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EDITORIAL

This seems to be the age of biotechnology or genetic engineering or molecular biology or called what-you-will. In almost every issue of many scientific magazines and newsletters as well as trade and business magazines, there are always some reports of breakthroughs and advances made in this area. There has also been an endless spate of meetings and conferences held on this subject in the past few years, but it has become a bit quieter these days. Perhaps most of these people are settling down to serious work after all the "sales-pitching".

Many a research institute and university has its own biotechnology/molecular biology unit including PORIM and Unilever. Undoubtedly this is an exciting field, and perhaps most important it is an excitement which appeals to politicians, administrators and industry bosses, (rather than some "crummy" field trials) resulting in greater availability of R & D funds. Is it any wonder that many has hopped onto this "bandwagon" or "gravity-train"?

Perhaps let us join the "bandwagon" and devote this issue to biotechnological news and what biotechnology has to offer or influence our field of work. In December there was a world conference on biotechnology of fats and oils held at Hamburg, and attended by many scientists from our industry. Dr. Cheah, Suan Choo, biotechnologist at PORIM has graciously provided us with a short write-up on the highlights of this conference. Her paper presented at the 1987 International Oil Palm Conference is reproduced here as a feature article. The abstract of her paper presented at the SABRAO International Symposium & Workshop on Gene Manipulation in Plant Development in Developing Countries is also included. Perhaps at some future date, she can also oblige us with a write-up on the biotechnology R & D thrusts at PORIM's new Plant Science and Biotechnology Unit. We should perhaps also get Dr. W. de Greef, formerly oil palm breeder at Unilever Plantations in Zaire and

presently with Plant Genetic Systems, a plant genetic engineering firm in Belgium, to share his thoughts on what biotechnology/genetic engineering has to offer to the oil palm. Included in the biotechnology news, are notes on companies and projects involved in the biotechnology of oils and fats.

To give some counterbalance to the positive bias on biotechnology on the issue, I have extracted certain relevant sections of "Issues forum: Biotechnology and patenting" from the congressional update section of Agronomy News, and also included the abstract of Rasmusson's paper on his twenty-years' experience in ideotype breeding in barley, as one of the key pitches of plant molecular biologists is the ability to engineer super or ideal crop plants with the new genetic engineering techniques.

Editor

Feature Articles

A. WORLD CONFERENCE ON BIOTECHNOLOGY FOR THE FATS AND OILS INDUSTRY

The World Conference held in Hamburg, West Germany, 27 September - 2 October 1987, attracted about 500 participants from 40 countries. It was jointly organised by the American Oil Chemists' Society (AOCS) and the German Society for Fat Research (DGF). More than 50 plenary lectures were delivered by invited speakers from the U.S., Japan, Canada, Australia and Europe. There were 41 posters presented in 3 sessions on the biochemistry and genetics of plant lipids, lipases and industrial applications. Accompanying the conference was an exhibition by 20 companies featuring products and services for biotechnological applications as well as mainstream processing equipment for the fats and oils industry.

The main emphasis of this conference was on biotechnological approaches for the modification of oils and fats via genetical and enzymic means. The discussions covered plant, animal and microbial lipids.

Genetic Engineering of Oil Crops

The keynote addresses on the opening day had Professor Paul K. Stumpf (University of California - Davis, USA) delivering a lecture on the basic biochemistry of plant lipid biosynthesis and Dr. Colin Ratledge (University of Hull, U.K.) speaking on the same topic but in microbial systems. In the opinion of the speakers, there is sufficient basic knowledge to initiate studies in the gene manipulation of lipid synthesis in plants by in vitro methods. Several biosynthetic reactions in plant fatty acid synthesis were identified as "rate-limiting" by Dr. D. Guerra (Biotechnica Canada). He suggested that these may be the targets for genetic manipulation for increasing oil yield. However, Dr. Ratledge cautioned against such an idea as it has been shown in bacteria that changing rate-limiting steps in lipid biosynthesis was ineffective in increasing yield. A change in one rate-limiting reaction usually has cascading effects on the reactions that follow.

