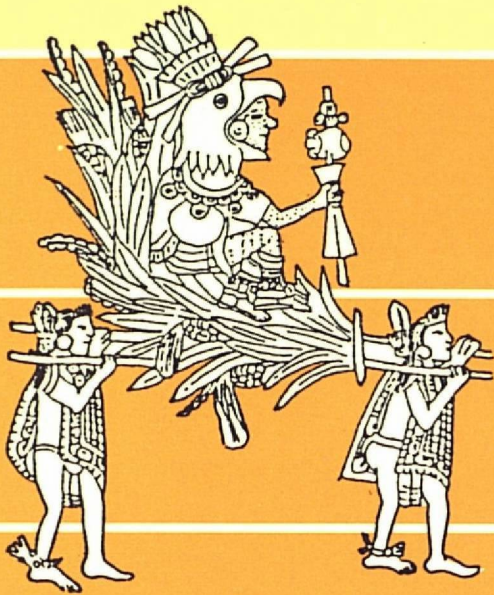

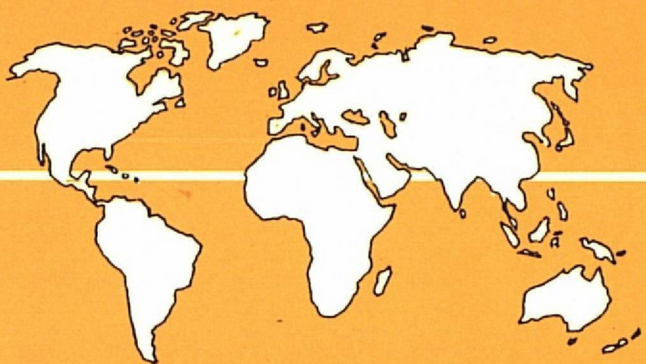

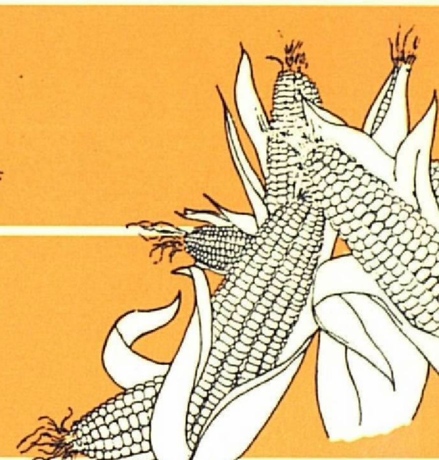

MAIZE

in human nutrition

Food and Agriculture Organization of the United Nations

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FOOD AND AGRICULTURE ORGANIZATION OF THE UNITED NATIONS
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Preface

Over the years FAO has published a series of nutrition studies. *Maize and maize diets*, a nutritional survey published in 1953, covered available information and knowledge on maize up to that date. Since then a vast amount of research information on breeding, varietal improvement, storage, processing, consumption and improvement of the nutritional quality of maize has become available.

The need to update and revise the old edition to include this information was keenly felt by FAO, which has decided to publish it under a new title, *Maize in human nutrition*, and to aim it at a more technical level of readership.

The current edition provides expanded information on the chemical composition of maize, including the make-up of maize protein and dietary fibre, on grain quality and storage and on the effects of lime-cooking of maize and the manufacture of foods such as tortillas, *arepas* and *ogi*. It reviews evidence of the association between maize consumption, bound niacin and pellagra and presents the evidence of amino acid deficiencies in maize and results obtained from experiments with both humans and animals. It discusses the importance of raising the protein quality of maize through incorporation of the opaque-2 gene and its probable contribution to improving the diet of maize-eating populations, and it makes a strong case for commercial production of quality protein maize (QPM). Finally, it provides a more up-to-date account of how maize diets can be improved following the old principle of nutrition: consume a balanced diet containing food legumes, animal protein, fruits and vegetables.

