# Agricultural and Food Chemistry

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The Division of Agricultural and Food Chemistry is composed of people with probably more varied interests than any other division of the American Chemical Society; this logically results in its being a less closely knit division than some others. Even so, the individual activities of most of its members have the common objective of feeding people. And we all know that the number to be fed, both in the United States and in the world, is increasing at a rapid rate.

How long this rate will continue, no one knows. To offset this rapid growth there are many potential technological advances in the food field that promise much further improved quality and increased quantity of Improvements have already crops. changed the growing of crops and preparing and marketing of foods almost unbelievably in the past 50 years. But the time must come when the cost of improvements and increases will be greater than can be afforded and, therefore, there will be a slowing down of progress.

I was born on a farm in Central Illinois-long enough ago to be able to tell first-hand of a half-century of changes. Fifty years ago farming was done with horses and mules, and meat was grown and slaughtered on the farm. About all that we could afford to buy at the grocery store was salt, sugar, flour, and baking soda. We made our own vinegar and syrup, and ground our own corn meal. We canned our own strawberries, cherries, and peaches; and apples and vegetables were stored in earth cellars.

Plant nutrients were used much more sparingly 50 years ago than today, and applications of trace nutrients were of little economic significance. Pesticides were extremely limited. If the grasshoppers were going to get your crop-they got it-and there was not too much to be done about it; and this happened a good deal more often than today's "organic farmers and gardeners" who oppose all use of chemical pesticides and fertilizers would have you believe.

During the past 50 years, advances in animal breeding and nutrition have been most outstanding. Through proper balancing of nutrients and the use of growth promoters and medicaments, efficiency of weight gain per pound of feed has been greatly increased. The commercial feed business of today is extremely complicated because of the great variety of trace materials being added to feeds.

In other fields whole new terminologies have sprung up. Such expressions as plant regulators, growth promoters, and defoliating agents were not in the farmer's vocabulary 50 years ago.

Perhaps by touching even very briefly on some of the many aspects of food production, I can illustrate how important it is that each of us realize that what he does, although it may be confined to an isolated segment, is part of and essential to the solution of the over-all problem of delivering an adequate food supply to an expanding population. Whether in fertilizers, pesticides, or food processing, I am sure that we must dovetail our efforts, must have a common meeting ground, and must have an adequate mechanism of exchanging scientific information, if we are to meet future food requirements.

Fertilizers, insecticides, fungicides, herbicides, other pesticides, plant hormones and regulators, and even tranquilizers all affect the amount and quality of the food processed.

Even though annual consumption of fertilizers has increased from 2 million tons (primary plant nutrient basis) in 1940 to 6.5 million tons at present, only about 10% of farmers, according to a survey by the National Plant Food Institute, use as much fertilizer on their two main crops as recommended by their state experiment stations. With more than ample production facilities for fertilizers, the problem is one of better communications between the fertilizer manufacturer and the farmer. The need for fertilizers must be demonstrated through more soil testing, and reasonable assurance must be given to the farmer that his use of more fertilizer will be profitable. In many instances financial credit must be given to the

farmer until the crop is harvested. On this angle, the fertilizer industry could step up its cooperative efforts with bankers.

Crop production depends not only upon the weather and fertilizer, but also upon control of the various pests that may devour the crop. Even after crops are harvested, insect losses on stored crops are over \$300 million per year. The time will come when we cannot afford and will not tolerate this luxury. There are available today grain fumigants which will protect stored crops. Forced ventilation bins are growing in use because of their quicker and more efficient use of fumigant, as well as better assurance of eliminating toxic vapors before the grain is used. Carbon tetrachloride, carbon bisulfide, ethylene dibromide, methyl bromide, sulfur dioxide, and hydrogen phosphide are among the popular fumigants of today.

