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Some Observations on Vitamins A and D Activation of Yeast by X-Ray and Other Radiation.

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During the course of experimentation with the production of varying degrees of erythema of the skin, induced by x-ray over a small area of the body of the white rat, it was noted that animals fed a ration deficient in vitamin A and given a mild erythema dose developed symptoms of deficiency later as compared to control series.[1] It had been noted in previous experiments that a moderately large quantity of x-ray given in the form of a general body dose induced to a certain extent a vitamin A deficiency.[2] I assumed at that time that the x-ray played a rôle in the activation or utilization of vitamin A in the body and might be a source of activation of vitamin A in other forms in nature.

In the literature the greater amount of investigation has been centered upon activation of vitamin D or the antirachitic vitamin in various substances by means of the quartz light and the Cooper-Hewitt, or carbon arc lamp.

During the last two years I have studied the activation of yeast by the carbon arc, the quartz light and the x-ray. Various periods of exposure were used with each set-up. The yeast used was that of compressed Brewer's variety.* The test for vitamin A has resorted itself into a biological one in that the chemical tests with trichloracetic acid and similar chemicals as described by Fearon[3] and others, have proven to be unreliable.[4] I have used the white rat in all of these experiments and for the determination of vitamin D deficiency I have studied the bony structure by means of radiograph, silver impregnation in the case of amputated extremities, and as a check, hematoxylin and eosin stain in a few cases. The radiographic evidence will give data at varying periods of time in an animal under a period of observation.

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[1] Jorstad, L. H., and Lane, C. W., *Arch. Derm. and Syph.*, 1929, xix, 954.

[2] Burrows, M. T., Jorstad, L. H., and Ernst, E. C., *Radiol.*, 1928, vi, 7.

* The yeast was furnished by the laboratories of Anheuser-Busch, Inc. The radiation with the Cooper-Hewitt light and the quartz light was done by the same laboratory, who also granted certain amounts of money for materials used.

[3] Willimott, S. G., and Moore, T., *Biochem. J.*, 1926, xx, 651.

[4] Hawk, Phil. B., *Science*, 1929, lxix, 200.

In our studies with the Cooper-Hewitt lamp the yeast was exposed for periods from 5 to 60 minutes. There was some protectivity against rickets in the yeast exposed for 30 minutes, but complete protection was not afforded with the samples radiated less than 50 minutes. The 60 minute radiated yeast was not as protective as the 50 minutes. In regard to the vitamin A activation it was noted that xerophthalmia occurred in animals fed samples radiated up to 30 minutes and protection was not afforded completely until the radiation period was 50 minutes.

With the quartz light complete protection against xerophthalmia and rickets occurred with 5 to 7 minutes' radiation of the yeast sample. This yeast was radiated in the same manner.

In each case 5% of the yeast by weight was added to the Steenbock ration in one series and to a ration composed of egg albumin, potato starch and McCollum's salt mixture in another series. The second diet is a better one for determining vitamin A deficiency in that the cracked corn used in the Steenbock ration contains a certain quantity of vitamin A. Crisco was also added to the diet in that this supplies fat free of the fat soluble vitamin. Here again as in the experiments with the Cooper-Hewitt lamp the higher exposure, that is, 9 and 11 minutes, with the quartz light, did not give the same protection as the lower periods of radiation.

In the former experiments the amount of x-ray which had protected the rat against a vitamin A deficiency was 20 MAM, given to an area on the animal 1 cm. in diameter. Thus, in this set-up it was decided to radiate yeast cake 1 cm. thick for periods of 20, 25 and 30 MAM. The set-up used was 10 inch distance, 9 inch spark, 90 kilovolts; 4 minutes' exposure giving 20 MAM, etc. With this yeast greatest protection was afforded against rickets by that given the 20 MAM exposure.

The interesting factor in these experiments is that there is a point wherein activation reaches its maximum and then becomes less. Thus, we may speak of an activation curve with a rounded contour, the apex being reached at a different period of time with the different lights used. From that one can assume that the question involved is that of energy, the energy output of the Cooper-Hewitt lamp being less than that of the quartz and the quartz being less than the x-ray.

In former experiments we have shown that with different types of x-radiation, that is, the high voltage type with filtration, the vitamin A liberation occurs at 10 MAM and with a higher radiation, such as 75 MAM, vitamin A has to be added to the organism in

order to prevent loss of body weight during a course of several treatments. Here again we have the same energy curve as we have in the above experiments. All these experiments tend to refute the idea that certain lights are specific in the liberation of certain products. It also tends to establish that vitamin D is closely allied with vitamin A and that the question of liberation of these vitamins from yeast is a question of the differences in energy given.

Conclusions. The Cooper-Hewitt lamp, quartz light and x-ray in low voltage cause an activation of the vitamin D and the vitamin A in yeast fed to rats along with a deficient vitamin D and vitamin A ration. The period of exposure of the light in each case varies with the type of light used. The maximum point of activation is found with each light.