September 11, 1978

Number 37

Citation Classics

Loomis W D & Battaile J. Plant phenolic compounds and the isolation of plant enzymes. *Phytochemistry* **5**:423-38, 1966.

Inability to isolate active enzymes from certain plants was found to be due to plant phenolic compounds. These bind to proteins more strongly than biochemists had realized. Polymeric adsorbents, adapted from brewing chemistry, released the plant enzymes from these complexes. [The *SCI*[®] indicates that this paper was cited 304 times in the period 1966-1977.]

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January 19, 1978

"This paper was born of frustration. We had set out to study the biosynthesis of monoterpenes in plants, and peppermint, which is an important crop in Oregon, looked like an ideal subject. Eventually we realized that the lack of publications in this area was not due to lack of effort, but to lack of results. Labeled precursors weren't incorporated effectively, and the only enzyme we could demonstrate was polyphenoloxidase, i.e., extracts turned brown.

"We tried every conventional technique of enzyme extraction, and several unconventional ones. We could prevent the browning, but we still couldn't detect other enzymes. We think the idea of adding insoluble protein to the homogenates came from a plant pathology paper describing the use of hide powder to adsorb tannins in isolating a virus from cocoa leaves. We didn't have any hide powder, but a neighboring laboratory had some beef heart muscle. We tried it, and it seemed to work. We then tested hide powder, collagen, and cottage cheese. They all gave us colorless extracts, with active enzymes, but we wanted something non-biological so we could be sure that we weren't adding contaminating enzymes. Discussing this with a colleague who does research on hops, I learned that the brewing chemists had recently solved the chill-haze problem in beer by adding synthetic polymers to adsorb plant phenolics. I went to the brewing literature and found that the brewing chemists had already done the basic plant biochemistry for us. Their best phenol adsorbent was insoluble PVP (Polyclar AT), and it was the key to isolating active enzymes from peppermint.

"The solution to our problem had come from unexpected sources. The background literature to explain the results came from even more diverse areas, including insect physiology, leather chemistry, and food technology. More recently we have improved our methods further by using adsorbent polystyrene beads that were originally developed for industrial waste treatment. We have become convinced that scientific advances often result from combining information from seemingly unrelated fields.

"We have also become convinced that there is much useful information and valuable insight in the older scientific literature. The older work should not be forgotten, but should be regarded as a starting point for current research. This is especially true in plant biochemistry, because plants were very popular subjects with the early biochemists. Their popularity waned because those biochemists had the same problem we did: it was hard to get active enzymes. The interest in our paper indicated that we were not the only ones still frustrated by plant enzymes."