The only protein in the fatty acid biosynthetic pathway in plants that has been studied at the molecular level is the acyl carrier protein (ACP). In his paper, Dr. John Ohlrogge (Michigan State University, USA) reported that his group had successfully synthesized and cloned an oligonucleotide probe for an ACP gene. Using this probe, he measured that ACP messenger RNA (mRNA) levels during seed development in soyabean. This was compared to the mRNA levels for the seed storage protein lectin, another gene which is switched on during seed development. The results showed that the mRNA for ACP accumulated at high levels before that for lectin. This demonstration of temporal regulation of gene expression has relevance to attempts to use the promoters of storage protein genes to drive fatty acid biosynthetic genes in transgenic plants, as is being done by some groups. These promoters may not be the right ones after all even though they ensure tissue specific expression. Some preliminary data was also presented to show that enzyme specificity is the likely mechanism for the targetting of fatty acids in triacylglycerol synthesis. This result is indeed encouraging for those endeavouring to genetically manipulate oil composition for it would appear that the genes of these enzymes could well provide the "handle" for this purpose.

Several papers were given by plant breeders who argued that it may not be necessary to resort to in vitro gene manipulation methods for changing oil composition. Dr. G. Robbelen (Georg-August University, FRG) considered the new gene technology as only a little better than mutation breeding. As an alternative, the breeders recommended that new genetic resources for oil crops be exploited. An oil crop which has been identified as having great potentials for the production of mid-chain fatty acids is cuphea. From studies made so far, it was concluded that cuphea will probably not be commercialised as an oil crop. However, there is interest in introducing the genes of cuphea into rape.

Tissue Culture of Palms

Two papers were delivered on the above topic. A representative of Kao Corporation (Japan) spoke on the efforts made to tissue culture the coconut palm. Attempts to regenerate plantlets from calli derived

from the meristem of the male inflorescence have so far proved to be unsuccessful. Direct embryogenesis appeared to be a more promising route for the clonal propagation of this palm.

In the oil palm, Dr. L.H. Jones (Cambridge University, U.K.) discussed several aspects of the work that had been carried out at Unilever when he was with the company. It was said that the field trials are still not sufficiently long enough to give actual increase in yield in the clonally propagated palms. The speaker elaborated upon the importance of selecting the right clone for a particular environment. With regards to the abnormal fruits produced by some clones, Dr. Jones claimed that they have been able to correlate a number of changes made at the factory as the causes giving rise to the abnormality.

New Novel Oil Crop Varieties

High Oleic Sunflower

Sungene Technologies Corporation (California, USA) reported their success in developing varieties of sunflower that produce oils with over 95% oleic acid. It was claimed that Sungene produced these for a client who will control its distribution. From their experience, mutation breeding did not give any fatty acid mutants but cell culture was able to generate both high and low oleic acid lines.

High Linoleic Linseed

Dr. A. Green of CSIRO (Canberra, Australia) spoke at the conference on the efforts made to convert linseed oil from an industrial to an edible oil. For this purpose, his group used the mutagen, ethyl methanesulphonate (EMS), to induce low linolenic acid mutants in the flax plant. By screening for mutants with reduced desaturase activity, they isolated two mutants which had low linolenic and high linoleic acid contents. They have developed a "zero genotype" with 3% linolenic and 63% linoleic by crossing plants carrying these mutations in the Gleneig variety. At trials conducted near Canberra,

this variety produces a yield of 1 tonne of oil per hectare. Currently, studies are being made on the effect of growth temperature on the fatty acid composition and the oxidative stability of the oil produced.

Soyabean

The USDA and Purdue University (USA) have used mutation breeding to produce high stearic and high oleic mutants in the Century line of soyabean. It was found that there is a reciprocal relationship between oleic and linoleic acid contents. For better oil stability, low linolenic lines are being developed. In the effort to improve oil flavour by reducing lipoxxygenase (LOX) activity in soyabean, work is currently being carried out at the molecular level to determine the LOX isozyme responsible for the development of the "grassy flavour". Results showed that the isozyme L-2 may be the enzyme involved. There are also efforts being made to use recombinant DNA techniques to engineer the storage proteins of soyabean.

Enzymic Modification of Lipids

In his keynote address, Dr. T. Yamane (Nagoya University, Japan) spoke on the applications of lipases, phospholipases and lipoxxygenases in the modification of oils and fats. He introduced the term "micro-aqueous" to describe the system for lipase action in which only trace amounts of moisture is present. The effect of moisture on lipase-catalysed reactions was demonstrated with respect to enzymic glycerolysis and aminolysis. It was claimed that micro-aqueous environments conferred operational stability to the reaction and increased yield and selectivity. The engineering aspects of micro-aqueous systems were discussed in relation to application in batch, fluidised bed and membrane reactors.

