 1	1.69	1.66
Examination 2	1.85	1.87

Table II

Analysis of Variance for Plaque Scores of Group I (Non-Orange Eaters) and Group II (Orange Eaters) and Between the Examinations 1 (Before the Oranges were Added to the Diet) and Examination 2 (After the Oranges were Added).

Source of Variation	Degrees of Freedom	Mean Square	F	
Between Groups	1	1.620	3.90	$p>.05$
Subjects in groups	244	0.415		
Between examinations	1	2.070	19.53	$p>.001$
Group X examination interaction	1	0.694	6.55	$p>.05$
Subjects X examination in groups	244	0.106		

Means	Group I	Group II
Examination 1	2.69	2.65
Examination 2	2.89	2.70

Table III

t Tests for Analysis of Comparisons Between Examinations 1 and 2 for Groups I and II. Also Analysis of Comparisons in Plaque Scores Between Group I and II at Examination 1 and 2.

Comparisons	t	
Examination 1 vs Examination 2 for Group I	4.76	p>.001
Examination I vs Examination 2 for Group II	1.19	N.S.
Group I vs Group II for Examination 1	0.77	N.S.
Group I vs Group II for Examination 2	2.92	p>.05

Table IV

Analysis of Variance for Transformed Calculus Scores of Group I (Non-Orange Eaters) and Group II (Orange Eaters) and Between Examinations 1 and 2.

Source of Variation	Degree of Freedom	Mean Square	F	
Between groups	1	1716.6	0.16	N.S.
Subjects in groups	244	11002.7		
Between examinations	1	225581.7	34.49	p>.001
Group X examination interaction	1	2288.1	0.35	N.S.
Subjects X examinations in groups	244	6540.8		

Means	Group I	Group II
Examination 1	4.97	4.66
Examination 2	1.91	2.24

Table V

Analysis of Variance for Transformed Decayed, Missing, and Filled Surfaces Scores of Group I (Non-Orange Eaters) and Group II (Orange Eaters) and Between Examinations 1 and 2.

Source of Variation	Degree of Freedom	Mean Square	F	
Between groups	1	2423.7	0.28	N.S.
Subjects in groups	244	8759.2		
Between exams	1	26.78.7	2.55	N.S.
Group X examination interaction	1	125.0	0.12	N.S.
Subjects X examination in groups	244	1050.8		

Means	Group I	Group II
Examination 1	11.54	10.89
Examination 2	11.81	12.08

Table VI

Analysis of Variance for Transformed White Spot Scores of Group I (Non-Orange Eaters) and Group II (Orange Eaters) and Between Examinations 1 and 2.

Source of Variation	Degree of Freedom	Mean Square	F	
Between groups	1	1984.0	1.29	N.S.
Subjects in groups	244	1535.1		
Between examinations	1	3.3	0.01	N.S.
Group X examinations interaction	1	173.3	0.27	N.S.
Subjects X examinations in groups	244	643.4		
Means	Group I	Group II		
Examination 1	.15	.15		
Examination 2	.12	.17		

Table VII

One Way Analysis of Variance for Estimates of Reliability of Gingival Scores by the Examiner

Source of Variation	Degree of Freedom	Mean Square	Mean Square Estimate*
Between subjects	20	2888.8	$\sigma_e^2 + 2\sigma_s^2$
Between scores within subject	21	433.2	σ_e^2

* σ_s^2 = variance attributable to difference between subjects.

σ_e^2 = variance attributable to scoring error.

$\sigma_s^2 = 2888.8 - 433.2/2 = 1227.8$

$\sigma_e^2 = 433.2$

$$\text{Reliability} = \frac{\sigma_s^2}{\sigma_s^2 + \sigma_e^2} = 0.74 = 74\%$$

$$\text{Scoring error} \quad \frac{\sigma_e^2}{\sigma_s^2 + \sigma_e^2} = 0.26 = 26\%$$

Table VIII

Blood Sampling Results for Plasma Ascorbic Acid, Plasma Calcium, and Hemoglobin at the Two Testing Periods, 23 Weeks Apart.

Beginning of Study	Ascorbic Acid (mg/100 ml)	Calcium (mg/100 ml)	Hemoblogin (gm)
Group I average	1.2	10.8	13.6
Group II Average	0.9	10.7	13.5
23 Weeks Later			
Group I average	0.8	10.2	13.7
Group II average	1.1	10.1	13.6

DISCUSSION

Table I illustrates a highly significant increase in the gingival scores from the first to the second examination. In close relation to this, it can be noted in Table II that there was an increase in plaque scores. Several explanations may be given for these two findings. First an increased interest in home dental care may have occurred as the project was initiated. Then during the 23 weeks of the study, a general relaxing of oral hygiene habits may have resulted. Another possibility could be a seasonal change from summer to winter. Changes like this do occur in some blood components, such as ascorbic acid.[49] Table III shows that the two groups did not differ significantly through the first examination on plaque scores. However, at the second examination Group I (control group) had a significant rise in plaque scores, while Group II had only a slight rise, with the only apparent difference between the two groups being the consumption of the oranges.