FAO acknowledges the contribution of Prof. R. Bressani of the Institute of Nutrition of Central America and Panama for the extensive revision and rewriting of the book with the technical assistance of Ms Maria Antonietta Rottman. Dr M.A. Hussain, officer-in-charge of Nutrition Programmes Service, Food Policy and Nutrition Division, was responsible for technical editing and preparation of the final manuscript. Valuable suggestions were made by other staff members of the division and of the Plant Production and Protection Division and Agricultural Services Division.

This book is intended for nutritionists, agronomists, food scientists, dieticians and others concerned with maize. It is hoped that they will find it useful and worthwhile.

Paul Lunven
Director
Food Policy and Nutrition Division

Contents

Chapter 1
Introduction

TYPES OF MAIZE

Maize, the American Indian word for corn, means literally "that which sustains life". It is, after wheat and rice, the most important cereal grain in the world, providing nutrients for humans and animals and serving as a basic raw material for the production of starch, oil and protein, alcoholic beverages, food sweeteners and, more recently, fuel. The green plant, made into silage, has been used with much success in the dairy and beef industries. After harvest of the grain, the dried leaves and upper part, including the flowers, are still used today to provide relatively good forage for ruminant animals owned by many small farmers in developing countries. The erect stalks, which in some varieties are strong, have been used as long-lasting fences and walls.

Botanically, maize (*Zea mays*) belongs to the grass family (Gramineae) and is a tall annual plant with an extensive fibrous root system. It is a cross-pollinating species, with the female (ear) and male (tassel) flowers in separate places on the plant. The grain develops in the ears, or cobs, often one on each stalk; each ear has about 300 to 1 000 kernels, weighing between 190 and 300 g per 1 000 kernels, in a variable number of rows (12 to 16). Weight depends on genetic, environmental and cultural practices. Grain makes up about 42 percent of the dry weight of the plant. The kernels are often white or yellow in colour, although black, red and a mixture of colours are also found. There are a number of grain types, distinguished by differences in the chemical compounds deposited or stored in the kernel.

Special crops grown primarily for food include sweet corn and popcorn, although dent, starchy or floury and flint maize are also widely used as food. Flint maize is also used as feed. Immature ordinary corn on the cob either boiled or roasted is widely consumed. Floury maize is a grain with a soft endosperm much used as food in Mexico, Guatemala and the Andean countries. The dent type of maize has a vitreous horny endosperm at the sides and back of the kernel, while the central core is soft. Flint kernels have a thick, hard and vitreous endosperm surrounding a small, granular, starchy centre.

ORIGIN OF MAIZE

The cultivation of maize or Indian corn most probably originated in Central America, particularly in Mexico, from whence it spread northward to Canada and southward to Argentina. The oldest maize, about 7 000 years old, was found by archaeologists in Teotihuacan, a valley near Puebla in Mexico, but it is possible that there were other secondary centres of origin in the Americas. Maize was an essential item in Mayan and Aztec civilizations and had an important role in their religious beliefs, festivities and nutrition. They claimed that flesh and blood were made from maize. The survival of the oldest maize and its distribution depended on humans who harvested the seed for the following planting. At the end of the fifteenth century, after the discovery of the American continent by Christopher Columbus, maize was introduced into Europe through Spain. It then spread through the warmer climates of the Mediterranean and later to northern Europe. Mangelsdorf and Reeves (1939) have shown that maize is grown in every suitable agricultural region of the world and that a crop of maize is being harvested somewhere around the globe every month of the year. Maize grows from latitude 58° in Canada and the former Union of Soviet Socialist Republics to latitude 40° in the Southern Hemisphere. Maize crops are harvested in regions below sea-level in the Caspian Plain and at altitudes of more than 4 000 m in the Peruvian Andes.

In spite of its great diversity of form, all main types of maize known today were apparently already being produced by the native populations when the American continent was discovered. All maize is classified as *Zea mays*. Furthermore, evidence from botany, genetics and cytology has pointed to a common origin for every existing type of maize. Most researchers believe that maize developed from teosinte, *Euchlaena mexicana* Schrod, an annual crop that is possibly its closest relative. Others, however, believe that maize

originated in a wild maize that is now extinct. The closeness of teosinte to maize is suggested by the fact that both have ten chromosomes and are homologous or partially homologous.

