COMMENTARY

by Bernard Dixon

WASTE HAY BECOMES RESOURCE, THANKS TO MICROBIAL PARTNERS

Perhaps the most valuable bequest Selman Waksman left to future generations of microbiologists was an enthusiasm for studying mixed populations of organisms as they occur naturally, in addition to the highly artificial pure cultures usually prepared for laboratory investigation. It was ecological thinking of this sort which led the exiled Ukrainian to soil microbiology and his 1943 discovery of streptomycin, which in turn ushered in the golden age of antibiotics.

Not all researchers have adhered closely to Waksman's philosophy. Indeed, there are cases of investigators being carried astray simply by overlooking crucially important interactions between microorganisms in their natural habitats. But others have been wiser—not least James Lynch of the U.K. Agricultural and Food Research Council's Letcombe Laboratory in Oxfordshire. For some time now, he and his colleagues have chosen to focus their attention on microbial communities in the soil. And today that work is on the point of paying off in the form of techniques (first described briefly in BIO/TECHNOLOGY, **1**:389) for producing high-value agrochemicals from one of the most tiresomely awkward of all agricultural wastes: straw.

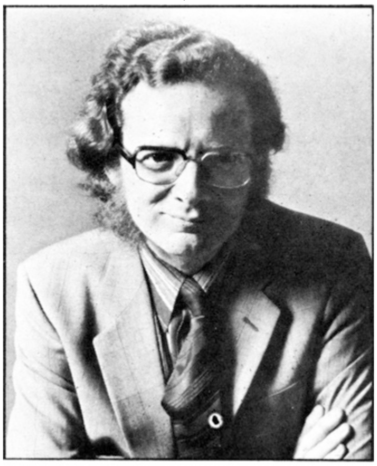
In recent years, this selfsame material has been at the center of increasingly acrimonious public and parliamentary controversy in Britain. The reason: farmers now sow over 70 percent of their cereals and oil-seed rape during the autumn, which means burning off stubble from the previous crop. The resulting conflagrations have caused anger, pollution, and road accidents throughout the country. But the only feasible alternative—ploughing in the straw—can lower cereal yields considerably. Particularly during wet years, stubble decomposing anaerobically in the soil generates toxins such as acetic acid that impair germination and retard the growth of roots and shoots. As a result, output falls by as much as 20 percent.

Is there a third option? That was the question that led Dr. Lynch to investigate a microbiological solution, such that straw could be exploited as a valuable resource rather than be disposed of, destructively or otherwise, as an unwanted by-product. Part of the difficulty with the natural decomposition of straw stems from its huge (130:1) ratio of carbon to nitrogen compared with that of microorganisms (about 10:1). This means that primary breakdown is usually incomplete and nitrogen-limited. Maybe a microbial cooperative would do the trick—one organism to break down the cellulose, releasing nutrients for a nitrogen fixer that would then raise the content of this essential element in the decaying material?

The Letcombe scientists decided to proceed on two fronts. They would scrutinize the breakdown of crop residues in nature, and then try to improve upon what they found by selecting the most desirable combination of organisms. Nitrogen fixation in rotting leftovers did, in fact, prove to be quite common. But most N-fixing heterotrophs cannot use cellulose, or the accompanying hemicellulose or lignin, as sources of carbon and energy. What actually happens in some situations is that they become associated with cellulolytic fungi which, as strikingly effective partners in symbiosis, provide them with nutrients. Because nitrogen fixation is most efficient in anaerobic conditions, and cellulolysis most rapid in aerobic conditions, the process tends to concentrate at interfaces between the two types of environment—usually in semi-waterlogged soil.

But could profitable biotechnology be built upon these ecological insights? Dr. Lynch and his collaborators experimented by chopping wheat straw, packing it into columns, and inoculating the system with soil, to assess the range of microorganisms emerging naturally. They found a wide variety of cellulolytic fungi, including species of *Penicillum*, *Fusarium* and *Trichoderma*, but only a single N-fixer—the anaerobic bacterium *Clostridium butyricum*. When studied in pure culture one of the fungi, *P. corylophilum*, decomposed straw polysaccharides (cellulose and hemicellulose) only when nitrogen was added. Likewise *C. butyricum* failed to multiply or fix nitrogen in pure culture. But if the two were cultured together the bacterium did grow and support cellulose breakdown by the fungus, presumably by providing a source of fixed nitrogen.

This carefully-selected bio-community is highly efficient, increasing the rate of cellulolysis and producing about 5 kg of nitrogen per metric ton of straw. In other words, it both accelerates the conversion of straw into compost and raises the nitrogen content handsomely. Full details of the work are due to appear in the *Journal of Applied Bacteriology*, but publication has been delayed while the British Technology Group (BTG) secures patent protection on this potentially invaluable idea. The Agricultural Genetics Company (in which the BTG has a stake) seems set to fund further research, and the first field trials will take place during the coming summer.


