XXXI. THE ANTISCORBUTIC AND ANTIBERI-BERI PROPERTIES OF CERTAIN SUN-DRIED VEGETABLES.

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THE present investigation is a continuation of work undertaken at the request of Sir Leonard Rogers to determine the antiscorbutic and antiberi-beri properties of certain sun-dried vegetables prepared at the Government Fruit Experiment Station, Quetta [Shorten and Ray, 1919]. This paper dealt with four varieties of vegetables, viz. carrots, spinach, onions, and cabbage. The period of experimental feeding—42 days—was afterwards considered too short. There were also certain accidental irregularities in the feeding which made it desirable to repeat the experiments.

As stated in our original paper [1919], the specimens of vegetables experimented with were prepared under the direction of Mrs G. L. C. Howard, Second Imperial Economic Botanist to the Government of India. A full description of the methods employed is to be found in Bulletin No. 8 of the Fruit Experiment Station, Quetta, dated March 1918. A short description of the procedure adopted may here be quoted from our original paper.

“The vegetables having been cleaned and prepared are placed in fresh water, or in certain cases in a steeping solution. The prepared material is then transferred to a wire basket and either scalded or steamed for a few minutes. The contents of the wire basket are now emptied on to the drying trays and dried in the sun. The dried product is collected in sacks or open boxes in which it is allowed to remain for about a week, during which time it is examined at intervals for signs of dampness. After about a week the dried product is ready to be compressed into bricks and packed in airtight tins.”

The present investigation comprises experiments with nine varieties of vegetable, viz. carrots, brinjal, spinach, cabbage, tomatoes, potatoes, turnips, turnip-tops and mixed factory-dried vegetables prepared by Messrs Swallow and Ariel, Port Melbourne, Australia.

In the case of the sun-dried vegetables the samples were from three to six months old. The age of the mixed factory-dried product could not be ascertained. All the samples were in good condition when opened. On opening the tins they were made up in 5 g. packets and stored at room temperature over sulphuric acid, beneath glass bell-jars. This precaution was necessary to prevent

deterioration in the damp climate of Calcutta. The daily temperature varied between 80° and 90° Fahrenheit. The time during which the vegetables were thus stored before consumption did not exceed two weeks in most cases.

Preliminary experiments showed that guinea-pigs readily consumed, and thrive on, 50 g. of fresh vegetable daily. Holst and Frölich [1912], and later Delf [1918], had previously shown that guinea-pigs remain scurvy free and put on weight on 30 g. of fresh cabbage daily in addition to a scurvy producing basal diet. In our experiments we thought it advisable to use a larger daily ration to allow for the less nourishing properties of vegetables grown in India, and for possible variations in the vitamin content of different varieties of fresh vegetable.

In the case of sun-dried vegetables it was found that 1 g. dried vegetable corresponds roughly to 10 g. fresh vegetable. The ration of dried vegetable chosen was therefore 5 g. and it was found that this amount was readily consumed in most instances.

Six guinea-pigs were used for each experiment, all the animals were placed on a basal diet consisting of crushed oats, bran, hay and water *ad libitum*, to which was added daily 30 cc. of whole milk autoclaved at 120° C. for one hour. Two animals were given 50 g. fresh vegetable daily; two 5 g. of the sun-dried product soaked in cold water; and two 5 g. of sun-dried product cooked for about 30 mins., according to the directions given by the producers (boiled in water and the liquor strained off). The animals were kept in open cages and were allowed to run about for a short time daily. They were weighed every fourth day under the supervision of one of us, and carefully inspected for clinical signs of scurvy, when the onset of the symptoms was expected. Post mortem examinations were carried out on all animals that died and histological examinations of the rib-junctions and other parts when necessary.

To determine the antineuritic properties, common fowls, weighing between 1200 and 2000 g., were used. These birds were given a basal diet of Rangoon polished rice with water *ad libitum*, to which was added 10 g. of the sun-dried product under investigation. Regular weighings and inspections were carried out as in the case of the guinea-pigs.

Sun-dried carrots (Exp. 1, Fig. 1).

A. Control; guinea-pigs Nos. 1 and 2—basal diet with 50 g. fresh carrots daily.

Remarks: Both animals remained healthy for 91 days. No. 1 gained 75 g. in weight, and No. 2 10 g.

B. Guinea-pigs Nos. 13 and 14—basal diet with 5 g. sun-dried carrots soaked in water, but not cooked.

Post mortem report: Both animals showed typical signs of scurvy—enlargement of the costo-chondral junctions with haemorrhages, destruction of the cartilage columns at the junctions, and haemorrhages into various joints.

