THE SCURVY OF GUINEA PIGS.*

II. EXPERIMENTS ON THE EFFECT OF THE ADDITION OF FRUITS AND VEGETABLES TO THE DIETARY.

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In a previous communication (1) we have reported on scurvy brought about in young guinea pigs by a diet of oats, hay, and water ad libitum. As far as the occurrence of this disorder was concerned we noted no difference whether this simple dietary was employed, whether it was supplemented by the addition of cod liver oil to supply fat and fat-soluble vitamine, or whether the white and yolk of egg were added, enriching the diet with water-soluble vitamine, fat-soluble vitamine, fat, and protein. The signs of scurvy appeared regularly at the same period on the simple as on the more complete ration. In the preceding paper the gross pathologic changes noted at autopsy were checked by careful microscopic examinations of the bones, in order to be certain that the lesions were truly scorbutic. This precaution was considered essential in view of our encountering cases which were regarded as scurvy at autopsy but which histologically resembled rickets. A similar control of the pathologic anatomic changes was carried out in the present investigation.

This study concerns itself with the effect of augmenting the dietary with fruits and vegetables, and more particularly with the alteration in the antiscorbutic potency of these foodstuffs occasioned by heating, drying, and aging. This question of the exact food value of dried fruits and vegetables is important from a nutritional point of view, and is of especial significance at this time when dehydrated foods are being employed to an increasingly large extent both among the civil population and in army rations.

* A preliminary report of this work was presented before the Society for Experimental Biology and Medicine, May 15, 1918.
It is therefore highly desirable that we should have as complete knowledge as possible of their food value in the dried state.

As orange juice is the prototype of an antiscorbutic, it seemed highly desirable to investigate whether aging affected to any degree its antiscorbutic value. Accordingly three guinea pigs, which had developed scurvy on the regular ration, were fed 3 cc. daily of orange juice which had been kept in the refrigerator for over 3 months. As will be noted from Chart 1 this addition cured the scurvy in the two pigs which lived long enough to permit us to judge of the result. It seemed, however, to have lost some of its potency as the result of aging, for when fresh orange juice, to only half the amount, was substituted in the case of the dietary of one animal, there was a sharp rise in weight and an improvement in general condition. We may add that a similar clinical result was noted when guinea pigs were given orange juice which had been subject to heat. Animals fed juice which had been autoclaved at a temperature of 110° under 10 to 15 pounds pressure for a period of 45 minutes, did not thrive so well as those which were

![Chart 1](http://www.jbc.org/)
fed the unheated juice. In both experiments, however, the signs of scurvy disappeared.

The active factor in orange juice can be extracted by means of 95 per cent alcohol. Chart 2 illustrates an experiment in which scurvy was cured by adding a small amount of such an extract to the dietary, and where it redeveloped when the residue was substituted for the extract. The extract was obtained as follows: 300 cc. of orange juice were evaporated at room temperature by means of a fan to about 40 cc. To this fluid 800 cc. of 95 per cent alcohol were added. This was allowed to extract at room temperature

**Chart 2.** During Period I all guinea pigs developed scurvy on a diet of hay, oats, and water *ad libitum*. These signs all disappeared during the second period when alcoholic extract of orange juice equivalent to 4 cc. of the juice was given daily. During the third period the residue of this extract was substituted in the same dosage, with the result that scurvy reappeared and there was marked loss of weight. A short fourth period was begun wherein the extract was given in place of the residue. However, the guinea pigs died, although the signs of scurvy were ameliorated.
for 7 days, being shaken frequently every day. It was decanted and once more evaporated down to about 40 cc., and then distilled water was added so that its volume again equalled 300 cc. (that of the original orange juice). The residue, which was of a gummy consistence, was taken up with distilled water so that it also corresponded with the original volume of the fruit juice. The water content of this extract varied somewhat in different preparations, but contained on an average 10 to 12 per cent of water. Its effect both on the scurvy and on the weight of the guinea pigs was striking when compared to the residue (Chart 2).