Introgression between teosinte and maize has taken place in the past and still does today in areas of Mexico and Guatemala where teosinte grows among the maize crop. Galinat (1977) indicated that of the various hypotheses on the origin of maize, essentially two alternatives remain viable: first, that present-day teosinte is the wild ancestor of maize and/or that a primitive teosinte is the common wild ancestor of both maize and teosinte or, second, that an extinct form of pod maize was the ancestor of maize, with teosinte being a mutant form of this pod maize.

In any case, most of the modern varieties of maize have been derived from materials developed in the southern United States of America, Mexico and Central and South America.

THE MAIZE PLANT

The maize plant may be defined as a metabolic system whose end product is mainly starch deposited in specialized organs, the maize kernels.

The development of the plant may be divided into two physiological stages. In the first or the vegetative stage, different tissues develop and differentiate until the flower structures appear. The vegetative stage is made up of two cycles. In the first cycle the first leaves are formed and development is upward. Dry matter production in this cycle is slow. It ends with the tissue differentiation of the reproductive organs. In the second cycle the leaves and reproductive organs develop. This cycle ends with the emission of the stigmas.

The second stage, also known as the reproductive stage, begins with the fertilization of the female structures, which will develop into ears and grains. The initial phase of this stage is characterized by an increase in the weight of leaves and other flower parts. During the second phase, the weight of the kernels rapidly increases (Tanaka and Yamaguchi, 1972).

The plant develops morphological characteristics and differences in the vegetative and reproductive stages as evolutionary consequences of natural selection and domestication. Some genotypes have adapted to specific ecological zones and so have developed such barriers as day-length sensitivity and temperature sensitivity, which limit their adaptability to specific areas of latitude and altitude. Thus improvement programmes must be conducted within the areas where the improved varieties are to be grown. This does not mean, however, that specific genetic characteristics can be attained by backcrossing.

The morphology or architecture of the plant has also suffered evolutionary pressures that have resulted in great variability in the number, length and width of leaves, plant height, positions of ears, number of ears per plant, maturation cycles, grain types and number of rows of grain, among many other characteristics.

This variability is of great value in improving the productivity of the plant and specific organic components of the grain. The main yield components include the number and weight of grains. These yield components are determined by quantitative genetic effects that can be selected relatively easily. The number of grains depends on the ear and is determined by the number of rows and the number of kernels per row. The size and shape of the kernel determine its weight in the presence of other constant factors such as grain texture and grain density. The ratio of grain weight to total plant weight for most maize lines is about 0.52. From 100 kg of cobs, about 18 kg of grain is obtained. One ha of maize yields about 1.55 tonnes of stalk residue. In field-dried maize plants from three locations in Guatemala, plant dry weight varied from 220 to 314 g. This weight comprised 1.8 percent dried flowers, 14.7 to 27.8 percent stalks, 7.4 to 15.9 percent leaves, 11.7 to 13 percent husks, and 9.7 to 11.5 percent cobs. The field-dried grain represented 30 to 55.9 percent of the whole plant dry weight. These data show the significant yield of plant residues that are often left in the field. The distribution may change, however, since it is accepted that about half of the dry matter is grain and the other half is made up of plant residues excluding roots (Barber, 1979).

STRUCTURE OF THE MAIZE KERNEL

Maize kernels develop through accumulation of the products of photosynthesis, root absorption and metabolism of the maize plant on the