An interesting new concept introduced at this conference was the continuous use of lipase for fat splitting without the need for enzyme immobilisation. In the system described by Dr. M. Buhler of Henkel (Dusseldorf), the process was carried out in a multistage set-up in which hydrolysis occurred in the first reactor followed by separation of the reactants in a second reactor. In the latter, the reaction

mixture was allowed to separate into the glycerol/water phase and the oil phase. The lipase collects at the interphase, and therefore, by recycling this phase into the first reactor, one effectively recycles the enzyme. The advantage of such a system is that one can do away with the cost of immobilisation and the problems associated with the process. Dr. Buhler mentioned that the system has been tested in a pilot plant consisting of a two-stage mixer-settler set-up capable of producing 171 kg of fatty acids per day.

Single Cell Oil

Several speakers touched on the topic of producing oil by microbial fermentation. Single cell oil (SCO) production from bacterial, yeast, filamentous fungal and algal sources were described. It was concluded that the process will only be economical for the production of specialty oils. The recently commercialised production of a gamma-linolenic acid-rich oil by fungal fermentation was quoted as an example of such a premium value oil. Dr. N.K.H. Slater of Unilever (Vlaardingen, The Netherlands) gave the estimated cost of production of SCO as Dfl. 10/kg (that is, about M\$12/kg). At this price, it will not be economical to produce commodity oils. However, the use of waste products as feedstock may reduce the cost of production. It was cited that New Zealand has developed a process for the production of a cocoa butter-like fat using yeast fermentation of whey. This process is being tested at the pilot plant stage. There appears to be much controversy as to what this product should be labelled as.

Commercialising Biotechnology

On the subject of commercialising biotechnology for the fats and oils industry, Dr. R. Dull of Experience, Inc. (USA) said in his lecture that the process will be very complicated in view of the fact that this market is already complex with the high degrees of substitution possible. The important economic considerations for commercialising any biotechnological product in the fats and oils market are:

- (a) one has to look carefully at the foreign exchange needs of the 80 exporting countries,
- (b) livestock cycles,
- (c) import regulations, and,
- (d) government subsidies.

The speaker posed the interesting question as to whether the oil palm should be engineered to produce high oleic acid oil for better nutritional value and for better competition with soyabean oil.

Regulatory Aspects of Biotechnology

In the last session of the conference, some regulatory aspects were discussed with respect to:

- (a) toxicological evaluation of biotechnological products,
- (b) regulations and enforcement in Europe, USA and Japan, and,
- (c) patenting.

Dr. W.E. Parish of Unilever (U.K.) said that biotechnology has become a "bogey" word due to lack of understanding on the part of the public. He stressed the need to differentiate between "perceived risk" and "real risk". In order to do this, there was the need for toxicological evaluation. He then went on to explain the three phases of examination of a biotechnological process:

- (a) identification of the properties and potential pathogenicity,
- (b) evaluation of the crude product, including the enzymes used, and,
- (c) evaluation of the final product.

Representatives from Europe, USA and Japan discussed the regulations imposed by each region for controlling biotechnological developments. It appears that Europe has no uniform approach to regulatory control of recombinant DNA products as there is no directive from the EEC on this issue. The situation appears to be better

organised in the USA. There is a Biotechnology Science Co-ordinating Committee that looks into regulatory aspects of biotechnology. Currently, the Committee's main activity is to monitor experiments dealing with the release of recombinant organisms into the environment. In Japan, four ministries are involved in regulating biotechnological developments.

The aspect of patenting genetically engineered plants was discussed by Dr. K. Downey (Agriculture Canada). Currently, plant breeders' rights (PBR) are taken care of by the International Union for the Protection of New Varieties (UPOV) which provides Plant Variety Protection (PVP). However, with the advent of non-conventional means of plant breeding, there is now the possibility that some investigators may resort to patenting. The speaker expressed the fear that such a trend may lead to problems later on. As an example, he cited the recent patenting of the high oleic sunflower variety. According to the speaker, in doing so, the investigator had practically patented all the genes involved in the synthesis of oleic acid. He suggested that there should be guidelines for the patenting of precisely described specific genes only. Protection of the variety itself should still come under UPOV.

Nutritional Aspects of Oils and Fats

During the World Conference, two lectures were given on the nutritional effects of lipids in the diet.