Even more costly than the damage they do to stored grains are insects' depredations on growing crops. Since World War II, and the introduction of DDT, man has developed and thrown into the battle a powerful array of synthetic organic insecticides. DDT is still used in greater quantities than any other, but many others have also proved almost indispensable to profitable modern farming. Any insecticide used should be effective in killing insects but should have no adverse effect on plants growing in the soil; the crops when harvested should contain no more than tolerable quantities of the residues.

To collect the information needed to establish these characteristics for any insecticide takes time and costs money. The cost of developing a successful new pesticide may range from \$750,000 to \$3 million, and the costs are rising. Sometime in the future we may, as the result of more experience, be able to predict with more accuracy from structures of molecules those which will be most likely to be useful and yet not have properties which make them unsatisfactory from the standpoint of health hazards.

The need for such information is urgent. Each year insects produce more protoplasm than all other terrestrial forms of animal life put together. In the United States perhaps one million people work just to feed the insects.

Ravages by insects are not new to this country. For example, grasshoppers nearly ate up the Middle West in the '80's and again in the



Those who work on problems associated with food production and those who work in food processing must dovetail their efforts to meet future's food needs

'90's. As one other example, insects twice almost permanently halted construction of the Panama Canal.

Where possible, insect control should be biological and ecological; the pests should be kept in balance. At the same time, chemicals will continue to play a more important role in fighting major outbreaks. Because of the terrific rate of multiplication of insects, scourges occur when a pest goes ou a rampage.

Relatively little is known today regarding diseases of insects, but basic research in this field has already resulted in real progress. There is also hope that further development of techniques for sterilizing insects could be more effective on a cost basis than outright killing.

The use of nontoxic silica aerogel as an insecticide (presumably acting by absorption of the insect's protective wax coating, and causing dehydration) is just one interesting new development in the insecticide field requiring a great deal more research to prove its economic effectiveness. Hormones, too, are the object of some fascinating current pest control research.

In the United States we have some very great food commodity surpluses, particularly in wheat and corn. These surpluses have resulted from greater efficiency in growing crops, government encouragement to produce them, and the use of herbicides and pesticides to protect them. One of the great contributions of pesticides has been in crop areas where high vitamin content foods are produced. We should not forget that without the use of pesticides we definitely could not produce the quantities of these crops which are required for adequate health.

In our battle with insects, we are being forced more and more to prescription entomology. In the past few years we have come from lead arsenate, pyrethrum, and nicotine to well over two hundred compounds used in this field. Farmers sometimes become confused with the many recommendations, and as a result they may use the wrong chemicals or apply them in the wrong way. Thus in pesticides as in fertilizers, a terrific educational program is needed if maximum benefits are to be gained. Since in many cases chemicals are the most practical if not the only means of controlling insects, more consulting entomologists will be required for the education of the producers of food.

Another problem in the pesticide business is inventory control. It is almost impossible to predict where a major pest outbreak will occur, and often the time lag between field survey and government report makes the information only history. The use of computing machines' and rapid relay of information as part of over-all inventory control is quite likely to play



a more important role in the future in decreasing the \$6-billion crop loss caused each year by insects and plant diseases.

Much progress has been made by building into plants genetic characteristics which make them resistant to diseases. Fungal diseases are difficult to control permanently through genetics because of the billions and billions of fungus spores and the probability that out of this large number a few will have characteristics enabling them to infect virtually any new plant strain produced.

Fumigants and fungicides as well as insecticides are applied to the soil to kill or control soil insects and rootinfecting organisms. In the use of soil fumigants, one nearly always kills the good with the bad. And, even though soil is sterilized, contamination from neighboring areas permits many forms of pest life to return very quickly.

Herbicides are growing rapidly in importance; they now are sprayed on some 35–40 million acres each year. In the use of all herbicides we are concerned lest they have a detrimental effect on useful soil microorganisms or on soil microbial decomposition. Most herbicides are removed by leaching, adsorption on soil colloids, and volatilization. In the development of new herbicides, their mechanism of disappearance should be carefully established.

