

This Week's Citation Classic™

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Brewbaker J L, Upadhyha M D, Mäkinen Y & Macdonald T. Isoenzyme polymorphism in flowering plants. III. Gel electrophoretic methods and applications. *Physiologia Plantarum* 21:930-40, 1968. [Dept. Horticulture, Univ. Hawaii, Honolulu, HI]

The article summarized inexpensive, do-it-yourself hardware and techniques for separating plant enzymes on horizontal gels. Applications were cited for several widely distributed enzymes of high stability and ease of handling in genetic and other biological studies. [The SC® indicates that this paper has been cited in over 130 publications since 1968.]

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"The discovery of gel electrophoresis was like the discovery of microcomputers to my team of plant breeders and geneticists, a looking glass through which we could step magically into a new world. Our research in tropical crop improvement often focused on plants that had not been studied genetically, plants with long life cycles. The ability to identify isoenzymes easily on gels provided a powerful genetic tool for trees, wild plant species, and other living organisms in which genetic study had previously been tedious or impossible.

"Our paper reflected the motivation of the College of Tropical Agriculture to provide techniques for colleagues abroad that could be widely and simply adapted. The co-authors were from India, Finland, and New England, each bringing his sense of minimal cost and maximal utility. We assembled our electrophoresis power sources from do-it-yourself kits, and bought most of our hardware in local grocery stores. We even tried replacing commercial (and expensive) starch for gels with supermarket cornstarch, since much of our work was on corn, but no matter how well purified, it failed! Our students, L. Espiritu, E. Hamill Johnson, J. Scandalios, and a visiting Swedish geneticist, Lars Beckman, contributed importantly to the refinement of these methodologies.

"Among the authors, we had prior experience applying gel electrophoresis in a surprising diversity of plants and plant tissues (reflecting our cosmopolitan backgrounds). We knew that

simple techniques were needed for the large scale screening of hundreds of genetic progenies. Enzymes of relative stability and easily sampled plant tissues were favored in these studies. We promoted isoenzymes as 'tools' for the plant breeder, a view that has been widely verified.

"Looking back, it seems to me we were overly modest in our assessment of the value of a paper such as this on techniques. It was widely cited primarily because it described simple and inexpensive methods adaptable to any research laboratory. So often, contributions of this type are 'put on the back burner' as we press to get out our hot new findings from application of the techniques. We published this as the third in a series of eight papers entitled 'Isoenzyme polymorphism in flowering plants,' and more than 30 papers have followed on uses and roles of plant isoenzymes. Among later papers reviewing this work is that entitled 'Polymorphisms of the major peroxidases of maize.'¹

"The preparation of a techniques paper is reserved in many instances until authors feel their techniques error-proof. Such is rarely the case, however, as modifications inevitably occur, often involving newly available equipment. Indeed, this paper would not have been published so soon had my conservative professorial view prevailed. Colleagues M.D. Upadhyha and Scandalios sensed much more the urgency of publication. As it was, the publication included two errors that have annoyed me for years, simple modifications that should have been refined before going to press.

"It seems in retrospect a classic enigma in my publishing experience, whether to wait until all the loose ends are tied up or publish with the view that other scientists may benefit immediately from your experience. This is a particular challenge for scientists in developing countries who cannot participate in international scientific workshops and annual meetings. In the future, we are challenged to develop an 'editable' publication format, with use of microprocessors to make minor corrections in manuscripts on techniques as the years add refinements. How delightful if computer-updated versions could be available annually as reprints!"

1. **Brewbaker J L, & Hasegawa Y. Polymorphisms of the major peroxidases of maize. *Isozymes—Curr. Top. Biol. Med. R.* 3:659-73, 1975.**