As it is not possible to define the nature of the antiscorbutic agent, it seems preferable, for the present, to term it "an accessory food factor." That its effect is not that of a mere laxative has been brought out in our previous paper (1) and is fortified by the negative results (which will be published elsewhere) obtained with laxatives in human scurvy. We may add that its remarkable therapeutic effect is not due to the mere salts frequently associated with it. In many instances we have attempted to cure the scurvy of guinea pigs by means of an addition of 1.5 cc. of "artificial orange juice" made according to the formula of McCollum and Pitz (2). This is composed of the various salts, citric acid, and sucrose in the proportions in which they are found in the natural juice. However, in every case it failed to show any curative properties. As we have mentioned in the preliminary communication, this same negative therapeutic effect was met with in the treatment of two cases of infantile scurvy treated for a period of about 3 weeks with appropriate amounts of this preparation. The most reasonable interpretation of the success obtained by others with "artificial orange juice" would seem to be that the guinea pigs which were benefited were suffering from some disorder not comparable with human scurvy.

A few years ago one of us reported the observation that the peel of the orange contains the antiscorbutic factor (3). This was of interest from a practical standpoint as it suggested the use of a waste product in place of or in addition to orange juice in connection with infant feeding. It seemed of interest not only to test further the value of orange peel in this respect, but to ascertain whether it would retain its antiscorbutic virtue in spite of drying (Chart 3). The peels which were used for this test were
kept in a burlap bag in an ordinary closet for a period of 3 months. By this time they had become dry and brittle. Their composition was as follows: total ether extract, 1.77 per cent; non-volatile ether extract at 100°, 1.58; volatile ether extract, 0.10; moisture 10.01 per cent. They were ground up before using, and 0.5 gm. of this substance was fed daily by means of a pipette, after thoroughly moistening with water, to a number of guinea pigs. Although all of these animals developed scurvy,

![Chart 3. Hay, oats, water ad libitum and desiccated orange peel (0.5 gm. daily). This peel was about 3 months old, and had been dried at room temperature. It was powdered each day and given with a pipette. It will be noted that it prolonged the life of the animals markedly. It also greatly delayed the onset of scurvy. After a period of 56 days an equivalent amount of fresh orange peel was substituted. In the case of Guinea Pig B 944 there was a prompt gain of weight, and a disappearance of the scorbutic signs. In the case of the two other animals the scurvy was cured in one (No. B 961) and was improved in the other.]

they did not evince signs of this disorder for 50 days, proving that the peel still retained considerable of its antiscorbutic power. Doubtless had a larger dosage been employed the development of symptoms would have been postponed still further. This result shows that under certain physical or chemical conditions, which deserve closer study, the accessory food factor is not destroyed for a considerable period.

The effect of an addition of prunes to the oats, hay, and water dietary was tested. This fruit was singled out as it is used so
extensively in infant feeding, and it is, therefore, important for physicians to know whether it can replace the orange juice, which is commonly prescribed when raw milk is not fed. The prunes were soaked in a small amount of water, crushed in a mortar, and then fed. Chart 4 shows the failure of this foodstuff which is seen to have retained almost none of its antiscorbutic power. It is interesting to compare the result with that obtained with orange peel, of which only about 5 per cent of the quantity was given.

The second part of this study was designed to test the efficacy of dehydrating vegetables. Some years ago Holst and Frolich (4) showed by experiment that dried vegetables had lost their antiscorbutic power. This had been the sad experience in our army during the civil war (5) and in other armies previous to this time. The experiments of Holst and his coworkers have been repeated and amplified recently at the Lister Institute by Chick and Hume (6), who came to the same conclusions. Nevertheless there is widespread propaganda claiming that "dehydrated vegetables and fruits differ from fresh vegetables in only one respect, namely, in that they have had the moisture taken out" and again that "the trouble is that we like our vegetables fresh." We learn furthermore that in all probability in the near future
dehydrated vegetables will become a staple in our army rations. In view of these various activities, the subject assumes added importance at the present time.

The very fact that guinea pigs promptly develop scurvy on a diet which includes an unlimited supply of hay, in other words sun-dried grass, is strong experimental evidence against the retention of antiscorbutic potency after drying.