Post-harvest losses prevent 25% of all fruits and vegetables harvested from reaching the intended consumer. Refrigeration is still the most effective and widely used method for protecting fresh produce from rapid spoilage, but chemicals and antibiotics are being widely studied. Evaluation of where they can be used effectively, and collection of data needed for clearance of residues in the products, require a technically trained staff.

The future role of radiation in the food industry is not yet clearly established. This development might be considered as a crash program because of the amount of money spent and the number of laboratories involved in investigations during the past few years. Probable high operating costs, development of off-flavors in the products, prolonged feeding tests needed to determine safety, and rigid control required for plant processes are some of the factors which must be overcome if radiation, even to the extent of partial sterilization-pasteurizationis to be of much significance.

Research workers are finding new chemicals that can dramatically change the basic life processes of plants. Some make plants grow large, others produce dwarfs. Some selectively destroy plants, prevent them from pollinating, retard sprouting, make them male sterile, or cause defoliation. Plants that can be made male sterile and remain female fertile offer great potential in development of new hybrids. This whole field will undoubtedly be exhaustively studied in the future, and much progress will be made.

As an example of plant stimulants, the gibberellins have a vitamin-like effect on many plants. They do not supply energy, initiate abnormal types of cell division, or change generally the chemistry of the plants. The specific effects depending upon time, quantity used, and plant to which applied are numerous. The next few years will see the further development of plant stimulants.

Chemicals are being used now in live animals on or from the farm. The phrase "contented cow" has a new meaning in the shipping of livestock which have been given tranquilizers to prevent shipping losses. If such products are to be widely used, they



Farmers must be shown need for fertilizer through wider use of soil test

must pay their way and show a profit. and meet FDA approval.

Just as farming is changed, the food store of today bears little resemblance to one 50 years ago when there were few middlemen between the farmer and the consumer. Recently the emphasis in food products has been on convenience in the home. And this emphasis has resulted in the complex food processing industry which can now deliver the complete frozen meal.

The use of many chemicals in the food industry all the way from fertilizers, soil fumigants, pesticides, and growth regulators to food additives has necessitated measures of control. Some people are pessimistic, fearing that the cost of control will greatly decrease the incentive for basic research and the development of new and better products. While granting that these checks will be expensive, I do not share this pessimism, because confronted with an expanding population we must make many advances just to hold our own with the demands.

Many of us are concerned with the problem of obtaining qualified technical people for the diversified areas of food production. The glamour of rockets and atomic bombs has made the field of agriculture seem less attractive to college students. This is particularly so if the salaries offered in agricultural sciences are on the low side. Yet, although the farm population has steadily decreased since 1920, the number of technical problems associated with the industry has steadily increased.

At present, in this country, the food program is partly out of balance. With certain crops, millions of dollars are being spent each year just to store the surpluses—and last year we added many millions of bushels to the surplus. Sufficiently large commercial or nonfood end uses for these surpluses are not in sight, and I do not believe that even an accelerated research program on crop surpluses can do more than put a dent in them.

In view of the present rate of population increase, many people believe that these surpluses will disappear in the near future. In most of the world the surpluses are already gone, and food shortages exist and are getting worse very rapidly. As the food problem becomes more acute, the incentives to solve the problem will be greater. There is no question but that controls on the rate of population increase, combined with vigorous research, can result in high standards of nutrition for many years to come.

The paper by E. T. Collinsworth in the December issue of AG AND FOOD, dealing with agricultural chemicals in the world market, is outstanding in pointing up what the U. S. could do in helping countries to better their food supply through insecticides, pesticides, and fertilizers.

