Catalase Photoinactivation

Abstract. *The enzymatic activity of catalase is lost during exposure to sunlight in the presence of oxygen. A simultaneous decline occurs in the absorption peak at 405 nanometers.*

Carotenoids accompany chlorophyll in all aerobic photoautotrophs able to survive in nature (*1*). Carotene protects chlorophyll from photodestruction. Eyster (*2*), working with another porphyrin, reported that the catalase activity of carotenoid-deficient seedlings was much lower than the catalase activity of normal seedlings. Evidence of the photodestruction of catalase by visible light is reported here.

Crystalline catalase from beef liver (Nutritional Biochemicals, Inc.) was suspended in 0.01*M* potassium phosphate, *p*H 7.0, the concentration during the subsequent exposure to light being 0.01 mg of enzyme per milliliter. The source of light was the sun. Intensities obtained by direct radiation were 99,000 to 110,000 lu/m^2. A Wratten filter (2c) was used to cover the solution during exposure. This filter absorbed virtually all radiation below 390 nm. Effects of oxygen during illumination were evaluated by leaving the samples open to the atmosphere. To obtain relatively oxygen-free conditions, samples were alternately flushed with nitrogen containing no oxygen and evacuated three times. Catalase was assayed according to the procedure of Chance and Maehly (*3*) with the following dilution modifications. Nine milliliters of 0.057*M* H_2O_2 in 0.01*M* potassium phosphate buffer, *p*H 7.0, was combined with 1 ml of enzyme preparation at zero time, and the reaction was allowed to proceed for 3 minutes. At the end of the reaction time, 1 ml of the mixture was placed in 10 ml of 0.4*N* H_2SO_4. This acid-denatured enzyme-H_2O_2 solution was then titrated with 0.002*M* $KMnO_4$ to determine the residual amount of H_2O_2.

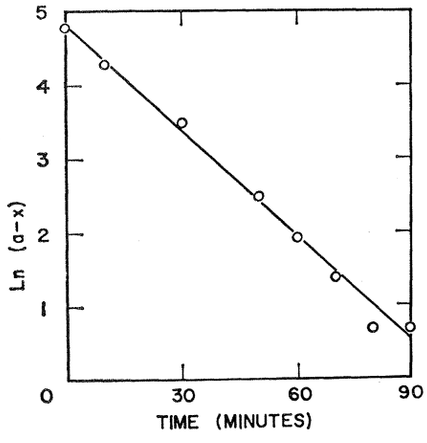

Fig. 1. Rate of catalase inactivation with increasing illumination time; *a*, initial concentration of enzyme in μg/10 ml; *x*, amount of catalase destroyed at time *t*.

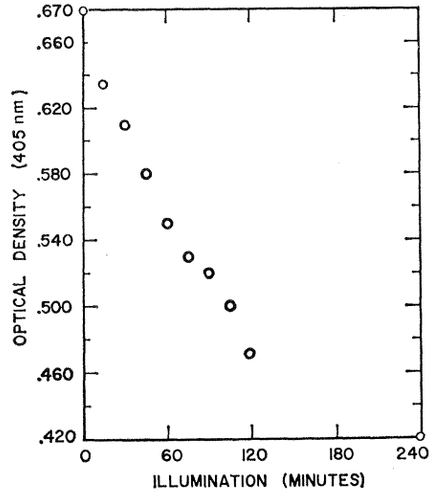

Fig. 2. The optical density decrease at 405 nm during a 240-minute interval.

Under vacuum, 92 percent of the activity of the catalase enzyme was retained after 30 minutes of exposure to sunlight. Aerobic samples lost all their activity from this sunlight exposure.

The rate of enzyme inactivation follows that for a first-order reaction (Fig. 1). Concomitant with the activity loss, the optical density of catalase at 405 nm (its visible absorption peak) declines as indicated in Fig. 2. The decrease in optical density follows a different kinetic pattern from that which describes the loss of enzymatic activity (*4*). We postulate that the enzyme was inactivated through alteration of one of the four porphyrin moieties within a catalase molecule, even though the remaining three retained their normal absorption spectrum. With prolonged illumination, all porphyrins would be altered.

Inactivation of catalase thus occurs in visible light in the presence of oxygen. There was a simultaneous decline in enzyme activity and absorption at the 405 nm peak during illumination.

ROGER L. MITCHELL
IRVIN C. ANDERSON
Department of Agronomy, Iowa State University, Ames

References and Notes

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Polonium-210 in Leaf Tobacco from Four Countries

Abstract. *Tobaccos grown in the United States, Rhodesia, South Africa, and New Zealand, have been measured for their polonium-210 content. Details of the method of measurement are given and the results are listed. A mean of 0.15 picocurie per gram has been found in New Zealand tobacco, compared with 0.49 picocurie per gram in United States tobacco. The concentration in South African tobacco was approximately the same as in United States tobacco, but the level in Rhodesian tobacco appeared to be significantly higher.*

The naturally occurring, alpha-emitting radioelement polonium-210 is found in plants generally, and its presence in tobacco is of special significance because this radioelement is volatile at the temperature of the burning cigarette, attaches itself readily to smoke particles, and thereby gains direct access to the lung. The report by Radford and Hunt (*1*) on the amounts of Po^{210} in four brands of American cigarettes and their assessment of the significance of inhaled Po^{210} as an initiator of lung cancer is of special interest.

Tobacco is normally aged for a period of 1 to 2 years between harvest and manufacture. This delay allows 138-day Po^{210} to approach equilibrium with its parent once removed—22-year lead-210 present in the leaf. The lead-210 in the plant may be derived from the soil or deposited on the leaves as "natural fallout" resulting from the decay of atmospheric radon-222. Any Po^{210} unsupported by lead-210 which might be present in the plant at harvest would largely disappear by radioactive decay during the ageing period.

Preliminary measurements of Po^{210}