weigh nearly twice as much as the heaviest individuals of most strains of laboratory albino mice.

This experiment was designed in the first place to throw light on the amount of improvement which might be made in the productive qualities of farm animals. When it is realized that the number of parent mice—some 1,200 males and 6,000 females—in the entire twenty-eight generations is very small compared to the hundreds of thousands or even millions of each kind of farm animal mated every year in the United States of America, it would seem that by adding a small amount, perhaps one per cent., to the labor now required to maintain these animals, their productivity can be changed from a level suited to medieval times to a level befitting the twentieth century. If the attained increase in the average weight of the mice is taken as a guide in estimating possible improvement, average milk yield could be increased 70 per cent., horses could be 70 per cent. stronger or speedier, pigs grow 70 per cent. faster, with a corresponding amount of improvement in other animals.[2] We may even wonder what the world would be like if the brain power of the human race were to be increased in like proportion.

Progeny-test breeding of the kind used with the mice may also be applied to plants in numerous ways, such as the development of races adapted to conditions of life to which the parent race is ill adapted, more rapid growth or less rapid growth, indeed to any continuously varying character—not that it will replace other methods of breeding, such as hybridization, which are so effective for some purposes, but that it can be used for purposes to which other methods of breeding are inapplicable, with good prospects of making progress in the desired direction.

The bearing of this work on evolution by means of natural selection is not found in the amount of change, which, after all, is not very large, or in the method of breeding, which does not occur in nature in so far as known, but in providing evidence that organisms may be modified at a rate and to an extent that makes further studies along this line worth the necessary time and effort. The means seem to be at hand for accelerating the rate of change which Darwin and his followers supposed occurred in nature at a very slow rate—too slow, indeed, to be observed in short periods of time. Progeny-test breeding, by accelerating the rate of change, offers a means of studying this aspect of evolution experimentally.

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2 Progeny-test breeding of poultry, under well-controlled conditions, by increasing egg yield by similar amounts, is another demonstration of the possibilities in progeny-test breeding.

ENZYMES IN ONTOGENESIS (ORTHOPTERA). XVII. THE IMPORTANCE OF COPPER FOR PROTYROSINASE[1]

AN inactive tyrosinase or protyrosinase yields a tyrosinase which can be poisoned by carbon monoxide, cyanide and diethyldithiocarbamate. The occurrence of copper in this protyrosinase is thus a reasonable expectation. The purpose of this paper is to test the effects on protyrosinase of the removal and subsequent addition of copper.

Protyrosinase was extracted from eggs of a grasshopper, *Melanoplus differentialis* (Thomas), according to a previously described method.[2] Its tyrosinase activity was measured in a Warburg apparatus. Aerosol, a commercially available synthetic detergent, was added in such excess that any protyrosinase was activated or converted into tyrosinase.

The procedure employed for removing copper from the protyrosinase was similar to that used by Kubowitz in studies of polyphenol oxidase.[3] An excess of potassium cyanide, 5 cc of .01 M KCN, was mixed with 20 cc of the protyrosinase extract. After a half hour 100 cc of saturated ammonium sulfate was added. The precipitate was centrifuged down, washed with 25 cc of saturated ammonium sulfate 0.05 M with respect to potassium cyanide, and dissolved in 0.9 per cent. sodium chloride. This solution was removed to a Cellophane tube and dialyzed against 0.9 per cent. sodium chloride for 50 hours at 0° C. A similar volume of protyrosinase extract received the identical treatment except that sodium chloride was substituted for potassium cyanide. The latter solution served as a control for the extraction of copper. After dialysis the volume of each of the two solutions was brought to 28 cc.

The copper content was estimated by means of a recently described method.[4, 5] A solution of copper sulfate in 0.9 per cent. sodium chloride was used as a source of copper. The total concentration of copper in the Warburg vessels is expressed in terms of moles per liter of reaction fluid (volume equals 3.0 cc).

The results of the treatment with potassium cyanide can be compared in terms of the change in amount of copper. During precipitation and dialysis the control protyrosinase preparation must have lost very little copper, since its 4.7×10^{-6} M represents a 98 per cent. recovery. Only one fifth as much, 0.9×10^{-6} M, remained in the extract treated with cyanide.