TABLE 1

Weight distribution of main parts of the kernel

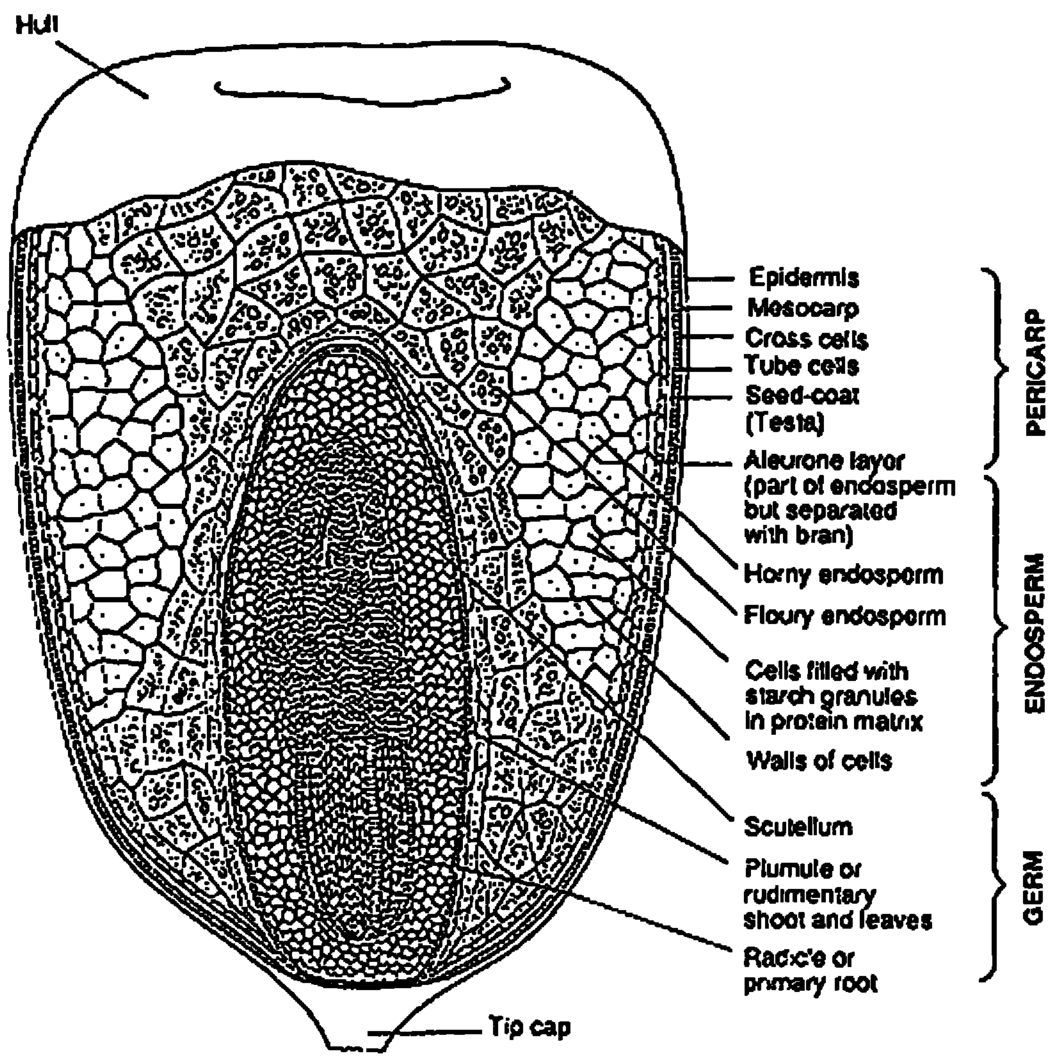
Structure	Percent weight distribution
Pericarp	5-6
Aleurone	2-3
Endosperm	80-85
Germ	10-12

female inflorescence called the ear. This structure may hold from 300 to 1 000 single kernels depending on the number of rows, diameter and length of the cob. Kernel weight may be quite variable, ranging from about 19 to 40 g per 100 kernels. During harvest the ears of maize are removed from the maize plant either by hand or mechanically. The husks covering the ear are first stripped off, then the kernels are separated by hand or, more often, by machine.