The Russians have no intention of relenting in their threat of world domination. With billions of dollars being spent in preparing for rocket and bomb warfare, we can only hope that no one will ever want to start a warfare of mutual annihilation. Hungry people will be swayed toward a supply of food. Our problem is very clear. Education and self-help will develop the principles of democracy —ignorance and starvation will result in the underdeveloped countries' selling their souls to communism by default. If we are to meet the requirements of an expanding population, it can be only by increase of our pioneering and fundamental research, and by dissemination of the information. A big job confronts us, not only in the United States, but also in competition with Russia in the underdeveloped parts of the world. Each bit of research in the food field has an effect on what we are doing, even when the separate bits are performed in apparently unrelated fields.

We need to know about that research as early as we can, which usually means keeping abreast of current literature. Like all scientific literature, that in the food field has increased in recent years. From the origin of the JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY in 1953 to the end of 1958, the Journal has published 797 technical papers, with 32% dealing with food processing, 26% with pesticides, 18% with nutrition, 17% with plant nutrients and regulators, and 7% with fermentation. The use of this published information has been of real value in the food producing and processing industries. We can safely pre-dict an increase in research, and a continuing need for distribution of the results of research.

When the Division of Agricultural and Food Chemistry was founded over 50 years ago, its charter members saw the value of having a common forum for discussion of their already diverse interests. It is gratifying to know that present members of the division have not lost sight of that value. When a questionnaire was distributed last fall to measure member opinion of the division as it is and ought to be, those responding voted overwhelmingly to keep the division in essentially its present form (about 70% voted to continue "as is," and another 15% to continue with some change).

For programs, members favorednot surprisingly-a balanced combination of symposia and research papers, with principal emphasis on new developments. Except for a minority of less than 15%, they did not favor splitting up the division to form smaller but more homogeneous groups. As one member wrote into his questionnaire, diversity can be a good thing.

In conclusion, our division and the JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY serve as a common meeting ground. There is a real need to make them stronger so that we can fulfill the obligations we assumed when we chose our careers in the field of adequately supplying food needs.

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# Supplementation of Foods with Amino Acids

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## Supplementation of food with essential amino acids is beneficial if it brings the first limiting amino acid into balance with the second according to body requirements

THE NUTRITIVE VALUE of the protein in a food depends upon many factors, including the protein's concentration in the food, its composition, particularly in terms of the essential amino acids, its digestibility, and the availability of its amino acids for protein synthesis. Few food items are ideal in all these respects. Foods can be improved in protein quality, however, by supplementation either with additional sources of protein or with essential amino acids. The most desirable procedure is often a suitable combination of these two methods.

The manner in which proteins supplement one another is well known. Less well known, however, is how the nutritive value of a protein may be improved by supplementation with an essential amino acid. This can be done effectively and economically provided a simple rule is observed: a protein should be supplemented with its first limiting amino acid and in such an amount as to obtain balance of the first limiting amino acid with the second. Balance of the two amino acids is achieved when they are available in the ratio in which the body needs them for protein synthesis.

If this rule is followed, and if all other essential nutrients are provided in adequate amounts, a distinct improvement in protein quality will be obtained. Markedly increased efficiency of protein utilization will be experienced. At the same time, proper amino acid supplementation will stretch considerably the world's protein supply, a goal of inestimable value in many countries.

The principles involved in supplementing proteins with essential amino acids were developed through studies with animals, primarily rats, chicks, and dogs. The feed industry, always alert to translate new scientific discoveries into practice, has made use of these laboratory findings and is using the essential amino acid methionine to supplement various poultry diets, thus improving the efficiency with which feed is utilized for the production of meat and eggs. As amino acid supplementation must be not only effective but also economical, the feed industry is weighing the benefits attainable from other amino acids against present and future potential costs of these materials.

The scientific basis for amino acid supplementation of animal feeds has recently been reviewed (10). Discussion of the importance of amino acids in foods for human consumption is a logical sequel to the earlier study.

Protein nutrition is amino acid nutrition. The amount of each essential amino acid present in the protein and liberated in the digestive process becomes available for tissue synthesis. However, not all available amino acids may be utilized. In effect only that portion which conforms to the body's requirements can serve for protein synthesis. All the essential amino acids must be supplied to the synthesizing site at the same time. For maximum efficiency, the essential amino acids must be present also in a definite ratio. The absolute amount of each amino acid needed for growth and maintenance constitutes the body's requirement for that amino acid.