1 Aided by a grant from the Rockefeller Foundation for research in cellular physiology.

2 T. H. Allen and J. H. Bodine, *Proc. Nat. Acad. Sci.* (in press), 1941.

3 F. Kubowitz, *Biochem, Zeitschr.*, 299: 32, 1938.

4 A. Eden and H. H. Green, *Biochem. Jour.*, 34: 1202, 1940.

5 The analyses were performed by E. B. Newell.

The removal of copper apparently produces a decrease in protyrosinase which can be restored by the addition of a copper salt (Fig. 1). In agreement

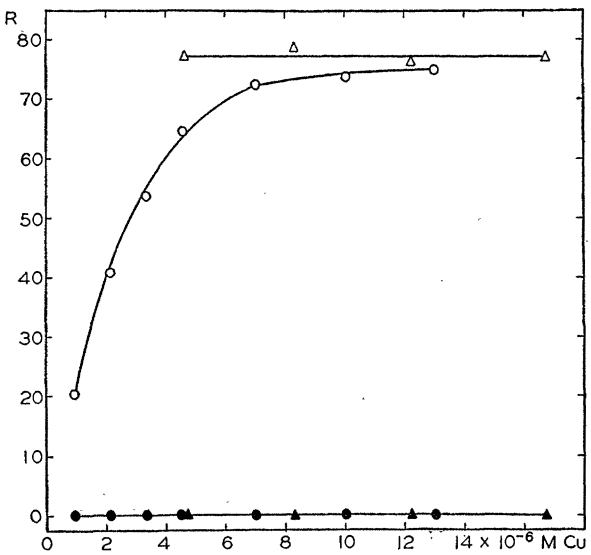

FIG. 1. Resynthesis of protyrosinase. Ordinate, reciprocal $\times 10^3$ of the time in minutes for the initial uptake of 100 c.mm. of oxygen during the oxidation of 6.9×10^{-3} mM of tyramine catalyzed by tyrosinase from 0.3 c.c. of protyrosinase extract. Abscissa, total concentration of copper. Open symbols, 0.07% Aerosol; closed symbols, no Aerosol. Circles, extract from which copper was removed; deltas, extract from which no copper was removed. pH = 6.8; Temp. = 24.9°C.

with Kubowitz[3] it was found that salts of Fe, Co, Ni, Mn and Zn were unable to replace those of Cu. The protyrosinase yields a tyrosinase which seems to be specifically dependent on copper for its enzymic activity. An excess of activator, Aerosol in this instance, seems to be necessary for conversion of protyrosinase into tyrosinase (Fig. 1). It is worthy of note that twice as much copper, that is 10 rather than 5×10^{-6} M, needs to be present for complete restoration of protyrosinase. The resynthesized protyrosinase and tyrosinase are destroyed by heating at 90° for 5 minutes; 18×10^{-6} M copper sulfate and the same with an excess of Aerosol have no catalytic activity. Thus it seems that copper unites with a copper-free substance to give a thermolabile protyrosinase, which in its turn must be activated before a thermolabile tyrosinase is produced. An anomalous and differential heat effect has also been found for protyrosinase extracts of low copper content. *i.e.*, heat treatment between temperatures of 60° and 70° not only inhibited but also activated.[6] The heat effects, therefore, appear to be independent of the copper.

Although the potentially active group of protyrosinase reacts with cyanide, it is unable to activate the enzymic oxidation of substrates. With the present knowledge of protyrosinase it seems hazardous to choose between whether the activation of protyrosinase is primary and direct or secondary and perhaps concerned with the removal of some material surrounding a core of tyrosinase. If the latter were so, it may be pointed out that the shell must be permeable to cyanide and its copper compound yet impermeable to substrates. This kind of semi-permeability would seem to be of a very peculiar order. Since Kubowitz[3] has shown that the active group of polyphenol oxidase indulges in electron exchange in order to catalyze the substrate's oxidation, it is suggested that a difference between protyrosinase and tyrosinase may be concerned with the state of its copper. An activator presumably overcomes some hindrance to oxidation and reduction of the active group.

When its copper is partially removed, a protyrosinase extract yields less tyrosinase. The return of copper leads to resynthesis of protyrosinase. The copper of protyrosinase seems to be a potentially active, prosthetic group.

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THE USE OF FATTY ACIDS IN INSECTICIDAL AEROSOLS

IN recent investigations[1] it has been shown that some relatively nonvolatile compounds show promise as fumigants against insects when applied in smoke or fog form. This development makes possible the use of safe and inexpensive insecticides that were formerly considered impractical because of difficulties in producing effective concentrations at room temperatures.

In practice a solution of the insecticidal material was sprayed on a heated surface. On coming in contact with the hot surface, the solvent was evaporated with explosive violence, and any dissolved material that did not vaporize readily was reduced mechanically to colloidal dimensions. That is, the insecticide was dispersed as an aerosol consisting of a suspension of the solid or liquid particles in air. By this method of volatilizing it was possible to keep the insecticide dispersed in an enclosed space for a long time. The rate of evaporation was also greatly increased, and the maximum vapor concentration was quickly obtained because of the tremendous surface of these

[6] J. H. Bodine and T. H. Allen, *Jour. Cell. and Comp. Physiol.*, 12: 71, 1938.

[1] W. N. Sullivan, L. D. Goodhue and J. H. Fales, *Soap*, 16 (6): 121, 123, 125, illus. 1940.