The maize kernel is known botanically as a caryopsis; a single grain contains the seed coat and the seed, as shown in Figure 1. The figure also shows the four major physical structures of the kernel: the pericarp, hull or bran; the germ or embryo; the endosperm; and the tip cap (dead tissue found where the kernel joins the cob). The gross anatomy and the microscopic structure of these anatomical components were well described by Wolf *et al.* (1952) and by Wolf, Khoo and Seckinger (1969). They also studied the structure of the improved opaque-2 maize and found differences between its endosperm and that of common maize. The protein matrix was thinner and there were fewer and smaller protein bodies, since there is a restriction in zein synthesis in opaque-2 maize. Robutti, Hoseny and Deyoe (1974) and Robutti, Hoseny and Wasson (1974) reported on the protein distribution, amino acid content and endosperm structure of opaque-2 maize.

The weight distribution of the different parts of the maize kernel is shown in Table 1. The endosperm, the largest structure, provides about 83 percent of the kernel weight, while the germ averages 11 percent and the pericarp 5

FIGURE 1
Maize kernel structure: longitudinal section enlarged approximately 30 times



(Diagram courtesy of the Wheat Flour Institute, Chicago, Illinois, 1964)

percent. The remainder is the tip cap, a conical structure that together with the pedicel attaches the kernel to the ear of maize. Table 2 shows the distribution of weight and nitrogen among the anatomical parts of common and selected kernel varieties, such as high-oil and high-protein maize and three quality protein maize (QPM) selections (Bressani and Mertz, 1958). The main difference in the high-oil variety is the size of the germ, which is about three times as large as the germ from common maize with a reduction

TABLE 2

Distribution of weight and nitrogen among parts of the kernel

Maize sample	Weight of 20 seeds (g)	Weight distribution (%)			Total N (%)	Nitrogen distribution (%)		
		Seed-coat[a]	Endosperm	Germ		Seed-coat	Endosperm	Germ
US 4251	5.62	6.3	86.3	7.4	1.31	3.3	81.2	15.5
US high oil (HO)	5.72	6.4	71.2	22.4	1.99	2.4	68.4	29.2
US high protein (H5)	4.32	6.9	82.7	10.4	2.24	2.2	83.2	14.6
US high protein (HP)	4.97	7.4	78.9	13.7	2.14	2.7	78.2	19.1
US normal-Sh1 PT	4.38	6.7	79.6	13.7	2.14	2.7	78.2	19.1
US normal mutant-Sh1 PT	2.50	10.7	70.6	18.7	2.21	6.1	64.6	29.3
Tiquisate (TGY)(Guat.)	8.24	4.9	83.9	11.2	1.37	2.8	75.2	22.0
San Sebastian (SSD)(Guat.)	8.24	4.9	83.9	11.2	1.37	2.8	75.2	22.0
Guatemalan 142-48	6.91	6.9	82.1	11.0	1.83	2.6	81.0	16.4
Guatemalan Cuyuta	5.95	5.7	82.5	11.8	1.28	2.9	72.4	24.7
Guatemalan teosinte	1.56	55.6[b]	44.4	—	1.81[c]	8.2	91.8[d]	—
Nutricta QPM	5.91	5.7	82.7	11.6	1.42	1.7	72.8	25.5
QPM yellow	6.49	5.9	81.6	12.5	1.48	2.4	73.4	24.2
QPM white	5.31	5.9	82.4	1.6	1.36	1.4	72.8	25.7

[a]Pericarp plus tip cap
[b]Includes the seed-coat (1.3%) and the hull (54.3%)
[c]The hull contained 0.26% nitrogen; dehulled teosinte contained 3.81% nitrogen
[d]Includes the germ
Source: Bressani and Mertz, 1958

TABLE 3

Weight and nitrogen distribution of parts of common and opaque-2 maize kernels

Part of kernel	Dry matter (%)			Nitrogen (%)		
	Common	Common	Opaque-2	Common	Common	Opaque-2
Germ	13.5	8.1	35	20.1	14.9	35.1
Endosperm	80.0	84.0	61	76.5	80.5	60.7
Seed coat	6.5	7.9	4	3.4	4.6	4.2