In order to assess the nutritive value of a food protein, it should be necessary to know only the requirements for the various amino acids, and the amino acid composition of the food. A comparison of supply with demand should identify the first limiting amino acid in a given protein as well as indicate the amount of the first necessary to utilize fully the second limiting amino acid. When this amount of the first has been added to the diet, this amino acid is considered "balanced" against the second.

To illustrate, Figure 1 shows the amino acid pattern of corn on a graph depicting the amino acid requirements of the growing rat. In order to identify the first limiting amino acid and the approximate amount needed to bring about balance with the second, the analytically determined amount of each essential amino acid is shown as a bar on a chart on which the essential amino acids are fitted in the order of the amounts required under a so-called requirement line which

#### Table I. Supplementation of Precooked Rice with L-Lysine HCI: Effect on Five Weeks' Rat Growth

(20 Animals per Treatment)

L-Lysine · HCL	% Total	Μ	<b>[</b> ALES	Fe	Females		
Added to	Lysine	Gain	Ratio	Gain	Ratio		
Basal	in Diet	(Gm.)	Feed/Gain	(Gm.)	Feed/Gain		
$\begin{array}{c} 0.00 \\ 0.05 \\ 0.10 \\ 0.20 \end{array}$	0.25	83	5.82	81	5.79		
	0.29	106	4.98	114	5.03		
	0.33	118	4.81	123	4.68		
	0.41	93	4.93	115	4.61		

## Table II. Supplementation of Corn Meal with L-Lysine HCI

Five Weeks' Data-Six Male Rats per Treatment

% L-Lysine · HCl	Total	Experi	MENT <b>R-2</b> 68	Experiment R-304	
Added to	% Lysine	Gain	Ratio	Gain	Ratio
Basal 117	in Diet	(Gm.)	Feed/Gain	(Gm.)	Feed/Gain
0 0.0125	0.16 0.17	36 41	10.21 9.31	40	7.90
0.025	0.18	45	7.95	44	7.56
0.05	0.20	51	7.45	55	
0.10	0.24	34	9.91	35	8.04
0.20	0.32	26	14.69	31	8.13

Table III. Supplementation of White Bread with L-Lysine HCI: Effect on Five Weeks' Rat Growth

% l-Lysine · HCl Added to Basal	% Total Lysine in Diet	М Gain (Gm.)	Ales Ratio Feed/Gain	Mg. Lysine per Gm. of Gain	Fe Gain (Gm.)	MALES RATIO FEED/GAIN	Mg. Lysine per Gm. of Gain
0,00	0.33	71	6.00	20	69	6.01	20
0.10	0.41	117	4.58	19	93	4,92	20
0.20	0.49	159	3.70	18	130	3.99	20
0.30	0.57	188	3.30	19	140	3.61	20
0.40	0.65	228	2.97	19	139	3,60	23
0.50	0.73	203	3.04	22	138	3.58	26
0.60	0.81	174	3.24	26	134	3.62	29
0.70	0.89	223	2.93	27	140	3.67	32
0.80	0.92	186	3.13	30	140	3.62	35
Stock Diet		218	2.92		137	3.76	

cuts diagonally through the ordinate and the abscissa. The origin of the requirement line is placed on the abscissa. The ordinate is provided with a scale to measure the amounts of amino acids as percentage of diet, and the position of each essential amino acid on the ordinate is fixed, according to the established requirement, by the amount necessary to reach the requirement line. Thus, the requirement line cuts across each amino acid line at the requirement level. After the amounts of the various amino acids in corn are indicated on the chart, a line is drawn from the origin of the requirement line to the top of the bar for the amino acid in shortest supply, lysine. In order to determine the approximate amount of the first limiting amino acid needed to bring it into balance with the second, the line is turned clockwise around the origin until it reaches the second amino acid in shortest supply. This proves to be tryptophan. The

approximate amount of lysine, 0.04%, needed for balancing against the second limiting amino acid can then be read directly from the graph. This seemingly small amount when added to a corn diet causes an appreciable improvement in the rat's growth rate.