In her acceptance speech for the Normann Award at the DGF Meeting, Dr. Joyce Beare-Rogers of the Department of Health and Welfare, Ottawa, Canada, discussed the nutritional attributes of fatty acids. The studies carried out on the anti-thrombotic effect of eicosenoids (EPA) in Eskimo populations were described. There was a call for more investigations into the mechanism of this phenomenon. She also warned that one should be careful about diets high in polyunsaturated fatty acids. An interesting point raised was why saturated fats in infant diets are not causing cardiovascular effects.

In the lecture delivered by Dr. K.K. Carroll (University of Western Ontario, Canada), the tumourigenic effect of high fat diets was discussed. His studies showed the relationship between high intake of polyunsaturated fatty acids (PUFA) and tumour progression in experimental animals. However, epidemiological studies showed no correlation between breast cancer and PUFA. On the aspect of fatty acid in the diet and cardiovascular disease, the speaker concluded that monounsaturated fatty acids are just as effective as PUFA in reducing blood pressure. As PUFA-rich oils are easily oxidised, he expressed concern for large intake of these oils by the public.

Cheah, S.C.

PORIM

B. DEVELOPMENT OF DNA PROBES FOR THE OIL PALM

Cheah Suan Choo*

ABSTRACT

Genetic studies in the oil palm have been impeded by its long breeding cycle. As a result, there is little information on single gene loci in the palm genome even though several single gene mutations affecting fruit and vegetative characters are known. The advances made in recent years in molecular cloning techniques permit one to probe for specific deoxyribonucleic acid (DNA) sequences in the genome. The application of these techniques to the oil palm DNA should help overcome the problem of its long breeding cycle since progeny analyses can be made as early as the embryo stage. We are developing restriction fragment length polymorphic (RFLP) markers for the oil palm genome with the aim of using these, in the short term, for monitoring genetic uniformity in tissue culture, and, in the long term, to map single gene loci in the palm.

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INTRODUCTION

The advent of recombinant DNA techniques has made possible new methodology for genetic studies. Among these new developments is the technique for introducing parts of an organism's genome into yeast, bacterial or viral cells so that these genomic parts or DNA sequences can be multiplied within the microorganism for large scale preparation. The ability to generate large quantities of unique sequences in this manner has led to their use as genetic probes to complement biochemical and phenotypic markers in genetic analysis. These DNA probes can be either sequences of known genes or fragments of DNA with no known function. This paper describes the development and application of probes of the latter kind for the oil palm.

RESTRICTION FRAGMENT LENGTH POLYMORPHISM (RFLP)

Restriction endonucleases are enzymes which cut DNA at specific sites (termed restriction sites) to generate fragments which can be separated according to size by gel electrophoresis. In large, complex genomes, such as in plants, these fragments are seldom detectable as discrete bands. However, certain fragment bands can be highlighted by hybridising these with a labelled DNA probe. A characteristic pattern of bands is therefore generated for each set of restriction fragments when hybridised with a particular DNA probe. Genetic changes such as base changes, additions, deletions, inversions and transpositions can alter the banding pattern if these changes abolish restriction sites, create new restriction sites or change the distance between two restriction sites. This variability in the banding pattern of restriction fragments is known as restriction fragment length polymorphism, or RFLP.

RFLPs are inherited as simple codominant Mendelian genes and will therefore serve as useful genetic markers. Since they occur in the coding as well as the non-coding regions of the genome, it is evident that this type of polymorphism has the potential of creating a greater number of genetic markers than isozyme polymorphism. RFLP analysis has two components, namely, an array of restriction enzymes and a variety of DNA probes. The combination of these two further enhances the sensitivity of this technique.

The use of RFLPs as genetic markers was first proposed for the study of the human genome (Botstein et al., 1980), an application which has proven to be useful in the location of heritable disease gene loci (White et al., 1985; Cooper and Schmidtke, 1986). The commercialisation of RFLP probes in medicine is well under way (Van Brunt and Klausner, 1987). The concept of RFLPs as genetic markers should also be applicable to plants (Beckmann and Soller, 1986a). RFLPs have been reported for maize (Helentjaris et al., 1985; Evola et al., 1986), tomato (Helentjaris et al., 1985; Bernatsky and Tanksley, 1986), wheat (Muthukrishnan et al., 1985), peas (Domoney et al., 1986) and lettuce (Landry et al., 1987).