Charts 5, 6, and 7 portray the results of the addition to the regular dietary of three different types of dehydrated vegetables. In the first test dehydrated carrots, bought in the open market, were fed. In all these experiments the vegetable was mixed with as much water as it would absorb, precaution being observed not to add a surplus and thus wash out any of the salts. For the vegetables made use of in the two other experiments, a grinder was employed so that they would more readily take up moisture. It was found that this procedure rendered them more palatable to the animals. In all cases careful note was made of the amount of vegetable consumed. In general it may be stated that the carrots employed in Experiment 6 were not well taken, but that in Nos. 5 and 7 they were consumed in the amounts provided, excepting towards the end when the animals were weak and sick. The "commercial" dehydrated carrots possessed practically no scurvy-protecting power. It is not possible to state the age of this
preparation, or the degree of heat to which it had been subjected. The second lot tested (Chart 6) had been prepared 7 months previously by means of drying at room temperature with the aid of electric fans. It had meanwhile been carefully preserved in waxed cartons. These carrots likewise failed to ward off scurvy. When, however, fresh carrots were substituted all scurbutic signs rapidly disappeared and there was a prompt change in the general condition. The last experiment of this series (Chart 7) was the most conclusive as the carrots were excellent in appearance and flavor. They had been dehydrated about 3 to 5 weeks before the test, and meanwhile kept in well sealed paraffined paper bags. They were unable, however, to prevent or even notably to defer the onset of scurvy, and showed a sharp contrast in this respect to the potency of the fresh carrots which replaced them.

These experiments are not to be interpreted as showing that dehydrated vegetables are not a valuable foodstuff. On the contrary, in view of the fact that they are capable of furnishing a large amount of nutriment to the diet and their small bulk and ready preservation, they deserve commendation. They are not, how-

1 These vegetables were furnished by the Mrs. Oliver Harriman Food Research Laboratory of New York.
ever, the equivalent of the fresh vegetables, and unless this dis-
tinction is clearly realized it will result in nutritional disorder. It is quite possible that the process of dehydration and of sub-
sequent storing can be so improved as to obviate the loss of this food
accessory. The method includes a number of factors, among
which the degree of heat employed in dehydration seems less im-
portant than the rapidity of the process. The temperature and
state of moisture at which the vegetables are stored have also an

![Chart 7](http://www.jbc.org/)

**Chart 7.** Hay, oats, water *ad libitum,* and 7 gm. of dehydrated carrots
dried at a room temperature of about 130°F. about 1 month previously.
All animals developed scurvy. The marked gain in weight when an equiva-
lent amount of fresh carrots was substituted for the dehydrated vegetable
corresponded to the disappearance of scorbutic signs. Guinea Pig B 951
did not receive fresh carrots, as it was evidently ailing at the time the sec-
ond period began.

important bearing on their deterioration. Finally, it is probable
that certain vegetables will be found to withstand dehydrating
more successfully than others. It would seem that the onion and
the cabbage possess the highest degree of antiscorbutic power, so
that they might be expected to retain the greatest measure of this
quality after proper drying and storing.

In conclusion we may add that these experiments with guinea pig
scurvy coincide with our experience in the treatment of infantile
scurvy. Indeed, unless the results of animal investigation can
be brought into harmony with clinical observation—particularly in regard to the efficacy of therapeutic agents—they must be looked upon as doubtful and inconclusive.

SUMMARY.

Orange juice which had been allowed to age for some months in the refrigerator was found to have lost some of its antiscorbutic power. This factor may be extracted from orange juice by means of 95 per cent alcohol, and is entirely absent in the residue.

“Artificial orange juice” prepared according to the formula of McCollum and Pitz failed to protect guinea pigs from or to cure them of scurvy. It was also found to lack antiscorbutic power in infantile scurvy.

Orange peel possesses marked antiscorbutic potency, and withstands desiccation remarkably well, retaining considerable of this power after being dried for 3 months. Prunes possess practically no value as a preventive against scurvy.

Dehydrated vegetables were found to contain little or no antiscorbutic virtue. This experience coincides with that encountered in relation to human scurvy. As this food is of decided nutritive value efforts should be instituted to improve the methods of dehydrating and of storing so as to obviate this deficiency.

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