Studies by Knighton,[51] Clark and co-workers,[52] and Berdon and associates,[53] have not supported the effective cleansing ability of natural foods, especially fibrous foods. In the present study any cleansing effect or plaque removal by the oranges has been disregarded as a reason for less plaque accumulation in Group II as compared to Group I.

Table IV shows a highly significant decrease in transformed calculus scores. This appears to be directly attributable to the prophylaxis after the first examination. With a sufficient length of time, the calculus scores would probably have risen to the levels found at the first examination.

Table V indicates, that during the test period there was no significant rise in decayed, missing, or filled surfaces of teeth in either group. Over a period of 23 weeks, under normal conditions, it is doubtful that there would be a sharp increase in caries rate.

Normally a slight rise in the DMFS score would be expected, but when dealing with children during the time of exchanging primary to permanent dentitions, this may not hold true in every case. Also during this period of exchanging dentitions, it is possible to find a sharp increase in the DMFS if the primary teeth are missing and no permanent successor has erupted. Therefore an analysis of decayed, missing, and filled surfaces may be misleading when dealing with children ages six through 14 years. It would be very difficult to determine accurately when a primary tooth is missing due to caries or to normal exfoliation.

Another factor affecting DMFS is any restorative treatment performed. If a mesial or distal surface was diagnosed as carious, the restoration would have to cover an additional surface (occlusal, labial, or lingual) for access. The study group was accessible to restorative care, but the number of children receiving restorations was not computed.

It appears that the addition of the oranges to the diet of Group II had little or no effect of the caries susceptibility of the teeth during the 23-week test period. Table VI supports this finding by demonstrating no significant changes were present in the number of white spots of the teeth in either group. It is therefore assumed that the oranges added to the diet caused no significant rise in the etching or decalcification of the teeth which would cause white spots to appear.

Partial explanation for an examiner reliability of 74% is the type of examination used. The range of values for the gingival score was small (0 - 4). Also involved with this type of examination are judgement decisions that are bound to produce variations which will again affect reliability. Color changes, inflammation, texture and swelling are very difficult to assess and duplicate. Also a three-week period elapsed between the original gingival scoring and the retest for reliability. There may have been changes in gingival conditions during this period which could affect the reliability.

Another observation made through the 23-week study period was that members of Group II became increasingly dissatisfied with eating three oranges per day. A few minor attempts to avoid eating the oranges were made by the children, but most of these were thwarted. Overall, satisfactory cooperation was obtained from the entire study sample.

The diet analysis demonstrated that both groups were served foods containing more than the estimated average amount of nutrients needed by children. The oranges supplied additional nutrients, but only ascorbic acid totals were changed to any great degree. Group II received more than double the amount of ascorbic acid of Group I on a weekly basis due to the consumption of the oranges.

The results of this study should be qualified and not directly extrapolated to a non-institutionalized population. The entire study sample had access to a nutritious, balanced diet. Also there was supervision at all means. In all probability the diet consumed by these children was better nutritionally than that of the average child in the United States.

This study should be repeated and be conducted for a longer period, if a cooperative group of subjects could be obtained. It would also be desirable to use a more exacting method of determining gingival status in future studies. There appears to be a lack of refinement in most of the scales presently used to measure gingival status. It is further suggested that a method of controlling or monitoring home-care be performed to aid in eliminating a variable of the study. This study could also be conducted using individuals suffering from inherent body chemical imbalances.

On the basis of this study, using a restricted and semi-controlled group of children receiving a balanced diet, it appears that the additional oranges in the diet had a slowing effect on plaque build-up, but otherwise, had little effect on their oral health over a 23-week test period.

SUMMARY AND CONCLUSIONS

There has been much debate over the effects on the periodontium of providing additional fresh citrus fruit or ascorbic acid in the diet. Very few controlled clinical studies have been conducted using fresh citrus fruit over any substantial period of time.

Three hundred children, ages six through 20 years, were clinically examined, bite-wing radiographs were taken, and the children were scored for gingival status, plaque formation, decayed, missing, or filled surfaces of teeth, and white spots on teeth. Gingival status was determined using the PMA Index criteria. Plaque formation was assessed, after disclosing the teeth, using the Quigley and Hein Plaque scoring system.

The entire study sample was then divided equally into two groups according to age, sex, gingival status, and plaque scores. Both groups were given a thorough prophylaxis to obtain a baseline for measuring changes over the 23-week test period. A nutritional analysis was performed on the diet served to the children over a 46-week period. In support of the diet analysis, blood samples were taken on twenty children from each group to measure plasma calcium, plasma ascorbic acid and hemoglobin.

Group I continued on their normal diet with a substitution for any oranges that might occur in their diet with another non-citrus fruit or vegetable. Group II received the same diet but also had one sliced fresh #113 orange added to each meal.

After 23 weeks, 246 of the original 300 children were re-examined using the same criteria as in the original examination. Gingival scores increased significantly in both groups over the test period indicating a worsening of the periodontal condition. The mean gingival score of Group I increased from 1.69 to 1.85, while in Group II the mean score increased from 1.66 to 1.87. Plaque scores also increased in both groups, but Group I had a significant plaque score increase from 2.69 to 2.89 over the test period, and a significant increase over Group II, which also experienced a slight increase in plaque scores from 2.65 to 2.70 during the same period.

