our first paper[1] on this subject, we did *not* describe the formation of alcohols from esters over Raney nickel.[9] While we plan to continue our work with coumarin and other lactones, the investigations already mentioned[4,6] conclude our interest in the behavior of simple esters over Raney nickel and we now have no plans for extending this phase of the work.

(9) The communication of Ovakimian, Kuna and Levene[3] indicates a misunderstanding of this point.

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UNSATURATED FAT OXIDASE: SPECIFICITY, OCCURRENCE AND INDUCED OXIDATIONS

Sir:

An oxidase found in soybeans converts unsaturated fats into peroxides.[1-6] Carotenoid pigments or vitamin A present in the fat are oxidized by unstable intermediate products, not by direct enzymatic action nor by the relatively stable fat peroxides.[3-8]

This enzyme (soy beans ground with 5 parts of water and centrifuged) has now been found to oxidize directly only those compounds containing the $-\overset{H}{C}=\overset{H}{C}(CH_2)_7C(O)-$ group with *cis* configuration. For example, oleic, ricinoleic, linoleic and linolenic acids and their esters absorbed oxygen rapidly. Under the same conditions, oleyl alcohol, elaidic and erucic acids (in decalin) and a variety of other unsaturated compounds were not oxidized (many of these compounds were generously donated by Dr. George Burr and by Dr. C. R. Noller).

The unsaturated fat oxidase has been detected in seeds of various legumes: *Acacia melanoxyln*, alfalfa, scarlet runner bean, white clover, *Laburnum anagyroides*, *Lupinus albus*,[2] *L. Benthamii*, *L. Greyi*, *L. nanus*, garden peas (green and dry), purple vetch (green and dry). It was not detectable in seeds of castor bean, almond, maize, cucumber, flax or sunflower, nor in etiolated barley seedlings in which rapid oxidations of the carotenoids are also known to occur.[9]

Many factors influence the oxidation of organic substances induced by concomitant enzymatic oxidation of fats. Easily autoöxidizable carotenoids (as eschscholtzxanthin)[10] were decolorized more rapidly than stabler pigments (as zeaxanthin).[9] In highly unsaturated fatty compounds (ethyl linolenate), the oxidatiou of carotenoids was much slower than that of the same pigments dissolved in olive oil, an indication of competition between the fat and the carotenoid for the oxidizing agent.

Chlorophylls *a* and *b* were oxidized to colorless substances by the system of fat, enzyme and oxygen. *p*-Phenylenediamine (but not the *o* or *p* isomer) was oxidized to a blue pigment, a reaction similar to that produced by the cytochrome complex. Dihydroxyphenylalanine (dopa) was oxidized to the black pigment melanin, a reaction also catalyzed by the highly specific dopa oxidase. Ascorbic acid was oxidized at a slow rate. In the absence of fats or if heated enzyme preparations were used, none of these induced oxidations took place.

Polyphenols exhibited remarkable effects upon the enzymatic oxidation of fats and upon the induced oxidation of other substances. Hydroquinone, pyrogallol and catechol (10 mg. per 2–3 ml. of enzyme solution) prevented the absorption of oxygen by unsaturated fats and thus prevented the induced oxidation of other substances. Phloroglucinol and resorcinol, by contrast, did not inhibit absorption of oxygen by the fat or decolorization of carotenoids. They did, however, prevent oxidation of *p*-phenylenediamine to the blue pigment. Formation of this blue pigment resulted from reaction between the amine and the fat peroxides. This reaction was inhibited by all the polyphenols, most of which were oxidized at a slow rate.

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