The same procedure has been used to assess the amino acid deficiency of many proteins and has been found useful as a first approximation. It is obvious, however, that such a procedure cannot give accurate values since both the amino acid analysis of the food's protein and the amino acid requirement are at best only approximations. As time goes on, we expect to obtain more reliable values for the amino acid requirements of the various species including man, and for the essential amino acid content of various food items. We can also increase our knowledge about the factors which affect digestibility of proteins and availability of the essential amino acids. In time, then, greater precision may be expected in our ability to predict the amount of amino acid required for balancing the first with the second limiting amino acid. At present it is necessary to determine experimentally the exact amount needed for balance. This has been done for a number of specific foods for human consumption.

As large segments of the population in different countries suffer in varying degrees from protein malnutrition, it is strange that it is not yet possible to state categorically to what extent amino acid supplementation of the locally available food supply could alleviate the suffering. The plain fact is that we are just now beginning to understand how to test for specific amino acid deficiencies in man, and how to apply laboratory-animal findings to man.

Important sources of protein for man are the cereals: rice, corn, and wheat. By calculation, as carried out more than a dozen years ago by Block and Mitchell (4), the first limiting amino acid in the protein of all three cereals is lysine. Yet, as far as rice and corn are concerned, there are essentially no data available to indicate whether or not supplementation with lysine alone would significantly affect people consuming these cereals. In fact, there are no recorded animal studies in which the amount of lysine alone needed to supplement effectively rice and corn was determined. Therefore, it was not known if it could be shown experimentally that lysine is the first limiting amino acid in rice and corn, and that a significant effect might be obtained from the addition of lysine. Actually, very beneficial effects can be demonstrated in experimental animals provided the established principle for amino acid supplementation is applied to these cereals. It is hoped that these studies may soon be extended to man.

Rice is considered the most important staple cereal of the world since it is the basic ingredient of the daily diet for more than half the human race. Through the experiments of Pecora and Hundley (8), it was known that when both lysine and threonine were added to rice in the amounts found by Rose (9) to be required for the laboratory rat, an impressive improvement in growth was seen. Supplementation with either amino acid alone, at the requirement level, gave no beneficial result.

In Table I is shown the result of one of our recent experiments (11) in which precooked rice was used rather than raw rice, in order to test this food in the approximate condition in which it is consumed. Small, graded levels of lysine hydrochloride





Figure 1. Bar graph of amino acid pattern of corn meal superimposed on a chart showing the amino acid requirements of the rat, according to Rose (9). Lysine is identified as the first limiting amino acid. About 0.04% additional lysine would bring the total lysine in the diet into balance with tryptophan, the second limiting amino acid

were added to a 90% rice diet complete with respect to vitamins, minerals, and essential fatty acids. Supplementation with 0.05% and 0.1%gave increasing growth responses. Supplementation with larger amounts was distinctly less beneficial.

When uncooked long grain and short grain rice and when "converted" rice was used, the largest beneficial amount was established also at approximately 0.1%, corresponding to 2 lb. of L-lysine HCl per ton of rice. At five weeks after weaning, an average improvement in weight gain of about 40% was seen in rat experiments. In a special study with selected litter mates, the growth rate of males increased as much as 100%. As the quality of rice varies considerably according to species, harvest, and other factors, as little as 20% improvement also has been observed in an isolated case. Particularly important, however, is the observation that after 25 weeks on the properly supplemented rice diet the animals were similar in size to normal animals, while the animals on the unsupplemented diet were definitely subnormal in size, weight (Figure 2), and appearance, with rough coats and scaly tails.

