Preliminary Studies of the Anthocyanins of *Phytolacca Americana* on Amberlite IRC-50

CLARK H. ICE, THOMAS B. GAGE and SIMON H. WENDER,
University of Oklahoma, Norman

Wender *et al.* (2) have reported the use of ion exchange resins for the isolation from natural products of one group of polyphenols, the flavonoid compounds. This paper reports the preliminary findings concerning the behavior on Amberlite IRC-50 resin of a related group, the anthocyanins.

Experimental

One kilogram of the berries from *Phytolacca Americana* was extracted with 8 liters of distilled water by passing through a wet grinder (1). The resulting mixture was filtered to remove solids, and the filtrate used for ion-exchange studies. This filtrate gave the usual anthocyanin color reactions on treatment with acid and base.

A two inch glass column filled to a depth of 30 inches with Amberlite IRC-50 resin in the hydrogen form was used in all the experiments. One hundred ml. of the extract was passed through the column. Much of the material passed through the column, but the anthocyanin was adsorbed on the upper portion of the resin. When the column was washed with distilled water, the purple band was slowly eluted. The rate of elution, however, was such that separation from water soluble constituents, such as sugars, was obtained. There was no adsorption of pigment from an alkaline solution.

In another experiment, 100 ml. of the extract was made 0.4 N in HCl and then passed through the column. The anthocyanin was more tightly adsorbed than from the neutral extract. It could be eluted, however, with

distilled water. In this way anthocyanins may be separated from flavonoid compounds since the latter are not eluted with water.

The combined yield of anthocyanin, weighed as the lead salt, was 30 grams.

It was observed that in prolonged reduction of an alcoholic solution of the extract a clear solution was obtained. This was also found to be the case in the reduction of delphinidum. A similar reduction of a flavonoid gives a red solution which retains its color on prolonged treatment. This fact is of interest since it is commonly thought an anthocyanin is responsible for the reddish color of the magnesium reduction test on flavonoids.

The success of this preliminary study indicates that ion exchange resins may prove to be useful for the isolation and purification of anthocyanins.

ACKNOWLEDGMENT

This investigation was supported in part by a grant from the Atomic Energy Commission.

LITERATURE CITED

1. GAGE, T. B., S. H. WENDER, and L. CIERESZKO. 1952. A convenient laboratory model wet grinder and extractor. Anal. Chem. (In Press)
2. MORRIS, Q. L., T. B. GAGE, W. DETTY, and S. H. WENDER. 1951. The use of ion exchange resins with flavonoid compounds. Science *113*: 522-523.