The degree of RFLP detectable in plants varies from species to species. A high degree of polymorphism is known to occur in the genome of maize (Riyin et al., 1983). The ability to detect RFLP in tomato, peas, beans and sunflower was, however, much reduced (Helentjaris et al., 1985; Beckmann and Soller, 1986b). It is suggested that the difference in the level of genetic variability in maize and the other crop plants may be due to the fact that maize is an outcrossing species while the others self-fertilise. Alternatively, this phenomenon of high variability may be a consequence of the transpositional events that commonly occur in the genome of maize (Walbot and Cullis, 1985). Isozyme polymorphism has been detected in the oil palm (Elaeis guineensis) in 15 loci (Ghesquiere, 1984; Ghesquiere and Meunier, 1986). Since RFLP probes have the potential to extend the number of loci to be tested for polymorphism, it will be interesting to test the hypothesis that outcrossing species show higher degrees of RFLP. The oil palm, like maize, is an outcrosser.

SELECTION OF RFLP PROBES

In RFLP analysis, one can use probes derived from a random complementary DNA (cDNA) or a genomic library. There is some debate as to which of these two libraries is the better source of RFLP probes (Beckmann and Soller, 1986b). cDNA libraries are particularly useful as they contain clones of single or low copy number gene sequences. Since the genome of most eukaryotes contain repeated DNA sequences, genomic libraries will have to be screened for clones which do not

contain such sequences. Both types of libraries have been employed by various researchers for the screening for RFLP probes. Whilst cDNA probes will only detect polymorphism within coding sequences of the genome, genomic clones are able to expand into the non-coding regions. In our experimental approach, we will use both cDNA and genomic libraries.

APPLICATIONS FOR RFLP PROBES

We are developing these DNA probes for several purposes. In the short term, we will use them as markers for monitoring genetic uniformity in tissue cultured oil palms. These markers will also be tested for linkage to the monogenic traits of shell thickness (sh locus), anthocyanin (*virescens*) and carotene (*albescens*) pigmentation, and the genetically mantled character (*Poissoni*). Linkage of these characters will then be extended for the long term mapping of agronomically important gene loci in the palm.

There are indications that RFLP probes can be used for varietal identification (Evola et al., 1986). This application deserves some attention in the oil palm. In view of the availability of clonal propagation techniques, there is the need to protect "breeders' rights".

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C. RNA EXTRACTION FROM OIL PALM TISSUES

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As a preliminary to cDNA cloning, total RNA has been extracted from various tissues of the oil palm (Elaeis guineensis). A modified Kirby salts solution was used as the extraction buffer. In the RNA extraction procedure, proteins were precipitated with phenol followed by salt precipitation of the RNA in either 2.23M sodium acetate buffer, pH 6.0, or 2M, lithium chloride. The yield and purity of the RNA obtained were found to be variable from tissue to tissue. Attempts are being made to obtain amounts of RNA in quantities large enough for the isolation of messenger RNA.

Abstract of paper presented at SABRAO's International Symposium and Workshop on Gene Manipulation for Plant Improvement in Developing Countries Kuala Lumpur, 1987.

NEWS

SABROA's International Symposium and Workshop on Gene Manipulation
for Plant Improvement in Developing Countries - Kuala Lumpur Nov.
30 - Dec. 3, 1987

This Symposium was expected to draw some of the leading international tissue-culturists and molecular biologists as speakers. It did manage to draw most e.g. Bajaj, Rhagavan, Hu han, Bhojwani, Lorz, Gerlach but a few very prominent speakers i.e. Murashige, Vasil, Helentjaris and Montague did not turn up for one reason or another. There were also some breeders among the speakers i.e. Mayo, Brar, Rajanaidu to give some balance to the theme.

A pleasant surprise was Dr. de Greef who replaced Dr. Montague as the speaker. Dr. de Greef until very recently was the oil palm breeder with Unilever in Zaire. Presently he is together with Dr. Montague at Plant Genetic System, a plant genetic engineering firm in Ghent, Belgium. It must have been a very drastic change from working at a very macro level in a low tech environment in the African bush to a micro level in neat sterile high-tech laboratory in Ghent, Belgium!

Participants were mainly from the countries neighbouring Malaysia i.e. Singapore, Thailand, Indonesia, Philippines, India although there were also representatives from Kenya and Libya. Most of the Malaysian oil palm breeders and tissue-culturists attended the Symposium.