Bernard Dixon, Ph.D.

Two immediate possibilities are on the agenda for investigation. One is that farmers could spray a mixed culture of *P. corylophilum* and *C. butyricum* onto the straw as it lays in their fields. They might do this as part of the routine cropping operation, using suitably adapted combine harvesters, or perhaps as a separate procedure afterwards. Dr. Lynch's studies suggest that one year's straw would produce as much as a quarter of the nitrogen required by the next season's crop—giving a splendid saving on (increasingly expensive) fertilizers.

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—Christopher G. Edwards

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The alternative is a more sophisticated procedure. Straw could be baled, as at present, and then be inoculated with the mixture of fungus and bacterium before being incubated under controlled conditions. The product would be a rich compost for use in horticulture.

But BTG chiefs no doubt have wider applications in mind, too. It may well be that the Letcombe discovery holds promise for the profitable conversion of straw and similar wastes into fertilizer at many other places and times than in stubble-burning Britain. Either way, this looks like being an elegant success for ecological thinking and the Selman Waksman approach to microbiology. ◪

FINAL WORD (Continued from page 192)

claim, even if the compound is obtained by a radically different synthetic approach. Under the patent law, the courts have held that an infringer is one who derives his own plants from those of the patentee, i.e., only clones infringe. One commentator has suggested that once the patentee has proven the similarity of the two plants and the defendant's access to the plaintiff's plants, that it be up to the defendant to establish his innocence of infringement by showing that his development was independent. Others have suggested that the judicial decisions inferring a "derivation" requirement were wrongly decided and have called for the elimination of that requirement.

A recent Patent Office Board of Apeals decision, *ex parte* Jackson, could have, if accepted by the courts, the practical effect of limiting the scope of claims to novel microorganisms to organisms derived from the deposited cultures, regardless of their taxonomic similarity.

The last major revision of the substantive patent law for chemical, and mechanical inventions occurred in 1952. The following year, James Watson and Francis Crick proposed a model for the physical structure of DNA, and thereby laid the groundwork for the molecular genetics industry. Clearly, the legislators did not have an opportunity to think about the problems of patenting DNA sequences or genetically engineered microorganisms when they drafted the 1952 statute.

The Plant Variety Protection Act (PVPA) of 1970, on the other hand, was written with classical plant genetics in mind. For that reason, despite its limitations, we may point to it as a model for a biological patent statute. The most attractive feature of the PVPA is its approach to the definition of a "new variety." Instead of the traditional patent requirements of novelty, utility, and nonobviousness, these are instead requirements of distinctness, uniformity, and stability. These concepts may be applied, not only to plant varieties, but also to animal varieties, cell lines, and microorganisms.

We may also commend the drafters of the PVPA for expressly allowing plant breeders to engage openly in experimental testing of seeds, without fear that they will lose the right to file a patent application. Under the utility patent law, there is a statutory bar to filing after one year of "public use." While there is also a judge-made exception for the experimental use of an invention, it is difficult for inventors to determine when they are protected within the exception. The Plant Patent Committee of the American Bar Association has expressed its concern that, since it is common to test-grow all new plant varieties, normally in open fields, this conventional testing might be regarded as public use under the general patent statute.

Another issue is the significance to be attached to written descriptions of a new organism. An early plant patent case held that a plant patent claim could not be anticipated by a mere catalogue description, and a microbiological case held that the use of a novel strain in a fermentation process could not be *prima facie* "obvious" if the strain were not available from a depository. The PVPA, however, makes a catalogue description effective as a reference if it clearly indicates a source from which a specimen of the new variety may be obtained.

The PVPA has its weaknesses, too. For example, it is not a model of legislative clarity when defining the protection afforded by a Plant Variety Protection Certificate. In particular, the farmers' exemption to the general infringement provision is both verbose and confusing.

Also the PVPA is concerned purely with the "production of a variety by seed." It is not really prepared to cope with the potentialities of "genetic engineering" at the molecular level as a means of obtaining new varieties, and offers no starting point for appropriate protection for genetic engineering techniques and DNA sequences themselves.

Still, the 1970 act was a significant step toward the broader goal of tailoring the patent system to encourage innovation in the field of biological invention. Scientists, patent lawyers, and businessmen interested in the future of biotechnology, working both individually and under the aegis of organizations like the Industrial Biotechnology Association and the Association of Biotechnology Companies, must lay the groundwork today for the law that will govern biological inventions in the future. ◪

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