Calculus scores decreased significantly in both groups during the observation period. In Group I the decrease was from 4.97 to 1.91 and in Group II the decrease was from 4.66 to 2.24. This observation is attributable to the prophylaxis given after the first examination. There were no significant changes in the number of decayed, missing, or filled tooth surfaces, or white spots of the teeth of either group.

The diet analysis demonstrated that both groups were exposed to a nutritionally well balanced diet. With the addition of the oranges to the diet, Group II received more than double the amount of ascorbic acid than available to Group I.

The blood analyses demonstrated slight fluctuation in values and over the 23-week test period Group I averages had a slight decrease in plasma ascorbic acid (.4mg/100 ml), while Group II averages had a slight increase (.2 mg/100 ml). The other two blood component averages changed in the same direction in identical amounts (plasma calcium decreased and hemoglobin increased).

Further study in this area is indicated using a more reliable and exacting index for measuring gingival status and an improved method of measuring DMFS during the change-over of primary to permanent dentitions.

This study indicates that additional citrus fruit (oranges) had no significant effect in decreasing gingival inflammation or plaque formation in children over the 23-week test period. However, additional oranges added to a nutritionally balanced diet did appear to keep the rise in plaque formation scores to a much lower level than the control group who had a significant increase in their plaque formation scores.

REFERENCES

1. Kyhos, E.D.: Minimum ascorbic acid need of adults. J Nutr 27:271, 1949.

2. Cohen, M.M.: The effect of large doses of ascorbic acid on gingival tissues at puberty. J Dent Res 34:750, 1955.

3. Shannon, I., and Gibson, W.: Ascorbic acid levels in blood and urine as related to oral health status. (S.A.M.-T.D.R. 63-79). USAF School of Aerospace Medicine, Aerospace Medical Center, Brooks Air Force Base, Texas, 1963.

4. Perlitsh, M.J., Nielsen, A.G., and Standmeyer, W.R.: Ascorbic acid plasma levels in gingival health in personnel wintering over Antartica. J Dent Res 40:789, 1961.

5. Mead, S.V.: Studies of the effect of ingestion of citrus fruit upon gingival hemorrhage. J Dent Res 23:73, 1944.

6. Thomas, A.E.: Some observations on the influence of orange juice ingestion on the teeth and supporting structures. Oral Surg 7:741, 1954.

7. Hanke, M.T.: Diet and Dental Health. Chicago, Univ. of Chicago Press, 1933.

8. El-Ashiry, G.M., Ringsdorf, W.M., and Cheraskin, E.: Local and systemic influences on periodontal disease. II. Effect of prophy and natural verses synthetic vitamin C upon gingivitis. J of Periodontol 35:250, 1964.

9. Wolbach, S.E., and Howe, P.R.: The effect of the scorbutic state upon the production and maintenance of intercellular substances. Dent Cosmos 67:1131, 1925.

10. Church, C.F., and Church, H.N.: Food Values of Portions Commonly Used. 11th ed., Philadelphia and Toronto, J.P. Lippincott Co., 1969.

11. Blockley, C.H., and Baenziger, P.E.: An investigation into the connection between the vitamin C content of blood and periodontal disturbances. Brit Dent J 73:57, 1942.

12. Burrill, D.Y.: Relationship of blood plasma vitamin C level to gingival and periodontal disease. J Dent Res 21:353, 1942.

13. Hard, M., Esselbaugh, N., and Donald, E.: Ascorbic acid nutrition assessed by serum level and subclinical symptoms in relation to daily intake. Am J Clin Nutr 6:401, 1958.

14. Linghorne, W.J., McIntosh, W.G., Tice, J.W., Tisdell, F.F., McCreary, J.F., Drake, T.G., Greaves, A.V., Johnstone, W.M.: The relation of ascorbic acid intake to gingivitis. J Can Dent Assoc 12:49, 1946.

15. Weisberger, D., Young, A.P., and Morse, F.W.: Study of ascorbic acid blood levels in dental patients. J Dent Res 17:101, 1938.

16. O'Leary, T.J., Gibson, W., and Shannon, C.F.: A screening examination for detection of gingival and periodontal breakdown and local irritants. (S.A.M.-T.D.R. 51-63). USAF School of Aerospace Medicine, Aerospace Medical Center, Brooks Air Force Base, Texas, 1963.

17. King, J.D.: Nutritional and other factors in "trench mouth" with special reference to nicotinic acid component of vitamin B_2 complex. Brit Dent J 74:113, 1943.

18. Ungley, C.C., and Horton, J.S.F.: Bleeding gums in Naval personnel, vitamin C and nicotinic acid intake. Lancet 1: 397, 1943.

19. Retarski, J.S., and Pijoan, M.: Gingivitis and vitamin C. J Am Dent Assoc 31:1323, 1944.

20. Parfitt, G.J., Speirs, D.M.: Role of nutrition in the prevention and treatment of periodontal disease. J Can Dent Assoc 36:224, 1970.

