This Week’s Citation Classic™

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This paper presents mobility data for 71 carbohydrates and derivatives in 8:8:4:1 isopropanol:pyridine:water:acetic acid solvent, at loads from 0.005 to 1.0 micromole. Spots are detected by four color reagents differing in selectivity. Ways of eliminating interference caused by ionic interactions are described. [The SCI® indicates that this paper has been cited in over 395 publications since 1980]

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“This was the first of six research papers in a ten-year collaboration by me (a biochemically oriented entomologist) and two analytical chemists working for CalPak (now the Del Monte Corporation, a subsidiary of RJR industries). Our friendship started with a mutual interest in pesticides. Sugars entered into the picture when CalPak found that pineapple from the Philippines often developed brown spots during the canning process, making it unacceptable to consumers. They assigned this problem to Thornburg, who enlisted me (among others) as a consultant. The browning turned out to be thermal degradation of 2,4-diketogluconic acid, a bacterial oxidation product of glucose formed during ripening of the pineapple. The techniques worked out in our paper were helpful in solving this puzzle.

“The starting point was a method I had developed earlier, with the assistance of a premedical student (now a prominent radiologist) for chromatographic separation of cations in insect blood.1 The solvent worked surprisingly well for sugars, polyols, and various organic acids and bases in crude biological extracts, since it handles high solute loads and minimizes ionic interactions that cause streaking of spots. We tried it on everything we could quickly lay our hands on, hoping to find something chemically similar to the mystery substance in pineapples. This required trying (and sometimes improving) many color reagents that had been used in prior carbohydrate work for the detection and characterization of sugars. We worked up a lot of data that we could see might be useful in later research by ourselves and others, so we decided to ‘do it up right’ for publication. This evolved into an unusually thorough study of the Rf and spot dimensions of varying loads of many compounds—in effect, measuring capacity, separability, and detection sensitivity and selectivity. The CalPak chemists (both of them perfectionists) deserve full credit for this. It led to recognition of their research skills by their company (Thornburg is now director of a large research group at Del Monte). Although I did much of the exploratory work, my ultimate role was in guiding and defining their experiments, and in writing the paper.

“We were surprised to learn that the paper has been frequently cited. It was perhaps the first ‘quick-and-dirty’ technique for learning something of the composition of biological extracts, and we later showed that it was applicable to an extraordinary variety of inorganic compounds.2 The early appearance of a technique of general utility (and also simple, fast, and inexpensive) in a large circulation analytical journal probably initiated a ‘chain reaction’ of citations, in multifarious contexts. Possibly only a few components of the method (such as the excellent color reagents, which did not require spraying) were widely adopted and so acknowledged.

“Modern chromatographic techniques use micron-size particles (of free or derivatized silica, synthetic polymers, etc.) instead of the matted cellulose fibers of paper, and have much higher resolving power. The plethora of TLC, GLC, and HPLC methods is reviewed by Churms,3 who does not cite our paper. Nevertheless, our method may still prove useful for the preliminary stages of some investigations, or in teaching laboratories that cannot afford costly state-of-the-art instrumentation.”