

## BIOCHEMICAL GENETICS: APPLICATIONS IN AGRICULTURE

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Biochemical genetics deals with the genetic control of the biochemical and physiological processes in the living system. As a discipline, therefore, it is more basic than applied. However, one finds that it has many immediate applications in many applied disciplines one of which is that related to food production.

1. *Plant and Animal Improvement.* It is a natural consequence that the findings in biochemical genetics will be of immediate use in plant and animal breeding. If one will take a closer look at the activities of the plant and animal breeders, he will immediately see that all that which they are hoping to improve are products of a series of biochemical reactions and each reaction is controlled by one or more genes, e.g.

(a) *Grain yield.* This is an accumulation of the starch, protein, fatty acids, vitamins, etc. that were synthesized by the plant and stored in the grain or kernels. All the genes involved in these biochemical pathways constitute the yield genes, and understanding these will definitely help the plant breeder not only in planning his breeding strategies but also his screening and evaluation procedures.

It is not only the quantity of the grain yield that are genetically determined. Quality is also influenced by the action and interaction of these genes which are reflected in terms of type of gene products that are finally synthesized.

(b) Resistance to pests and diseases may be manifested by the synthesis of specific metabolites. These resistance factors are generally gene products which may be easily identified. Screening for resistance may, therefore, use these metabolites as the selection indices. For example, in mungbeans an isozyme of peroxidase was observed to be associated with resistance to *Cercospora* leaf spot. Using the isozyme as a selection index, segregating populations in the mungbean hybridization program may be evaluated and resistant lines identified.

(c) The virulence factor produced by the parasite and its mode of action as well as the reaction of the host has been used in the breeding for resistance.

A tobacco pathogen, *Pseudomonas tabaci* produces a toxin structurally similar to the amino acid methionine. When tobacco

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tissue cultures were grown in the medium containing an inhibitory concentration of the methionine analogue, methionine sulfoxine, some variants resistant to *Pseudomonas tabaci* were produced. MSO is an analogue of the wildfire toxin which cause the formation of the same characteristic halo on the tobacco leaves as does the natural bacterial toxin. In this case, therefore, endogenous concentration of a specific metabolite, methionine, has been increased by selecting for resistance to the structural analogue, MSO.

- (d) It follows, therefore, that by using analogues of amino acids essential to human nutrition, variants of crop species which produce elevated levels of these compounds may be induced and selected in cultures, e.g. in rice.

Rice is of inferior nutritional quality because of low lysine, threonine, and isoleucine. In higher plants, these three essential amino acids and methionine are derived from aspartate. Using an analogue of lysine, S-( $\beta$ -aminoethyl) cysteine (SAEC), as the mutagenic agent, rice mutants resistant to SAEC were produced and these mutants have the lysine biosynthesis released from the regulatory control. Table 1 shows the improvement.

**Table 1. Free amino acid pools of rice tissue cultures (in nmoles/mg dry weight)**

AMINO ACID	WILD TYPE	EMS-5	EMS-6	EMS-8
Lysine	7.67	14.07	14.84	19.47
Histidine	10.76	6.21	5.97	6.96
Arginine	5.90	5.97	8.74	10.99
Glycine	6.89	11.17	17.27	17.89
Alanine	1.82	22.35	8.40	23.75
Valine	2.84	8.18	12.02	14.25
Methionine	0.71	1.86	1.80	2.06
Isoleucine	0.79	3.10	6.44	7.04
Leucine	0.80	3.88	6.26	6.67
Tyrosine	0.97	2.87	2.74	3.64
Phenylalanine	10.22	6.91	5.41	7.51
Total nmoles/mg dry wt.	104.10	182.32	149.31	193.06

When the total amino acid composition of the variants were compared to that of the normal, there was a definite improvement in protein quality as reflected by the increase in the relative levels of lysine, isoleucine, leucine and valine as follows (expressed as % moles of total)

	Wild Type	EMS-5	EMS-6	EMS-8
Lysine	5.19	6.54	5.86	6.26
Isoleucine	3.97	4.41	4.44	4.45
Leucine	6.79	7.87	7.88	7.81
Valine	6.37	6.94	7.42	7.23

(e) Measures of interaction between gene products may be a means of determining heterosis or hybrid vigor. In corn, Scandalios et al. showed the association of heterosis and the effects of intragenic and intergenic complementation on catalase structure.

Hybrid catalases between allelic genes of the  $Ct_{14}$  locus have been subjected to biochemical analysis. (specific activity, temperature sensitivity, photosensitivity, etc.). The heterotetramers generated by either intragenic or intergenic complementation exhibit improved physicochemical properties over the less efficient parental molecules. The hybrid proteins confer an advantage to the organism carrying them.

2. *Regulation of Biological Processes in Crops.* Knowledge of the genetic control of the physiological processes may facilitate the manipulation of these processes. For example, in *Arabidopsis*, flowering is enhanced by subjecting the plants to long day and higher temperature ( $28^{\circ}\text{C}$ ). This effect of day length and temperature is duplicated by bromodeoxyuridine. The mode of action is as follows:

Bromodeoxyuridine is a competitive analogue of thymidine. Since BdUR is an antimetabolite, its effect is based on selective interference with certain functions, i.e. BdUR is working against the flowering inhibitor. The mechanism of action of BdUR is through DNA. It preferentially inhibits DNA synthesis in the mitotically active cells of the vegetative apex, forming flank meristem. Consequently, the control initiation zone is activated, signaling the conversion of the vegetative apex into a prefloral apex. Upon elimination of the analog from the cells, mitotic activity may be resumed and floral development may thus take place.

3. *Food Processing.* Fermentation is one of the activities in food processing where knowledge of biochemical genetics may come in handy. Use of the biochemical mutants of yeast have been known to increase the alcohol yields in the wine industry. Certain qualities of the fermentation products are also attributed to the specific strain of the microorganism. For example, Champagne yeast strains give a definite alcohol quality quite different to that of other strains. In the improvement of the nutritional quality of some food items, the biochemical mutants of the microorganisms have been known to play an important role. For example, mutant strains that can synthesize high amounts

of lysine have been used for lysine production which in turn as food supplements.

4. *Genetic Engineering.* While the application of genetic engineering is yet to be seen in agriculture, the possibilities are quite obvious. For example, the transfer of the nitrogen fixing (*nif*) genes in bacteria into the crops that are heavy users of fertilizers would mean a reduction in production cost as well as the possibility of a more efficient use of the plant nutrients.

Successful intergeneric transfer of *nif* genes between prokaryotes has provoked widespread speculations on the possibility of constructing nitrogen-fixing eukaryotes. Before it is done, however, a great deal more knowledge is required about the genetic determination, regulation, physiological expression of *nif* genes in a very wide variety of microorganisms, as well as a great deal of new knowledge of plant physiology and genetics: for example, the regulation of *nif* gene expression, the uptake and incorporation of Mo and Fe, the protection of the *nif* gene products from oxygen, the synthesis of appropriate electron donor (such as ferridoxin or flavodoxin), the generation of ATP at an appropriate site, etc.

A more successful undertaking on gene transfers was that on sycamore using the bacteriophage as the carrier. The phage  $\lambda$ p lac 5 carrying the z gene for  $\beta$ -galactosidase was introduced to the suspension of sycamore cells. Smith, et al. were able to isolate some sycamore cells which temporarily grow on lactose. Sycamore cells do not normally grow on lactose. The results indicate, therefore, that the genes for lactose utilization were taken up and expressed by the plant cells.