## This Week's Citation Classic

Gorin P A J & Spencer J F T. Proton magnetic resonance spectroscopy—an aid in identification and chemotaxonomy of yeasts. Advan. Appl. Microbiol. 13:25-89, 1970. [Prairie Regional Laboratory, National Research Council of Canada, Saskatoon, Saskatchewan, Canada]

A brief description of the chemotaxonomy of yeasts in terms of nucleic acid, protein, and antigenic structures is given, and the polysaccharide structure/proton magnetic resonance (PMR) method of classification is detailed. PMR spectra of oligosaccharides are interpreted in terms of chemical structure, and yeasts are classified according to the PMR spectra of their mannosecontaining polysaccharides. [The *SCI*<sup>®</sup> indicates that this paper has been cited in over 110 publications.]

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Proton nuclear magnetic resonance (NMR) spectroscopy of derived mannose-containing polysaccharides, together with their sugar composition, can be used to identify and classify yeasts by means of the fingerprint H-1 region. NMR identification can be particularly applied to yeasts with few typical characteristics and to identify unknown chemical features in the polysaccharides.

This discovery was made almost by pure chance, arising from my association with Frank Spencer, which occurred when the Prairie Regional Laboratory of the National Research Council (NRC), Saskatoon, was relatively young. Researchers were allowed to develop their own projects without restriction, and we both fitted in well in this archaic system. Frank began his career in oxygen transfer, later graduating to yeast genetics and yeast classification and identification, owing much to a born-again year with Herman Phaff at the University of California, Davis. I was a carbohydrate chemist specializing in polysaccharides and looking for a distinctive line of research. So, with the proximity of A.S. Perlin's NMR machine, it was natural that we collaborated.

Despite indications that there was structural variation of cell-wall mannans,<sup>1</sup> we were still surprised that the first six yeasts extracted gave a different typical H-1 region in the spectrum. We therefore immediately dropped nearly everything else and in the next few months isolated the mannose-containing polysaccharides from 450 different yeast species, this being successful in 410 cases. We obtained 150 different fingerprint spectra. Needless to say, Frank's technician, Norm Gardner, and the NMR spectroscopist, Ted Mazurek, suffered a lot. And I was not an impressive figure extracting 12 or 15 yeasts a day on crutches.

Before writing the review, we published an embarrassing number of papers, but we bore up and Frank invented his system for the classification of yeasts. The fingerprints also served to identify yeasts that were present in polluted water bodies and that had few typical characteristics.<sup>2</sup> Previously, yeasts were classified and identified according to their morphology, spore shape and number, the nature of the conjugation process, the assimilation of six sugars, ethanol, and nitrate, and the ability to hydrolyze arbutin. Much later, RNA homology proved to be an excellent method for interspecies comparison and identification, but it is cumbersome for comparison of large numbers of yeasts. The only failure of the NMR method is in dealing with yeasts formed by the crossing of haploid strains.<sup>3</sup>

For the chemist, NMR spectroscopy served to identify novel structures, such as rhamnomannans, mannans containing  $\alpha$ - and  $\beta$ -linkages, and those containing a high proportion of *N*-acetyl-glucosamine. These systems received further attention because of their enzyme components.

As the initial reaction to the classification and identification methodology was not enthusiastic, it was a pleasant surprise to hear that the review has been widely cited. Either a younger generation of yeast researchers has taken note, or the citations have occurred in unrelated chemical or biochemical journals. Recent work in this field is being carried out by Nobuyuki Shibata.<sup>4</sup>

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<sup>1.</sup> Gorin P A J & Perlin A S. A mannan produced by Saccharomyces rouxii. Can. J. Chem. 34:1796-803, 1956. (Cited 85 times.)

Spencer J F T, Gorin P A J & Gardner N R. Yeasts isolated from the South Saskatchewan, a polluted river. Can. J. Microbiol. 16:1051-7, 1970.

Spencer J F T, Gorin P A J & Rank G H. The genetic control of the two types of mannan produced by Saccharomyces cerevisiae. Can. J. Microbiol. 17:1451-4, 1971.

Shibata N, Kobayashi H, Tojo M & Suzuki S. Characterization of phosphomannan-protein complexes isolated from viable cells of yeast and mycelial forms of *Candida abicans* NIH B-792 strain by the action of zymolyase-100T. Arch. Biochem. 251:697-708, 1986.