centration, as predicted by the present theory, and the limit + 58 mv. is approached according to both experiments and calculations.

Further discussions of this and other permeability problems, regarded as cases of "forced" diffusion, will, it is hoped, be presented in other communications.

8340 C

Apricot Seeds as a Source of Dehydrogenases.

CHARLES GURCHOT.

From the Pharmacological Laboratory of the University of California Medical School, San Francisco.

Increasing interest in various aspects of the mechanism of biological oxidation has developed a need for rich and reliable sources of dehydrogenases. An attempt was made to find such sources in appropriate plant material because of the possibility that such material might have an advantage in cheapness and in ease of handling. With the exception of yeasts, plants do not seem to have been much explored for dehydrogenases. Three samples of yeast from different manufacturers were found to be relatively poor sources for dehydrogenases, and samples from the same source varied greatly in such enzyme activity.

Preliminary search through representative available plant sources indicated the relative richness in dehydrogenases of the coatings of the seeds of various species of *Prunus*. With the skins removed, the seeds contain almost no dehydrogenases but are rich in lipases and emulsin. Such vegetables as beets and potatoes seem to be poor sources of dehydrogenases. Seeds of legumes and cereals are reported to be good sources of lactic dehydrogenase.[1] Representative assay data on dehydrogenase content are shown in Fig. 1 for extracts from beets, potatoes, yeast and seed coatings of almond, cherry, peach and apricot. Of these materials apricot seed coatings appear to be richest in dehydrogenases.

In extracting the dehydrogenases from fruit seeds, the seeds were first soaked for 24 hours in cold water to each liter of which one-half cc. of toluene was added. The skins were removed and extracted in a ball mill with disodium phosphate containing toluene, for 6 hours. To one liter of phosphate, 100 gm. of wet seed skins

[1] Anderson, B., *Z. Physiol. Chem.*, 1932, **210**, 15.

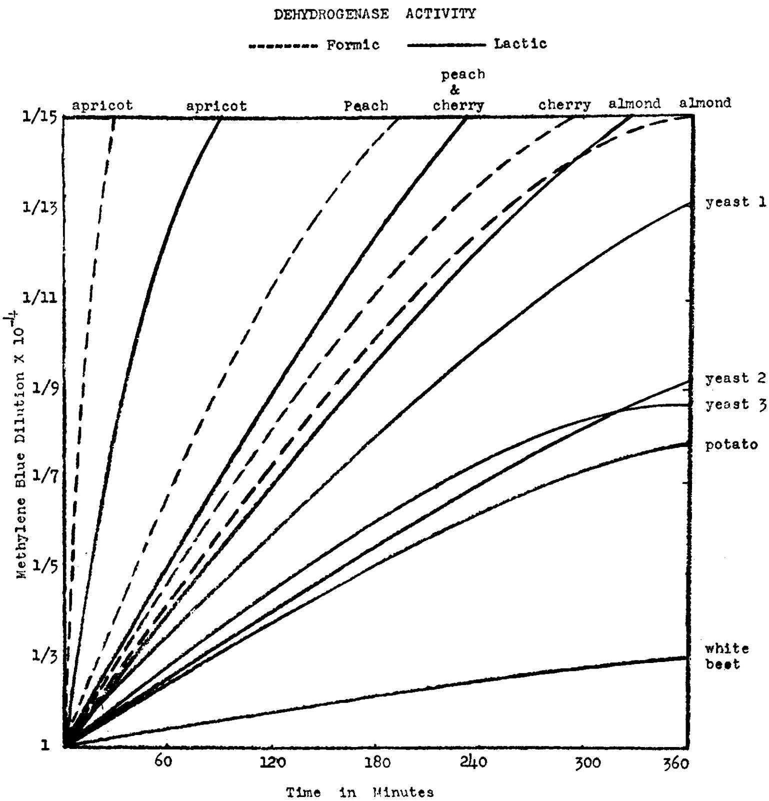

FIG. 1.
Dehydrogenase activity of various plant materials as determined by Thünberg methylene blue assay.

were added. The first extract contained most of the lactic and formic dehydrogenases. This was filtered and the skins extracted for another 6 hours with disodium phosphate. This second extract contained most of the malic, tartaric, and oxalic dehydrogenases. It was sometimes possible to separate lactic dehydrogenase from formic dehydrogenase by adsorbing the latter on alumina in acid buffer of pH 6.5. Formic dehydrogenase was then eluted with disodium phosphate. The phosphate filtrates were neutralized with dilute acetic acid and the enzyme precipitated with 5 volumes of 95% ethyl or methyl alcohol. The precipitate was centrifuged, washed with 95% alcohol, then ether, and dried in a vacuum desiccator over calcium chloride. The resulting material is gray and brittle. It can be purified to an almost white powder by dissolving in distilled water, dialyzing (to remove sulphydryl compounds which, when oxidized, compete with hydrogen acceptors and consequently interfere with the methylene blue assay), and re-recipitating twice with alcohol.

Preparations so made were assayed by the Thünberg technique.[2] Tests were made in a total volume of 10 cc. of phosphate buffer of pH 7.6, containing 10 mg. of enzyme powder, 1 cc. of 1% sodium lactate or its molecular equivalent of another appropriate substrate and 1:10,000 methylene blue. This was placed in a water bath at 36°C. for 6 hours and compared during this period with a series of methylene blue standards ranging from 1:10,000 to 1:150,000 which is almost colorless. The time required for the color to pale to the next lower standard was recorded. By adopting a suitable standard one may adjust samples of varying activity as determined by assay to a uniform degree of effectiveness per unit quantity.

All enzymes tested were inactivated by heating for 15 minutes at 60°C. They were not inhibited by HCN. It is interesting to note that formic dehydrogenase was found to be the most active dehydrogenase tested in the seeds of the 4 species.

8341 C

Effect of Cyclopropane on Isolated Intestinal Muscle.

S. A. PEOPLES* AND N. M. PHATAK. (Introduced by C. D. Leake.)

From the Pharmacological Laboratory, University of California Medical School, San Francisco.

It has been shown[1] that the *in vitro* effects of ether and ethylene on isolated intestinal muscle agree substantially with experimental evidence *in vivo*[2] and with clinical observations[3] that ether causes marked loss of intestinal tone and contractility while ethylene does not. It was also shown[1] that divinyl oxide, possessing a chemical structure combining characteristic features of both, is more like ethylene than ether in this respect as in other physiological actions. No reports so far seem to have been made regarding the effect of cyclopropane on intestinal muscle, although Waters and Schmidt[4]

[2] Thünberg, T., *Quart. Rev. Biol.*, 1930, **5**, 318.

* Merck Fellow in Pharmacology.

[1] Peoples, S. A., and Phatak, N. M., PROC. SOC. EXP. BIOL. AND MED., 1934, **32**, 378.

[2] Miller, G. H., *J. Pharm. Exp. Therap.*, 1926, **27**, 41.

[3] Luckhardt, A. B., and Lewis, D., *J. Am. Med. Assn.*, 1923, **81**, 1851.

[4] Waters, R. M., and Schmidt, E. R., *J. Am. Med. Assn.*, 1934, **103**, 975.