In No. 1 there was a subserous haemorrhage in the bladder. No. 2 had haemorrhages in the kidneys. All other organs were healthy.

Remarks: No. 1 lived 36 days and No. 2, 30 days, indicating a slight degree of protection as compared with animals on the basal diet only. Both animals developed diarrhoea a few days before death, which probably hastened the fatal issue.

C. Guinea-pigs Nos. 25 and 26—basal diet with 5 g. sun-dried carrots cooked as directed.

Post mortem report: No. 25 showed typical signs of scurvy, enlargement of the costo-chondral junctions with haemorrhages, subserous haemorrhages in the bladder. Subcutaneous and deep haemorrhages in the knee-joints. Other organs healthy.

Fig. 1. Experiment No. 1. Sun-dried carrots.

No. 26 had a haemorrhage in the broad ligament but no other signs of scurvy.

Remarks: No. 25 lived 32 days showing no protection as compared with controls on the basal diet.

The death of No. 26 was probably hastened by the fact that it aborted on the 21st day, giving birth to two dead foetuses. It died on the 27th day before the symptoms of scurvy were fully developed.

Sun-dried brinjal (Exp. 2, Fig. 2).

A. Control. Guinea-pigs Nos. 3 and 4—basal diet with 50 g. fresh brinjal daily.

Remarks: Healthy for 91 days. No. 3 gained 50 g., and No. 4 130 g.

B. Guinea-pigs Nos. 15 and 16—basal diet with 5 g. sun-dried brinjal soaked in water, but not cooked.

Post mortem report: Both animals showed typical signs of scurvy. In the case of No. 15 all the costo-chondral junctions were enlarged and haemorrhagic. There were haemorrhages into and around the knee-joints. No. 16 showed enlargement of and haemorrhage into the costo-chondral junctions. Microscopical examination of sections showed destruction of the cartilage columns, and haemorrhages at the costo-chondral junctions.

Fig. 2. Experiment No. 2. Sun-dried brinjal.

Fig. 3. Experiment No. 3. Sun-dried spinach.

Remarks: No. 15 lived 32 days and No. 16, 31 days, thus showing no protection as compared with the control animals.

C. Guinea-pigs Nos. 27 and 28—basal diet with 5 g. cooked sun-dried brinjal.

Post mortem report: Both animals exhibited typical signs of scurvy. No. 27 had haemorrhages in the bladder and broad ligament.

Remarks: No. 27 lived only 27 days; No. 28, 29 days, thus showing no evidence of protection.

Sun-dried spinach (Exp. 3, Fig. 3).

A. Control. Guinea-pigs Nos. 5 and 6—basal diet with 50 g. fresh spinach daily.

Remarks: Healthy throughout, but the gain in weight was not as marked as in some of the other groups.

B. Guinea-pigs Nos. 17 and 18—basal diet with 5 g. of soaked sun-dried spinach daily.

Post mortem report: Both animals showed marked signs of scurvy—enlarged and haemorrhagic costo-chondral junctions; haemorrhages into joints and various organs, subcutaneous and intra-muscular haemorrhages.

Remarks: No. 17 lived 32 days, and No. 18, 28 days. Both animals developed diarrhoea as a terminal symptom.

C. Guinea-pigs Nos. 29 and 30—basal diet with 5 g. cooked sun-dried spinach.

Post mortem report: Both animals had typical scorbutic lesions in the costo-chondral junctions. No. 30 had a haemorrhage into the broad ligament. There were scybalae in the large intestine.

Remarks: It is remarkable that both animals lived a shorter time than the controls on basal diet only. No. 29 died on the 28th day, and No. 30 on the 29th day. This may have been due to the fact that both animals were pregnant. No. 29 aborted on the 18th day, and No. 30 was delivered of a dead foetus on the 26th day.

Sun-dried cabbage (Exp. 4, Fig. 4).

A. Guinea-pigs Nos. 7 and 8—basal diet with 50 g. of fresh cabbage daily.

Remarks: Healthy throughout.

B. Guinea-pigs Nos. 19 and 20—basal diet with 5 g. of uncooked sun-dried cabbage.

Post mortem report: No. 19 (killed with chloroform), no evidence of scurvy.

No. 20. There was some slight enlargement of the costo-chondral junctions, but no haemorrhages were found.

Remarks: No. 19 lived 91 days without any signs, clinical or post mortem, of scurvy.