These data suggest that the effect of supplementing rice with lysine should also be studied with man. If lysine supplementation of rice should be found nearly as beneficial for man as for laboratory animals, large scale supplementation would appear to be economically feasible, considering the relatively small amount of lysine needed. It would be especially desirable in those areas where supplementation of rice with other protein foods such as fish is not possible.

The relatively low nutritive value of corn protein has been the object of much research, but the literature does not give a clear picture of the possibilities for improving corn protein by amino acid supplementation. Kwashiorkor is a protein deficiency disease known to occur among people whose main source of protein is corn. Consideration of all factors involved suggests that lysine should be the first limiting amino acid (Figure 1). This has, of course, been generally accepted; yet no available study has shown convincingly how proper lysine supplementation of corn might improve its nutritive value.

To test the thesis that lysine supplementation alone should give a beneficial effect, a number of rat growth experiments have been performed. The results of two such experiments, given in Table II, show that lysine Mrs. Catherine Clark, whose Brownberry Ovens is famous for its protein bread (supplemented with L-lysine), is shown at work in her experimental kitchen

alone and in very small amounts—up to 0.05% L-lysine HCl—will indeed improve a 90% corn meal diet. Growth on this diet is obviously poor; yet lysine improved the gain at five weeks up to 50%.

Other studies on corn are under way in several laboratories. Soon we hope to learn if lysine supplementation will prevent the onset of protein deficiency diseases in children. Tests carried out by Dr. Scrimshaw and his associates at the Institute of Nutrition of Central America and Panama suggest that children on corn masa experience improved nitrogen retention if lysine and the second limiting amino acid, tryptophan, are added simultaneously  $(\overline{13})$ . If it can be shown that properly supplemented corn reduces or prevents protein malnutrition, amino acid supplementation of corn may become a public health measure.

Studies on wheat are particularly important and timely. Wheat is not only the most important cereal food in the United States, but is rapidly increasing in importance in countries such as India and Japan. Wheat protein has been known for 40 years to be seriously deficient in lysine.

Table III shows the results of a recently completed experiment with bread, the most widely consumed form







Twenty-five weeks' growth response of rats to bread diets containing increasing amounts of lysine (added as pL-lysine HCl).

Figure 2 (top). Growth responses of rats (on precooked rice diet) to supplementation with optimum level of Llysine-HCI

Center: These rats, all the same age, have been on a bread diet. Two on the right weigh more than the other three, because their bread has been fortified with L-lysine

Figure 3 (bottom). Growth response of rats to bread diets containing increasing amounts of supplementary lysine

of wheat. This study was a repetition of an experiment carried out almost 10 years ago (12), with animals from the same colony and with a commercial white bread containing 5% nonfat milk solids.

When graded levels of lysine were added to the 90% air-dried bread diet containing all the other essential nutrients, increased growth responses were experienced. Maximum response by the males was obtained when 0.4%*L*-lysine · HCl was added to the approximately 0.3% lysine present in the bread, while 0.3% supplementary *L*-lysine · HCl was needed by the females. There is, then, a difference in the lysine requirement of the sexes.

Interestingly enough, this difference is apparent only when the requirement is expressed as a percentage of diet. When the amount of lysine required per gram of gain is calculated (Table III) there is no difference between the females and the males. It is also to be noted that at the optimum levels of supplementation the animals grew as well as those on the stock diet. Under the conditions of these tests, then, lysine was the only limiting amino acid in bread.

These experiments extended over a five-week growth period. Another study was then initiated in which the animals were kept on the various diets for half a year. Figure 3 shows, as a function of time, growth curves of groups of animals of both sexes from some of the treatments.