21. Grandison, W.B., Scott, L.B., and Cruickshank, D.B.: An investigation into the influence of synthetic vitamin C as a controlling factor in incidence of dental caries in already calcified teeth. Brit Dent J 72:237, 1942.

22. Todhunter, E.N., Robbins, R.C., Ivey, G., and Brewer, W.: A comparison of the utilization by guinea pigs of equivalent amounts of ascorbic acid (vitamin C) in lemon juice and the crystalline form. J Nutr 19:113, 1940.

23. Hicks, H.: Excessive citrus juice consumption: II. Clinical observation of its effects on superficial and deep tissues of the oral cavity. Oral Surg 4:858, 1951.

24. Massler, M., Cohen, A., and Schour, I.: Epidemiology of gingivitis in children. J Dent Res 31:496, 1953.

25. Benjamin, E.M., Russell, A.L., and Smiley, R.D.: Periodontal disease in rural children of 25 Indiana counties. J of Periodontol 28:294, 1957.

26. Moore, R.M.: A study of the effect of water fluoride content and socioeconomic status on the occurrence of gingivitis in school children. Thesis, I.U. School of Dentistry, 1963.

27. Bell, Carl: Personal communication, 1972.

28. James, P.M.C., Jackson, D., Slack, G.L., and Lawton, F.E.: Gingival health and dental cleanliness in English School children. Arch Oral Biol 3:57, 1961.

29. Carter, W.J., and Wells, J.E.: Epidemiology of gingival disease in Kansas City, Missouri school children. Midwestern Dent 36:21, 1960.

30. King, J.D.: Gingival disease in Dundee. Dent Record 65: 9, 1945.

31. Jameson, H.C.: Prevalence of periodontal disease of deciduous teeth. J Am Dent Assoc 66:207, 1963.

32. Miller, S.L., and Chrietzberg, J.E.: Caries experience among children in two fluoridated communities where the fluoride concentration was adequately maintained in only one. J Public Health Dent 22:61, 1962.

33. Wachowski, C.S.: County Dental Survey Results. Arizona Dent J 8:16, 1962.

34. Brucker, M.: Studies on the incidence and cause of dental defects in children. J Dent Res 29:148, 1950.

35. Easlick, E.: Dental caries, mechanism and present control techniques - a workshop. St. Louis, C.V. Mosby Co., 1948.

36. Shafer, W., Hine, M., and Levy, B.: A Textbook of Oral Pathology. 2nd ed., Philadelphia and London, Saunders Co., 1968, p. 313.

37. Graf, J.: The glycolytic activity of plaque and its relation to hard tissue pathology. International Conference on Dental Plaque. Morris Plains, N.J., Warner Lambert Pharmaceutical Co., 1969.

38. Shafer, W., et al., Op. Cit. p. 651.

39. Ash, M.M., Giltin, B.N., and Smith, W.A.: Correlation between plaque and gingivitis. J Periodontol 35:424, 1964.

40. Loe, H.: The formation and growth of dental plaque on gingival-dental structures. International Conference on Dental Plaque, Morris Plains, N.J., Warner Lambert Pharmaceutical Co., 1969.

41. Eichel, R.A.: A clinical television evaluation of plaque formation in children. Thesis, I.U. School of Dentistry, 1969.

42. Quigley, G.A., and Hein, J.W.: Comparative cleansing efficiency of manual and power brushing. J Am Dent Assoc 65:26, 1962.

43. Frueger, C.T., Cohen, P.J., and Foster, L.B.: Automated method for determination of serum calcium by use of Alizar. Clin Chem 16:816, 1970.

44. Marsh, H.H.: A modification of the Kleth-Summerson colorimeter. Am J Clin Path 37:115, 1962.

45. Brittin, G.M., Brecher, G., and Johnson, C.A.: Evaluation of the Coulter Counter Model 5. Am J Clin Path 52:679, 1969.

46. Freeman, M.F., and Tukey, J.W.: Transformations related to the angular and the square root. Annals Math Stat 21: 607, 1950.

47. Wever, B.J.: Statistical Principles in Experimental Design. New York, McGraw-Hill, 1962, p. 302.

48. Tietz, N.W.: Fundamentals of Clinical Chemistry. Philadelphia and London, W.B. Saunders Co., 1970, p. 176.

49. Diem, K.: Documenta Giegy Scientific Tables. Ardsley, N.Y., Geigy Pharmaceuticals, 1962, p. 568.

50. Diem, K.: Documenta Giegy Scientific Tables. Ardsley, N.Y., Geigy Pharmaceuticals, 1962, p. 578.

51. Knighton, H.T.: Effect of various foods and cleansing agents on elimination of artificially inoculated yeast from the mouth. J Am Dent Assoc 29:2012, 1942.

52. Clark, J.W., Cheraskin, E., and Ringsdorf, W.: Diet and the Periodontal Patient. Springfield, Ill., Charles Thomas Co., 1970, p. 383.