Biotechnology Update

1. Biotechnology Upgraded Oils

Vegetable oils command a world market of over US \$35 billion with a US\$4 billion U.S. market. The bulk of the oils are of low value e.g. palm oil, rapeseed oil, soyabean oil and sunflower seed oil. Scientists have begun to tap this vast potential using biotechnological methods to improve the properties and thus the value of oils and fats.

The two approaches adopted are the genetic modifications of oil seed plants to induce production of altered oils, involving genetic engineering, in vitro mutagenesis and tissue culture techniques; and the enzymatic or microbial modification of the oils and fats themselves.

Rapeseed (canola) will be the forerunner for genetically modified oils because it is easiest to genetically engineer, has the lowest saturated fat content, and the most efficient crop yielding 40% of its weight in oil. Genetic engineering may also extend its range of cultivation. Oil palm, being a perennial tree crop, will be the most difficult to genetically engineer.

Enzymatic modification of oils will use the least expensive oils as substrates, giving palm oil an advantageous position. However to be profitable, applications will have to be found that give a value increase of 25-50% over the price of unmodified oils.

70% of the oils and fats are used in edible products. Improvement in this area will include lowering saturated fat levels, lowering caloric value and reducing linolenic acid content. Vegetable oils also have industrial applications such as lubricants, detergents and plastic. The new technology would enable production of high value products e.g. esters, high purity monoglycerides, diglycerides, specialty detergents, emulsifiers, specific fats or oils. Tissue culture products are already in the market while enzymatic modification productions will appear in the market within the next five years.

Below is a sample of companies working on oils and fats modification.

- Calgene (California) - Genetic engineering of rapeseed for both edible and industrial oil markets.
- Lubrizol (Ohio) - Modifying sunflower, rapeseed and corn plants to upgrade oils. Collaborates with Sungene Technologies and others. Marketing high oleic acid sunflower seed oil.

- Sungene Technologies Corpn. (California) - Tissue-culture of corn, soyabean, rapeseed, sunflower and sesame. Developed high oleic acid and high linolenic acid sunflower strains.
- Biotechnica International (Canada) - Genetic manipulation of rapessed and flax to modify fatty acid composition.
- Unilever - Studies genetic modification and enzymatic processes. Produces cocoa substitute semicommercially.
- Ajinomoto Co. (Japan) - Enzymatic conversion of diglycerides to tryglycerides or replacement of one fatty acid with another.
- Asahi Denka Kogyo (Japan) - Enzymatic processes to produce cocoa butter substitute from palm oil.
- Cetus Crop. (California) - Has patents for use of enzymes to modify oils and fats.
- Genencor Inc. - Protein engineering of enzymes for use in modifying oils and fats.
- Fuji Oil Co. (Japan) - Has patent for use of lipase to make cocoa butter.
- Henkel Research Crop. (California) - U.S. research centre of German company. Uses molecular biology to develop microbes for oil and fat modification.

2. Plant Yield Enhancers

Biotechnica has genetically engineered symbiotic Rhizobium strains which have nitrogen fixation abilities. Regulatory approval was delayed because Biotechnica's system for monitoring the microorganisms employed a genetic marker carried by many natural soil microorganisms. The company has developed a new monitoring system based on a different marker.

Allelix's (Canada) approach was to screen for the best root associated bacteria, and then use them to inoculate the soil or seed to increase the number and mass of nitrogen fixing nodules to as much as 100%. From trials in Canada, U.S. and U.K., yield increases as much as 30% in soybeans and other legumes have been obtained. Genetic modification of the microbes could even boost crop yield higher and additional genes for production of toxins against pests could also be incorporated. Allelix is researching on the best method to deliver the inoculant to seeds or soil. Some of the effects do not even require the presence of the microbes but could result from spraying fermentation broth or some chemical derived from it into soil.

3. Biopesticides

Mitsui Chemicals (Japan) has developed a faster and easier process of producing insecticidal protein by using genetically engineered Bacillus subtilis instead of B. thuringensis.

With B. thuringensis the insecticidal protein is produced only during spore formation, and the spores have to be killed subsequently as they affect helpful insects. With the new process, production of the protein can be done without spore formation thus saving time and cost.

Ecogen (USA) will be marketing its biofungicide trade-named "Dagger G" to combat damping-off disease in cotton. The biofungicide is a selected Pseudomonas bacteria strain which works by chelating iron in the soil, depriving the fungi that cause the disease of this essential trace element.