Fig. 4. Experiment No. 4. Sun-dried cabbage.

No. 20 maintained its weight for 48 days and then entered on a rapid decline dying on the 57th day. This animal also suffered from diarrhoea. The cause of death is unknown, careful macroscopic and microscopic examination revealed no definite signs of scurvy.

C. Guinea-pigs Nos. 31 and 32—basal diet with 5 g. sun-dried cabbage cooked as directed.

Post mortem report: No. 31, slight enlargement of the costo-chondral junctions—no haemorrhages.

No. 32, typical signs of scurvy in the costo-chondral junctions. Haemorrhages over the scapula. Organs healthy.

Remarks: The degree of protection shown here is less than in the case with cabbage fed after simple soaking in water. Animal No. 31 was apparently healthy till the 36th day, when it developed diarrhoea. It showed practically no signs of scurvy after death on the 44th day.

No. 32 aborted on the 23rd day and from that date declined rapidly, dying on the 40th day.

Fig. 5. Experiment No. 5. Sun-dried tomatoes.

Sun-dried tomatoes (Exp. 5, Fig. 5).

A. Guinea-pigs Nos. 9 and 10—basal diet with 50 g. fresh tomatoes daily.

Remarks: Healthy throughout. No. 9 gained in weight 105 g.; No. 10, 110 g.

B. Guinea-pigs Nos. 21 and 22—basal diet with 5 g. of soaked sun-dried tomatoes daily.

Post mortem report (on No. 21 killed with chloroform). No signs of scurvy.

Remarks: Both guinea-pigs appeared to be perfectly healthy when the experimental feeding was discontinued. No. 21 gained 65 g. and No. 22 60 g. in weight. Protection against scurvy would appear to be complete.

C. Guinea-pigs Nos. 33 and 34—basal diet with 5 g. sun-dried tomatoes cooked as directed.

Post mortem report (on No. 33 killed with chloroform). No evidence of scurvy.

Remarks: Both animals were healthy when the experiment was stopped. No. 33 suffered from diarrhoea from the 52nd to the 55th day, but recovered without special treatment. This fact may explain its loss in weight—50 g. No. 34 gained 50 g. in weight.

Sun-dried potatoes (Exp. 6, Fig. 6).

A. Guinea-pigs Nos. 11 and 12—basal diet with 50 g. fresh potatoes daily.

Remarks: This vegetable was not relished to the same extent as the preceding kinds. The average quantity consumed by both animals was about 20 g. daily. Both were healthy at the conclusion of the experimental period. No. 11 gained 35 g. in weight; and No. 12, 65 g.

B. Guinea-pigs Nos. 23 and 24—basal diet with 5 g. of soaked sun-dried potatoes daily.

Fig. 6. Experiment No. 6. Sun-dried potatoes.

Post mortem report: On No. 23. No evidence of scurvy, costo-chondral junctions normal. No haemorrhages. Organs all appear healthy. Cause of death unknown.

Remarks: No. 23 appeared to be perfectly well till about the 20th day, when it began to lose weight. There was no diarrhoea. The animal refused to take food and died on the 28th day. There were no clinical manifestations of scurvy at any time. No. 24 continued to do well and had gained 155 g. in weight at the conclusion of the experiment.

C. Guinea-pigs Nos. 35 and 36—basal diet with 5 g. sun-dried potatoes cooked as directed.

Post mortem report: No. 35, no signs of scurvy, cause of death unknown.

No. 36. Enlargement of costo-chondral junctions with haemorrhages. Haemorrhage into bladder, subcutaneous and intra-muscular haemorrhages around the knee-joints.

Remarks: The soaked product would appear to afford almost complete protection against scurvy, the boiled product only a moderate degree.

Mixed factory-dried vegetables (Exp. 7).

A. Guinea-pigs Nos. 41 and 42—basal diet with 5 g. daily of mixed factory-dried vegetables soaked in water.

Initial weights: No. 41, 460 g.; No. 42, 415 g. *Final weights:* No. 41, 270 g.; No. 42, 320 g.

Post mortem report: Both animals showed the usual typical signs of scurvy—enlarged and haemorrhagic costo-chondral junctions, haemorrhages into joints and various organs.

Remarks: No. 41 lived 29 days; No. 42, 37 days. Both showed marked symptoms of scurvy before death.

B. Guinea-pigs Nos. 43 and 44—basal diet with 5 g. daily of mixed factory-dried vegetables cooked as usual.

Initial weights: No. 43, 580 g.; No. 44, 570 g. *Final weights:* No. 43, 350 g.; No. 44, 250 g.