53. Berdon, J.K., and Hammer, J.W.: The cleansing effect of selected foods on human dentition. Va Dent J 47:30, 1970.

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ABSTRACT

The Effects of a Dietary Supplement of Fresh Oranges on the Oral Health of Children

By

Gary J. Dilley

The effects of additional citrus fruit in the diet on the periodontium have been a debated subject for some time. This study attempted to measure the effects of eating three additional oranges per day by 123 children ages six through twenty years and an equal number of controls over a 23-week period. To measure any changes that might take place, the following were evaluated clinically, and the decayed, missing and filled surfaces were also evaluated radiographically:

1. gingival status
2. plaque formation
3. D.M.F.S. and d.m.f.s.
4. white spots

Results after the 23 week test period showed that the gingival scores increased significantly in both groups (increased inflammation). The plaque formation score also increased in both groups, but only the non-orange eaters' score increased significantly over their original score and over the orange eaters' score. The decayed, missing, and filled surfaces and white spots did not change significantly in either group.

Therefore with this study sample over the 23-week test period, the additional oranges in the diet had limited measurable effect on the hard and soft tissues of the oral cavity.

ADDENDUM

THE EFFECTS OF A DIETARY SUPPLEMENT OF FRESH ORANGES ON THE ORAL HEALTH OF CHILDREN

By

Gary J. Dilley

The possible effect on oral health of adding citrus fruit to the diet has been a controversial topic. This study of children ages 6 through 20 attempted to measure the effects of eating three oranges per day over a 44-week period. The children were residents of the Soldiers and Sailors Children's Home of Knightstown, Indiana. The original study of 23 weeks was continued for an additional 21 weeks to determine if any further changes or trends would appear or continue in either Group I (the non-orange eaters) or Group II (the orange eaters). To measure any possible changes, the following were evaluated clinically, and the decayed, missing and filled surfaces were also evaluated radiographically:

1. gingival status
2. plaque formation
3. calculus accumulation
4. D.M.F.S. and d.m.f.s.
5. white spots

After the 44-week test period, both groups had a significant increase in gingival scores as represented by increased inflammation (Table IX). The orange eaters had a smaller increase in inflammation than the non-orange eaters; however, the difference was not statistically significant. Plaque scores fluctuated but at the end were nearly the same as at the beginning of the study. Again the orange eaters had less (non-significant) plaque formation (Table XI). Similarly, no significant difference was found between groups on calculus scores (Table XIII). The decayed, missing or filled surfaces for both groups increased significantly, as did the frequency variable for white spots (Tables XV and XVII). In this study, the addition of oranges to an already well balanced diet had limited beneficial effects on the variables measured.

The third and final re-examination was performed on the entire remaining sample of 206 children (103 from each group) 44 weeks after the experimental group had begun consumption of the oranges (July 5, 1972 to May 8, 1973). The children were re-examined for gingival status, plaque formation, calculus formation, presence of white spots, and decayed, missing or filled surfaces of each tooth, as had been done for the two previous examinations. In addition, 31 of the original 40 children (17 from Group I and 14 from Group II) who had provided blood specimens were resampled for the same hemotologic components (Table XIX). There was again a decrease in the total study sample because children were being returned to their natural parents.

Statistical analyses were repeated on the same five variables. The Analysis of Variance tests were used to measure any differences between the two groups at the third examination. This test also measured any differences between the three examinations and any group-by-examination interaction. If there were any significant differences between the three examinations, the Newman-Keul Sequential Range Test* was used to determine where the differences occurred.

*Newman-Keul Sequential Range Test: Statistical analysis submitted to the Research Computation Center of Indiana University-Purdue University, Indianapolis, Indiana.

RESULTS

Gingival Scores: The analysis of variance in Table IX shows a highly significant difference ($F = 45.52$ when $F_{2,204} = 4.71$ at the .01 level of confidence) between the three examinations. There were no significant differences between the two groups or in the group-by-examination interaction.

The differences between the gingival scores for the three examinations were further tested by the Newman-Keul Sequential Range Test (Table X). For both groups, gingival scores for examination two were significantly higher than for examination one and scores for examination three were significantly higher than those for both examinations two and one.

Plaque Scores: The analysis of variance in Table XI shows a highly significant difference ($F = 13.31$ when $F_{2,204} = 4.71$ at the .01 level of confidence) between the three examinations. No other significant differences in scores were noted either between groups or in group-by-examination interaction. The Newman-Keul test (Table XII) showed that for both groups the plaque scores for examination three did not differ from those for examination one, but the scores for examination two were significantly higher than those for both examinations one and three.

Transformed Calculus Scores: The analysis of variance in Table XIII shows that there was a highly significant difference ($F = 28.80$ when $F_{2,204} = 4.71$ at the .01 level of confidence) between the calculus scores for the three examinations. Sources of variation between groups and group-by-examination interaction were not significant. The Newman-Keul test (Table XIV) demonstrated that the scores for examination three did not differ significantly from those for examination one, but that the calculus scores for examination two were significantly lower than those for examinations one and three for both groups.