Particularly important were the results obtained from the addition of 0.2% lysine. This small amount, equivalent to 0.25% L-lysine  $\cdot$  HCl, gave a growth response which although suboptimal during the first few weeks after weaning, permitted the animals eventually to reach a stature not much different from that obtained with optimum supplementation. Many experiments have been carried out in which bread baked from flour supplemented with 0.25% L-lysine · HCl was tested for improvement in nutritive value in comparison with unsupplemented bread. The results have always been consistent and clear-cut. The improvement in protein quality of bread attainable by a modest supplementation with lysine is so striking that nutritionists and clinicians in many laboratories, and in many countries, have studied the possible effects of large-scale supplementation. A recent report (5) may be of special interest in this connection. A reproduction and lactation study has been carried out with rats, comparing the effects of commercial bread containing 6% milk solids with an identical bread supplemented with 0.25% L-lysine · HCl based on the weight of flour and baked into the bread. From many observations over a three-year period, on the parent generation (Figure 4) as well as on six successive generations (Figure 5) involving ultimately several thousand animals, it was obvious that commercial bread as the only source of dietary protein did not support adequate reproduction.

There was a reduced rate of conception as well as poor lactation, resulting in very low body weights at the time the animals are normally weaned. Some symptoms of lysine deficiency were exhibited including nervousness, irritability, and perverted appetite. The rats chewed on everything they could reach and ate their own hair, thus denuding those parts of their bodies which they could reach. The hair accumulated in their stomachs; upon death, hair boluses were found in the stomachs of all animals on the commercial bread diet. No such hair boluses or other symptoms were seen in the animals reared on lysine-supplemented bread. Moreover, lysine supplementation of the bread improved reproduction and lactation considerably and uniformly.

Through this study and many others, some with experimental animals and some with humans, lysine supplementation has been shown to alleviate a number of lesions and pathological conditions. Bread has figured prominently in these studies as a suitable source of better balanced protein. For example, lysine supplementation of bread in the earlier described reproduction studies improved considerably the longevity of the malesa very important observation which needs further study. Lysine supplementation speeds bone growth (3) and prevents accumulation of fat in the bones (7). Other evidence suggests that lysine supplementation reduces incidence of dental caries (6).

These and other reports are being surveyed critically to learn more about specific lysine deficiencies in man. Because of the importance of protein during periods of growth and stress, many specialized uses for lysinesupplemented bread are indicated, including its use in hospital diets and



Figure 4. Growth response of seven litters of rats on commercial bread diets

as food for infants and adolescents (1). In the field of geriatrics, lysine supplementation is highly recommended by the authors of a recent study (2). Many workers with experience of their own seem to feel that even in this country lysine supplementation of bread may help many who suffer to some degree from protein deficiency. Lysine-supplemented bread has been on the market as a specialty loaf for several years, and may be available soon to everyone.

Protein deficiency, the world's top nutritional problem, affects at least two-thirds of its population. Hitherto, deficiencies, which may seriously injure health, could be corrected only by supplementation with animal or vegetable products, either unavailable or too costly in many countries. Now it appears on the basis of experimentation with animals that protein deficiency may be corrected simply by appropriate supplementation of the available foods with the first limiting

Figure 5. Growth response of six successive generations of rats on commerical bread diets



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amino acid. This constitutes a new approach to amino acid supplementation of foods. Until this procedure was developed it was common practice to supplement proteins with arbitrary amounts of the essential amino acids. Usually that amount was chosen which was considered to constitute the requirement for the species. Thus excessive supplementation took place, and often intensified amino acid imbalances. Proper supplementation, on the other hand, leading to a balance of the supplementing amino acid with the second limiting amino acid, is always beneficial.

All the important cereals, including rye, oats, and barley, are deficient in lysine. With full-scale production of lysine, supplementation of cereals would cost only a few dollars per person per year. The first limiting amino acid of peas, beans, peanuts, and many other protein foods is methionine. DL-Methionine and its  $\alpha$ -hydroxy analog, which can replace methionine in the presence of an amino-nitrogen donor, are available as low-cost food supplements.

To learn how to make proper use of these and other amino acids to improve the protein quality of foods and to help in overcoming protein deficiencies is a tremendous challenge.

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