Post mortem report: Both animals, more particularly No. 43, had enlarged and haemorrhagic costo-chondral junctions, subcutaneous and joint haemorrhages.

Remarks: No. 43 lived 26 days; No. 44, 23 days. Both had marked clinical signs of scurvy before death.

Sun-dried turnip-tops (Exp. 8).

A. Guinea-pigs Nos. 45 and 46—basal diet with 5 g. soaked sun-dried turnip-tops daily.

Initial weights: No. 45, 350 g.; No. 46, 370 g. *Final weights:* No. 45, 305 g.; No. 46, 290 g.

Post mortem report: Both animals had typical signs of scurvy—enlarged and haemorrhagic costo-chondral junctions, haemorrhages into joints and other parts.

Remarks: Both animals lived 47 days and exhibited well-marked clinical signs of scurvy before death. No. 45 developed diarrhoea on the 38th day.

B. Guinea-pigs Nos. 47 and 48—basal diet with 5 g. of boiled sun-dried turnip-tops daily.

Initial weights: No. 47, 520 g.; No. 48, 480 g. *Final weights:* No. 47, 310 g.; No. 48, 310 g.

Post mortem report: No. 47 had enlargement of the costo-chondral junctions with extensive haemorrhages into knee-joints, muscles, etc. No. 48 had enlargement of the costo-chondral junctions, but no haemorrhages.

Remarks: No. 47 lived 44 days and showed well-marked clinical symptoms of scurvy before death. No. 48 did not have any definite symptoms of scurvy, when it died on the 47th day.

Sun-dried turnips (Exp. 9).

A. Guinea-pigs Nos. 49 and 50—basal diet with 5 g. uncooked sun-dried turnips daily.

Initial weights: No. 49, 570 g.; No. 50, 520 g. *Final weights:* No. 49, 355 g.; No. 50, 400 g.

Post mortem report: There was some enlargement of the costo-chondral junctions in both cases. Haemorrhages were not marked.

Remarks: No. 49 lived 32 days and No. 50, 37 days; neither showed very marked clinical or other evidence of scurvy.

B. Guinea-pigs Nos. 51 and 52—basal diet with 5 g. of boiled sun-dried turnips daily.

Initial weights: No. 51, 400 g.; No. 52, 450 g. *Final weights:* No. 51, 225 g.; No. 52, 250 g.

Post mortem report: Both animals, especially No. 51, showed typical signs of scurvy.

Remarks: Practically no protection.

Control; basal diet only (Exp. 10).

Guinea-pigs Nos. 37 and 38.

Initial weights: No. 37, 530 g.; No. 38, 520 g. *Final weights:* No. 37, 350 g.; No. 38, 390 g.

Post mortem report: Both animals exhibited the typical post mortem signs of scurvy. The costo-chondral junctions were enlarged and haemorrhagic, and under the microscope showed destruction of the cartilage columns. There were extensive subcutaneous haemorrhages in both legs, haemorrhages into and around the knee-joints. No. 38 had submucous haemorrhages in the bladder. The teeth were loose and the gums congested. The jaw bone was brittle and broke easily. No. 37 had no marked changes in the teeth and gums.

Remarks: Before death both animals exhibited the typical symptoms of scurvy—lameness, face ache position, etc. Signs of scurvy first appeared about the 25th day and both animals died on the 32nd day.

Antineuritic properties (Exp. 11, Fig. 7).

Fowl No. 1. Basal diet—Rangoon rice with 10 g. boiled sun-dried carrots daily (see Fig. 7).

Remarks: At the conclusion of the experiment the bird was quite healthy and had gained 180 g. in weight.

Fowl No. 2. Basal diet with 10 g. boiled sun-dried brinjal daily (see Fig. 7).

Remarks: The bird, though it had lost 290 g. in weight, was apparently healthy and showed no signs of neuritis.

Fowl No. 3. Basal diet with 10 g. of boiled sun-dried spinach daily (see Fig. 7).

Remarks: This bird remained healthy and gained 120 g. in weight.

Fowl No. 4. Basal diet with 10 g. of boiled sun-dried cabbage daily (see Fig. 7).

Remarks: This bird gained 430 g. in weight and was healthy and vigorous at the end.

Fowl No. 5. Basal diet with 10 g. of boiled sun-dried tomatoes daily (see Fig. 7).

Fig. 7. Antineuritic properties.