Source: Landry and Moureaux, 1980

in endosperm weight. Germ of the high-protein varieties is larger than that of common maize but about half the size of high-oil varieties. There are also differences in the weight of the seed-coats. Table 2 also shows some data for teosinte, the closest relative to maize. The seed weight is much lower than that of maize seed, and the endosperm weighs about half that of maize. The three QPM selections are similar to maize in weight per seed and in weight of the seed-coat, the endosperm and the germ. Similar data have been reported by other authors. Table 3 summarizes data for two common varieties and one opaque-2 maize (Landry and Moureaux, 1980). The two common samples have the same general characteristics as those reported above; the opaque-2 sample, however, has a larger germ providing more nitrogen than the QPM selections in Table 2. With respect to the germ, the increase of weight and of nitrogen amounts in absolute as well as relative terms is consistent with other results (Watson, 1987).

WORLD PRODUCTION

World maize production increased from 1979-1981 to 1987, as shown by continent in Table 4. The land area planted with maize increased from 105 million ha in 1961 to about 127 million ha in 1987. Although part of the increase resulted from additional land area planted, significant increases in production resulted from genetic improvement and more efficient

TABLE 4

World maize production

Region and year	Area harvested (*1 000 ha*)	Yield (*kg/ha*)	Production (*1 000 MT*)
Africa			
1979-81	18 193	1 554	28 268
1985	19 099	1 522	29 069
1986	19 580	1 575	30 840
1987	19 512	1 395	27 225
North and Central America			
1979-81	39 399	5 393	212 384
1985	40 915	6 092	249 258
1986	37 688	6 116	230 511
1987	35 187	5 690	200 211
South America			
1979-81	16 751	1 928	32 369
1985	17 813	2 182	38 859
1986	18 799	2 021	38 001
1987	19 413	2 143	41 595
Asia			
1979-81	36 815	2 296	84 531
1985	35 246	2 628	92 629
1986	37 474	2 729	102 274
1987	37 399	2 788	104 269
Europe			
1979-81	11 738	4 668	54 792
1985	11 556	5 423	62 673
1986	11 539	6 207	71 621
1987	11 405	6 039	68 901

(continued)

TABLE 4 (continued)

Region and year	Area harvested (1 000 ha)	Yield (kg/ha)	Production (1 000 MT)
Oceania			
1979-81	76	4 359	332
1985	124	3 804	471
1986	107	4 402	471
1987	84	4 302	363
USSR			
1979-81	3 063	2 989	9 076
1985	4 482	3 214	14 406
1986	4 223	2 955	12 479
1987	4 600	3 217	14 800
World			
1979-81	126 035	3 345	421 751
1985	129 235	3 771	487 367
1986	129 411	3 757	486 198
1987	127 605	3 584	457 365

Source: FAO, 1988

technological field practices and fertilizer applications, as well as from the introduction of new, more highly reproductive varieties.

The developing countries have more area given to maize cultivation than developed countries, but yield in the latter is about four times higher. Since 1961, yields per ha in the United States, for example, have increased significantly, while yields in Mexico, Guatemala and Nigeria (selected as countries where maize intake by the human population is high, particularly in the first two) have increased only slightly. While most of the production in developing countries is for human consumption, in the developed world it is mainly for industrial use and animal feed. The high yields and

production in North and Central America are mainly attributed to the United States, which outproduces countries such as Mexico where maize is the most important staple cereal grain. With changing rural-to-urban populations and lifestyles in developing countries, there is a continuous shift to the consumption of wheat, which may influence maize production. There is a slow increase in its use in industry and as an animal feed, particularly for poultry and other monogastric animals. A comparison of the available data for wheat, maize and rice put maize as the second most important cereal grain, after wheat and before rice. In terms of yield per hectare, however, maize outyields the other two. The only food crop outyielding maize in tonnes per hectare is potatoes in their unprocessed state, though not on an equal moisture basis.