Transformed, Decayed, Missing and Filled Surfaces: The analysis of variance in Table XV shows that there was a significant difference between the scores for the three examinations ($F = 3.43$ when $F_{2,204} = 3.04$ at the .05 level of confidence). No significant differences were found either between groups or group-by-examination interaction. The Newman-Keul test (Table XVI) demonstrated that the scores for examination two did not differ significantly from those for examination one or three, but the scores for examination three differed significantly from those for examination one. Examination three had the highest mean score.

Transformed White Spots: The analysis of variance in Table XVII shows a significant difference between the scores for the three examinations ($F = 3.98$ when $F_{2,204} = 3.04$ at the .05 level of confidence). No significant differences were found between the two groups or in the group-by-examination interaction. The Newman-Keul test (Table XVIII) further showed that the white spot scores for examination three were significantly higher than those for both examinations one and two.

Blood Analysis Results: Table XIX demonstrates that at the end of the 44-week test period, the average blood values continued to be very similar between the two groups and between the examination periods. The plasma ascorbic acid levels were stabilized at 1.1 mg per 100 ml for both groups. The average level in Group II remained the same as during the 23-week sampling period. The average in Group II increased slightly from that of the 23-week sampling period. The plasma calcium levels returned to the approximate levels that were measured at the beginning of the study. The new calcium averages were 10.9 mg per 100 ml and 10.8 mg per 100 ml for Groups I and II, respectively. The hemoglobin concentration continued to rise slightly, with the new averages being 14.1 grams and 13.9 grams for Groups I and II, respectively.

Since menus were rotated and the same food served every 26 weeks, further diet analysis was not required.

TABLES

Table IX

Analysis of Variance for Gingival Scores of Group I (Non-Orange Eaters) and Group II (Orange Eaters) Between Examination 1 (Before Oranges Were Added to the Diet), Examination 2 (23 Weeks After the Addition of Oranges), and Examination 3 (44 Weeks after the Addition of Oranges)

Source of Variation	Degrees of Freedom	Mean Square	F	
Between groups	1	0.067	0.20	N.S.
Subjects in groups	204	0.333		
Between examinations	2	4.916	45.52	p<.01
Group x examination interaction	2	0.195	1.81	N.S.
Subject x examination in groups	408	0.108		
Mean Scores				
Examination	1	2	3	
Group I	1.68	1.83	2.02	
Group II	1.66	1.87	1.94	

Table X

Neuman-Keul Sequential Range Test of Comparisons between Examinations 1, 2 and 3 for Gingival Mean Scores Over Groups I and II

Examination	1	2	3
Mean	1.67	1.85	1.98

The examination scores for each group were significantly different at the .05 level.

Table XI

Analysis of Variance for Plaque Scores of Group I (non-Orange Eaters) and Group II (Orange Eaters) Between Examination 1 (Before the Oranges Were Added to the Diet), Examination 2 (23 Weeks After the Addition of Oranges), and Examination 3 (44 Weeks After the Addition of Oranges)

Source of Variation	Degrees of Freedom	Mean Square	F	
Between groups	1	1.696	3.27	N.S.
Subjects in groups	204	0.518		
Between examinations	2	1.571	13.31	$p < .01$
Group x examination interaction	2	0.187	1.58	N.S.
Subject x examination in groups	408	0.118		

Mean Scores

Examination	1	2	3
Group I	2.67	2.88	2.71
Group II	2.63	2.72	2.60

Table XII

Neuman-Keul Sequential Range Test of Comparisons Between Examinations 1, 2 and 3 for Plaque Mean Scores Over Groups I and II

Examination	1	3	2
Mean	2.65	2.65	2.80

Underlined mean scores are not significantly different. The scores for examination 2 are significantly higher than the scores for examinations 1 and 3 at the .05 level.

Table XIII

Analysis of Variance for Transformed Calculus Scores of Group I (Non-Orange Eaters) and Group II (Orange Eaters) Between Examination 1 (Before the Oranges were Added to the Diet), Examination 2 (23 Weeks After Addition of Oranges), and Examination 3 (44 Weeks After the Addition of Oranges)

Source of Variation	Degrees of Freedom	Mean Square	F	
Between groups	1	540.6	0.03	N.S.
Subjects in groups	204	15726.9		
Between examinations	2	173432.9	28.80	p<.01
Group x examination interaction	2	1608.6	0.27	N.S.
Subject x examination in groups	408	6020.9		

Mean Scores

Examination	1	2	3
Group I	97.34	47.86	106.49
Group II	99.51	55.17	102.62

Table XIV

Neuman-Keul Sequential Range Test of Comparisons Between Examinations 1, 2 and 3 for Transformed Calculus Mean Scores Over Groups I and II

Examination	2	1	3
Mean	51.51	98.43	104.55

Underlined mean scores are not significantly different. The scores of examination 2 are significantly lower than the scores of examinations 1 and 3 at the .05 level.