Post mortem report: Organs healthy, cause of death could not be ascertained. On histological examination the sciatic nerves were found to be normal and there was no sign of any degeneration.

Remarks: The bird was quite healthy until it suddenly died on the 79th day, having gained 70 g. in weight. There were no clinical signs of neuritis.

Fowl No. 6. Basal diet with 10 g. of boiled sun-dried potatoes daily (see Fig. 7).

Post mortem report: Organs healthy. On histological examination no signs of nerve degeneration could be found.

Remarks: This bird lost 70 g. in weight, otherwise it was quite healthy until the 68th day when it suddenly died—clinically there was no sign of neuritis.

Fowl No. 7. Control—basal diet only (see Fig. 7).

Post mortem report: Much emaciated. The skin of the legs was dry and scaly. No appreciable swelling was noticed. Macroscopically the organs were normal. On histological examination the sciatic nerves showed extensive degeneration—the axis cylinders of practically all the nerves had broken up, and the medullary sheath was converted into myelin droplets.

Remarks: On the 16th day the bird began to look unhealthy and gradually developed neuritis with its clinical symptoms. On the 33rd day he could scarcely stand, and moved with high stepping gait. He lost 500 g. in weight and died on the 40th day.

DISCUSSION.

From the short description of the process by which sun-dried products are prepared, quoted from our original paper, it will be noted that the drying process is carried out at the ordinary atmospheric temperature at Quetta, which varies between 75° and 100° F. during the drying season. They would therefore correspond in the main to the low-dried products of Givens and Cohen [1918] and Givens and McClugage [1919]. To what extent the preliminary scalding or steaming affects the process can only be determined by further experiments which we propose to carry out shortly. It is probable however that, in the case of thick slices, only the superficial cells are raised to a high temperature, and the central cells may to some extent escape the effect. The structure of the vegetable cells themselves may also have an influence, the coarser thick-walled types being more resistant.

From Table I we see that sun-dried tomatoes afford complete protection when fed both uncooked and cooked. Cabbage and potatoes afford partial protection, the degree of protection being more marked with the uncooked material. Tomatoes and potatoes are cut in comparatively thick slices and cabbage has a somewhat coarse leaf. The texture of brinjal and spinach, two varieties which show absolutely no antiscorbutic properties, is comparatively delicate.

A point which appears to emerge from Table I is the influence of cooking. Animals on the uncooked product lived longer in almost every instance than those on a similar ration fed in the cooked state.

Table I. *Antiscorbutic properties.*

Vegetable		No. of days guinea-pigs lived on 5 g. daily in addition to basal diet	
		Uncooked	Cooked
Sun-dried carrots	...	36, 30	32, 24
„ brinjal	...	33, 31	27, 29
„ spinach	...	32, 28	26, 27
„ cabbage	...	91, 57	40, 44
„ tomatoes	...	91, 91	91, 91
„ potatoes	...	26, 91	37, 47
„ turnips	...	37, 32	39, 35
„ turnip-tops	...	50, 50	46, 43
Mixed factory-dried	...	32, 23	—
Control	...	32, 32	—

Table II. *Antineuritic properties.*

Vegetable		No. of days fowls lived on 10 g. daily in addition to basal diet	Remarks
Sun-dried	carrots ...	91	No evidence of neuritis
"	brinjal ...	91	" "
"	spinach ...	91	" "
"	cabbage ...	91	" "
"	tomatoes ...	79	No evidence of neuritis. Cause of death unknown
"	potatoes ...	68	" "

SUMMARY.

1. Nine varieties of Quetta sun-dried vegetables have been investigated as regards their antineuritic and antiscorbutic properties.

2. Sun-dried tomatoes, potatoes and cabbage retain to a considerable extent the antiscorbutic properties of the fresh vegetable, when fed as a daily ration of 5 g. dried product to guinea-pigs weighing on an average 500 g.

3. Sun-dried carrots, brinjal, spinach, turnips and turnip-tops have little or no antiscorbutic properties.

4. Mixed factory-dried vegetables are entirely devoid of antiscorbutic properties.

5. Cooking before consumption diminishes any antiscorbutic vitamin which may be present. These products should therefore be given in the form of salads when possible.

6. Six varieties of Quetta sun-dried vegetables, viz. carrots, brinjal, spinach, cabbage, tomatoes and potatoes, were investigated as regards their antineuritic properties. In no instance was there any evidence of neuritis in common fowls weighing on the average 1600 g. on a daily ration of 10 g.

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