FURTHER OBSERVATIONS ON THE EXPERIMENTAL PRODUCTION OF URINARY CALCULI*

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In previous communications,[1,2,3] it has been demonstrated that if white rats were maintained for a period of time on a diet deficient in vitamin A, urinary calculi would develop in a large percentage.

In our researches, it was found that when animals were fed a diet deficient in vitamin A for a period of 250 days, postmortem study revealed the presence of vesical calculi in 88 per cent and of renal calculi in 42 per cent of the animals. It likewise was shown that if vitamin A alone were added to the deficiency diet, the formation of calculi was prevented. After the presence of calculi, which were too large to pass spontaneously from the kidney or bladder of the rats, had been demonstrated roentgenographically, an excess of vitamin A was added to the deficiency diet, and in every instance, provided the experimental animal survived, the calculus then underwent spontaneous solution and disappeared. This solution occurred in a comparatively shorter length of time when a co-existing urinary infection was not present. The calculi produced in this experimental work varied in diameter from 0.5 to 12 mm. They were light brown in color, spherical in shape, and they were composed chiefly of calcium phosphates with traces of carbonates. No oxalates or uric acid were present. A small amount of mucoid substance was noted also.

After these researches, the question arose as to whether the chemical constituents of the calculi could be changed by altering the diet. As reported previously, a decrease in the phosphorus in the diet in relation to the calcium caused a complete reversal in the chemistry of the stone, and under these circumstances the principal constituent of the stones was then calcium carbonate. Traces of calcium and magnesium phosphate were found. Traces of oxalates were present, but no uric acid was found.

The question which then presented itself was: "Why can uric acid calculi not be produced in the white rat by further modification of the diet?" The absence of uric acid in the calculi produced in the white rat may, of course, readily be explained by the fact that these animals possess the power of oxidizing approximately 80 per cent of the circulating purines to allantoin. Allantoin is much more soluble in water than uric acid and thus it never forms a constituent of the calculi in rats.

It has been stated that calculi have been produced experimentally only in white rats—a species quite remote phylogenetically from man.

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Therefore, a similar series of experiments with dogs has been in progress for the past year. In the dog, as in the rat, care must be taken to maintain a mild deficiency diet for a long period of time. If more than a moderate degree of vitamin A deficiency is continued, the animals die from pulmonary complications or from diarrhea—a disease which simulates distemper (Fhloring[4]). Thus, only a very mild degree of vitamin A deficiency is permitted for a long period. Such symptoms as loss of weight, xerophthalmia, and marked weakness of the legs should not occur.

A group of seven dogs has been maintained on the vitamin A deficiency diet for a period of eleven months. Recently, under ether anesthesia, cystotomy was performed on each of four of these animals. Multiple stones were found in the bladders of three, and no stones or sand were noted in the fourth dog. One large stone which was too large to pass spontaneously was allowed to remain in the bladder of each of the three dogs and the bladder was closed by the usual technic. The dogs are now being fed a high vitamin A acid-ash diet in order that we may ascertain whether the stones will undergo solution.

Chemical analyses of the calculi removed from these dogs revealed that they were composed of ammonium and calcium phosphate with small amounts of carbonates. No urates or uric acid were present. Although this series of experiments is too small to warrant definite conclusions, at least we may infer that bladder calculi may be produced in dogs maintained on a mild vitamin A deficiency diet for a considerable period of time.

The possibility of producing calculi composed of uric acid and urates was then considered. It is well known that birds eliminate uric acid and that a greater part of the urea is converted into uric acid. Benedict and Behré[5] discovered the unique mechanisms of uric acid metabolism in pure bred Dalmatian dogs by which considerable uric acid is eliminated in the urine. Investigations are now in progress in our laboratories in an attempt to produce calculi in this animal. Emmett and Peacock[6] in 1923 learned from autopsy findings that avitaminosis in chicks was associated with the presence of urates in the kidneys and at times on the surface of the heart and spleen.

Cruickshank, Hart and Halpin[7] found kidney lesions in practically all chickens that died during their experiments with avitaminosis. The kidneys were pale and contained an accumulation of urates. Likewise Elvehjem and Neu[8] reported an elevation of the blood urea when the chicks were maintained on a diet deficient in vitamin A.

Therefore, we used the following diet in a series of experiments with twenty chickens:

58 parts ground white corn	1 part precipitated calcium carbonate
25 parts wheat middlings (standard)	1 part precipitated calcium phosphate
12 parts crude casein	2 parts dried yeast
1 part sodium chloride	water ad libidum

Irradiation was administered for twenty minutes three times a week. The control chickens were fed green yellow corn instead of white corn. In the test animals, the white corn was replaced occasionally by yellow corn in order to maintain only a slight deficiency for a long period of time. The growth curves and symptoms were indicative of mild deficiency. At intervals, the birds were allowed to progress to the stage in which they developed a staggering gait, ruffled feathers and weakness of the legs. No xerophthalmia was noted.

At autopsy five months later, the kidneys were found to be somewhat pale, urates and sand were present in the kidneys, and the ureters contained urates and small uric acid calculi. Hard concretions of uric acid were present in the cloaca. The blood uric acid in the controls averaged 5.1 mg. per hundred cubic centimeters of blood and after the birds studied had been on the deficiency diet for a period of five months, just prior to postmortem study, the blood urea averaged 14 mg. per hundred cubic centimeters of blood. In order to produce calculi similar to vesical stones, a colostomy has been performed in chickens thus diverting the fecal stream and utilizing the lower segment of the bowel and the cloaca as a bladder. From this structure, small calculi can easily be removed for chemical examination which has shown that they were composed purely of uric acid and urates.

In conclusion, while an insufficient number of dogs and birds has been studied to warrant definite conclusions, the relationship between vitamin A deficiency and the formation of calculi seems probable and the mechanism probably is similar to that which occurs in the white rat.

References

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