Table XV

Analysis of Variance for Transformed Decayed, Missing, or Filled Surfaces Scores of Group I (Non-Orange Eaters) and Group II (Orange Eaters) Between Examination 1 (Before the Oranges were Added to the Diet), Examination 2 (23 Weeks After Addition of Oranges), and Examination 3 (44 Weeks After the Addition of Oranges)

Source of Variation	Degrees of Freedom	Mean Square	F	
Between groups	1	650.4	0.05	N.S.
Subjects in groups	204	13132.4		
Between examinations	2	3451.7	3.43	$p<.05$
Group x examination interaction	2	133.0	0.13	N.S.
Subject x examination in groups	408	1005.4		
Mean Scores				
Examination	1	2	3	
Group I	192.87	197.84	199.18	
Group II	189.22	195.78	198.75	

Table XVI

Neuman-Keul Sequential Range Test of Comparison Between Examinations 1, 2, and 3 for Transformed Decayed, Missing or Filled Surface Mean Scores Over Groups I and II

Examination	1	2	3
Mean	191.05	196.81	198.97

Underlined scores are not significantly different. Scores for examination 3 were significantly higher than the scores for examination 1 at the .05 level.

Table XVII

Analysis of Variance for Transformed White Spot Scores of Group I (Non-Orange Eaters) and Group II (Orange Eaters) Between Examinations 1 (Before Oranges Were Added to the Diet), Examination 2 (23 Weeks After the Addition of Oranges), and Examination 3 (44 Weeks After the Addition of Oranges)

Source of Variation	Degree of Freedom	Mean Square	F	
Between groups	1	365.1	0.15	N.S.
Subjects in groups	204	2514.0		
Between examinations	2	2905.0	3.98	$p < .05$
Group x examination interaction	2	238.6	0.33	
Subject x examination in groups	8	729.0		
Mean Scores				
Examination	1	2	3	
Group I	12.84	11.47	17.99	
Group II	11.93	14.59	20.39	

Table XVIII

Neuman-Keul Sequential Range Test of Comparisons Between Examinations 1, 2, and 3 for Transformed White Spot Mean Scores Over Groups I and II

Examination	1	2	3
Mean	12.39	13.03	19.19

Underlined mean scores are not significantly different. The scores for examination 3 were significantly higher than the scores for examinations 1 and 2 at the .05 level.

Table XIX

Blood Sampling Results for Plasma Ascorbic Acid, Plasma Calcium and Hemoglobin at the Three test times: 1 (Before Oranges were Added to the Diet), 2 (23 Weeks After Addition of Oranges to the Diet of Group II), and 3 (44 Weeks After Addition of Oranges to the Diet of Group II)

	Ascorbic Acid (mg/100 ml)	Calcium (mg/100 ml)	Hemoglobin (gm)
Beginning of Study			
Group I	1.2	10.8	13.6
Group II	0.9	10.7	13.5
23 Weeks			
Group I	0.8	10.2	13.7
Group II	1.1	10.1	13.6
44 Weeks			
Group I	1.1	10.9	14.1
Group II	1.1	10.8	13.9

DISCUSSION

Table IX demonstrates a statistically significant increase in gingival scores over the three examinations. The rates of change for the two groups varied, but these can probably be accounted for as normal fluctuations within group variations as they were fairly equal. This lack of differences between the two groups may be accounted for by the fact that both groups received nutritious and balanced diets, or by the possibility that, given a nutritious and balanced diet, ascorbic acid does not provide measurable beneficial effects. Correlating with this observation, the blood samples demonstrated nearly identical ascorbic acid levels at the end of the 44-week testing period. One other possible consideration might be that Group II did not regularly eat their ration of oranges. However, the good cooperation of the children and supervisors makes this explanation unlikely.

Table XI illustrates that the plaque formation averages for both groups at the first and third examinations were nearly identical and that the averages over Groups I and II were identical to two decimal places, as shown in Table XII. It must be pointed out that the orange-eating group tended to lag behind the non-orange eaters in their plaque accumulation averages. This might be accounted for by the citrus fruit consumption. Group averages for plaque accumulation were consistently lower in Group II after the orange consumption started but were not statistically significant over the test period.

Table XIII shows a predictable pattern of calculus redeposition from the time of the original prophylaxis, after examination one to the end of the test period. The rates of calculus accumulation for the two groups were slightly different; however, these fluctuations could be normal group variations.

Table XV shows the trend for an increase in D.M.F.S. This increase was not significant from examination one to examination two or from examination two to three, but there was a statistical difference between the first and third examinations. The steady rise in D.M.F.S. can be expected in this age group of children.

Table XVII shows a significant increase for both groups in transformed white spot scores at the third examination. This could be explained by further eruption of permanent teeth with white areas becoming visible. Since both groups experienced an increase in detected white spots, the citrus fruit consumption probably is not responsible for this increase.

Table XIX shows slight fluctuations in the representative blood samples taken at the end of the study. The mean plasma ascorbic acid level of Group II (1.1 mg/100 ml) remained the same as the 23-week sample, which had increased from the original sampling of 0.9 mg/100 ml. The plasma ascorbic acid level of Group I (1.1 mg/100 ml) fluctuated to above the 23-week sampling but never returned to the original level of 1.2 mg/100 ml. It could be that the effect of additional oranges in the diet of Group II stabilized the plasma ascorbic acid level, and there may be a saturation level which prevents utilization of all the ascorbic acid from citric fruit.