Issues on Biotechnology and Patenting

'Biotechnology' is enduring as a magic word in the halls of Congress and in university administrative offices across the nation. Funding biotechnology is much more exciting than funding variety trials, or field studies on disease or pest resistance. The general public isn't quite sure what those are for anyway. A few years down the road we may find ourselves in the amusing position of having a biotech research center at every land grant college, but with many of the schools lacking a breeder that knows how to raise plants out in the field. The new tools of biotechnology research are, of course invaluable, and research on the application of biotechnology to agricultural production should be pursued. However, traditional breeders and agronomy departments will need to be vigorous in communicating the fact that these techniques are tools to be used in conjunction with breeders.

Patenting is an important tool in stimulating research and development and technology transfer. Patents allow investors to benefit from their efforts, protecting their right to a return to their efforts while moving the product rapidly into the market place. Patents may be granted on any new, useful and nonobvious composition of matter, or article of manufacture, machine or process. In 1980, the Supreme Court held that the mere fact that subject matter is "living" does not render it unpatentable. However, "products of nature" are not patentable because they lack novelty. A claim to the entire genetic material of a single cell would be rejected; but one may properly seek a patent on an isolated gene encoding a protein of interest.

In its new book, "Agricultural Biotechnology: Strategies for National Competitiveness, the National Research Council notes that biotechnology patents make up about 2 percent of the patents granted. Of these patents, about 40-45 percent are awarded to foreign parties, about 40 percent to U.S corporations and 18 percent go to U.S universities, government agencies, nonprofits and individuals. Biotechnology patents may constitute a large portion of the patents received by universities, ranging as high as 37 percent at the

University of Wisconsin. There is however, a considerable disparity in the number of universities receiving such patents. In 1985, seven universities obtained half of the biotechnology patents granted to all U.S universities; the University of California, MIT, the University of Wisconsin, Stanford, Harvard, Cornell and Purdue.

In addition to the difficulties with technology transfer, universities will have to struggle with their historical preference for free information flow. For plant breeders, the Plant Variety Protection Act of 1970 provided protection for the development of particular genotypes while allowing the free exchange of germplasm from cooperating programs. Ian Edwards of the National Wheat Improvement Committee warned in Sept. 5, 1987. Genetic Engineering News, that ready access to germplasm may be restricted as groups begin to patent traits within their materials.

As we move toward patenting as a mechanism of protecting research interests, we are also moving toward litigation as a mechanism for decision making. We may be moving toward an endless eries of court cases that may hamper and prevent research. As a further complication, few geneticists or breeders are fluent in the legal process or the legislation that underlies it. Likewise, few in the legal profession are fluent in biological realities underlying plant breeding and the new biotechnology. Among the difficult questions that may need to be addressed: Is a patent issued for a trait; or for the DNA that codes for the trait? If it is for the trait, as some argue, what happens when an alternative combination of genes express a patented trait? If the patent is for a gene, does a gene on another chromosome that is involved in the expression of the trait require a separate patent? "

An Evaluation of Ideotype Breeding¹

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ABSTRACT

Plant breeders have attempted to enhance yield by selecting for individual traits since the beginning of plant breeding. This approach has been broadened to encompass the breeding of model plants or ideotypes. An ideotype is hypothetical plant described in terms of traits that are thought to enhance genetic yield potential. Ideotype breeding is defined as a method of breeding to enhance genetic yield potential based on modifying individual traits where the breeding goal (phenotype) for each trait is specified. The purposes of this paper are to elaborate on and evaluate ideotype breeding as a method to enhance genetic yield potential and to describe an ideotype for barley (Hordeum vulgare L.). Successes that have occurred in breeding to enhance yield with individual traits, the value of genetic diversity for individual traits, and benefits from goal setting are presented as arguments in support of ideotype breeding. Alternatively, information is presented on the requirement of symmetry in size among plant parts, compensation among plant parts, pleiotropy, and genetic background, all factors that may slow progress in ideotype breeding. Ranges of genetic diversity, heritability estimates, and introgression information are presented for 27 barley traits. A barley ideotype consisting of 14 traits and the target or goal for each trait are described. Ideotype breeding is recommended as a methodology to augment traditional plant breeding, when the breeding goal is enhancing genetic yield potential. Breeding experience and research to date suggest that ideotype breeding is not a suitable substitute for traditional yield breeding.

Additional index words: *Hordeum vulgare* L., Yield breeding, Plant design, Plant architecture, Plant breeding methods, Model plants, Small grains.