The plasma calcium levels of both groups dropped at the 23-week examination but returned to their original levels at the 44-week examination. The hemoglobin levels of both groups increased slightly at each examination. All three of the blood component averages remained well within the normal ranges for children of this age group. The blood sample analyses were completed for only a representative portion of both groups.

Throughout the last 21 weeks of the study there were increasing complaints in Group II about eating the three oranges per day. However, the overall cooperation was good.

From the results of both the 23-week and 44-week testing periods, it appears statistically that the addition of oranges to the diet had limited effect on the oral health of the children of Group II. These results should not be directly correlated to other non-institutionalized groups of children. Both groups had access to a nutritious and well-balanced diet before and after the oranges were added to the diet. This one factor may have had the greatest impact on the lack of differences between the two groups during the test period. Two other factors which may have affected the results are the lack of refinement in measuring gingival conditions and the variable of personal oral hygiene by both groups. It is hoped that a similar but more exacting study can be performed to confirm or invalidate these results.

SUMMARY AND CONCLUSIONS

Opinions differ concerning the possible effects on oral health of adding fresh citrus fruit to the diet of children. This study was instituted because few clinical investigations have attempted to correlate additional fresh citrus fruit consumption and oral health.

Three hundred children, ages six through twenty years, were clinically examined, bite-wing radiographs were taken, and the children were scored for gingival status, plaque formation, calculus accumulation, decayed, missing, or filled surfaces of teeth, and white spots on teeth. Gingival status was determined using the PMA Index criteria. Plaque formation was assessed by the Quigley and Hein Plaque scoring system, after the teeth were disclosed.

The study sample was then divided equally into two groups according to age, sex, gingival status, and plaque scores. Both groups were given a thorough prophylaxis to obtain a baseline for measuring changes over the 44-week test period. A nutritional analysis was performed on the diet served to the children over the test period. In support of the diet analysis, blood samples were taken on twenty children from each group to measure plasma calcium, plasma ascorbic acid and hemoglobin.

Group I continued on their normal diet with a non-citrus fruit or vegetable being substituted for any oranges that might occur in their diet. Group II received the same diet but had one sliced fresh #113 orange added to each meal.

After 23 weeks and 44 weeks the children were re-examined using the same criteria as the original examination. The results at the end of the 44 weeks on the 206 remaining children follow:

Gingival mean scores increased significantly for both groups. Group I had an increase in mean score from 1.68 to 1.83 after 23 weeks, and another increase to 2.02 after 44 weeks. Group II had an increase from 1.66 to 1.87 at the 23 week examination and a further, but lesser increase to 1.94 at the conclusion of the study. Therefore, in both groups there was an increase in the inflammatory condition of the gingiva, but there was no statistical difference between the two groups.

Mean plaque scores fluctuated significantly for both groups but examinations one and three were not statistically different. Group I had fluctuations from 2.67 to 2.88 and back to 2.71 at the 23-week and 44-week examinations, respectively. Group II had slightly less plaque accumulations, from 2.63 to 2.72, and then a reduction to 2.60 at the 44-week examination. No statistical differences were noted between the two groups.

The frequency variable calculus scores showed significance only at the interim examination. This significant decrease in calculus scores (Group I decreased from 97.34 to 47.86 and Group II decreased from 99.51 to 55.17) could be expected because a thorough prophylaxis was given after the first examination. At the final examination the calculus scores returned to the approximate levels found at the beginning of the study. Group I had a score of 106.49, while Group II was slightly lower at 102.62. No significant difference was observed between the two groups.

The frequency variable D.M.F.S. scores showed a constant increase over the 44-week study period. The increases were not significant when measured at intervals, but from the first examination score (191.05) to the third examination score (198.97) there was a significant increase at the .05 level. There was no significant difference between the two groups.

The figures for the frequency variables for white spots showed no significant difference between the groups. Significant differences were noted between examination three (19.19) and both examination one (12.39) and examination two (13.03).

The blood analyses demonstrated slight fluctuations over the 44-week test period. The plasma ascorbic acid levels were stabilized for both groups at 1.1 mg per 100 ml at the end of the 44-week test period. The plasma calcium levels dropped at the 23-week examination, but returned to the original levels at the 44-week examination. Hemoglobin concentrations increased in both groups by nearly identical amounts. All three blood components were within the normal ranges for children.

For future studies on this topic it is recommended that more precise indices be used for measuring gingival inflammation and scoring d.m.f.s. - D.M.F.S. in the mixed dentition stages. It would also be beneficial if some method of controlling or conforming the methods of oral hygiene for the subjects could be instituted.

This 44-week study demonstrated that adding citrus fruit to the diet had no statistically significant effect on decreasing gingival inflammation, plaque accumulations, or calculus deposits. However, there were trends for less rapid increases in these three measurements for the group consuming the oranges, as their average scores were lower at the end of the 44-week study.