USES

As indicated in previous sections, maize has three possible uses: as food, as feed for livestock and as raw material for industry. As a food, the whole grain, either mature or immature, may be used; or the maize may be processed by dry milling techniques to give a relatively large number of intermediary products, such as maize grits of different particle size, maize meal, maize flour and flaking grits. These materials in turn have a great number of applications in a large variety of foods. Maize grown in subsistence agriculture continues to be used as a basic food crop. In developed countries more than 60 percent of the production is used in compounded feeds for poultry, pigs and ruminant animals. In recent years, even in developing countries in which maize is a staple food, more of it has been used as an animal feed ingredient. "High moisture" maize has been paid much attention recently as an animal feed because of its lower cost and its capacity to improve efficiency in feed conversion. The by-products of dry milling include the germ and the seed-coat. The former is used as a source of edible oil of high quality. The seed-coat or pericarp is used mainly as a feed, although in recent years interest has developed in it as a source of dietary fibre (Earll *et al*., 1988; Burge and Duensing, 1989). Wet milling is a process applicable mainly in the industrial use of maize, although the alkaline cooking process used in manufacturing tortillas (the thin, flat bread of Mexico and other Central American countries) is also a wet milling operation that removes only the pericarp (Bressani, 1990). Wet milling yields maize starch and by-products such as maize gluten, used as a feed ingredient. The maize germ processed to produce oil gives as a by-product maize germ meal, used as an animal feedstuff. Some attempts have been made to use these by-products for humans in food mixes and formulations.

Although the technology has been available for a long time, the increase in fuel oil prices has resulted in much research on the fermentation of maize to produce alcohol, popular in some states of North America. Fermentation also provides some alcoholic beverages.

Finally, maize plant residues also have important uses, including animal feeds as well as a number of chemicals produced from the cobs, such as furfural and xylose. These residues are also important as soil conditioners.

Chapter 2

Chemical composition and nutritional value of maize

There are significant amounts of data on the chemical composition of maize. Many studies have been conducted to understand and evaluate the effects of the genetic make-up of the relatively large number of available maize varieties on chemical composition, as well as the effects of environmental factors and agronomic practices on the chemical constituents and nutritive value of the kernel and its anatomical parts. Chemical composition after processing for consumption is an important aspect of nutritive value (see Chapter 5); it is affected by the physical structure of the kernel, by genetic and environmental factors, by processing and by other links in the food chain. In this chapter, the chemical nature of maize, of both common and quality protein types, is described as a basis for understanding the nutritive value of various maize products consumed throughout the world.

CHEMICAL COMPOSITION OF PARTS OF THE KERNEL

There are important differences in the chemical composition of the main parts of the maize kernel as shown in Table 5. The seed-coat or pericarp is characterized by a high crude fibre content of about 87 percent, which is constituted mainly of hemicellulose (67 percent), cellulose (23 percent) and lignin (0.1 percent) (Burge and Duensing, 1989). On the other hand, the endosperm contains a high level of starch (87.6 percent) and protein levels of about 8 percent. Crude fat content in the endosperm is relatively low. Finally, the germ is characterized by a high crude fat content, averaging about 33 percent. The germ also contains a relatively high level of protein (18.4 percent) and minerals. Some information is available on the chemical composition of the aleurone layer (see Figure 1), which is relatively high in

TABLE 5

Proximate chemical composition of main parts of maize kernels (%)

Chemical component	Pericarp	Endosperm	Germ
Protein	3.7	8.0	18.4
Ether extract	1.0	0.8	33.2
Crude fibre	86.7	2.7	8.8
Ash	0.8	0.3	10.5
Starch	7.3	87.6	8.3
Sugar	0.34	0.62	10.8

Source: Watson, 1987

protein content (about 19 percent) as well as in crude fibre. Tables 2 and 3 provide some additional details on nitrogen distribution in the maize kernel. The endosperm contributes the largest amount, followed by the germ, with only small amounts from the seed-coat. About 92 percent of the protein in teosinte comes from the endosperm. Protein in the maize kernel has been reported on by a number of researchers (e.g. Bressani and Mertz, 1958).

From the data shown in Tables 2 and 3 it is evident that the carbohydrate and protein contents of maize kernels depend to a very large extent on the endosperm, and crude fat and to a lesser extent protein and minerals on the germ. Crude fibre in the kernel comes mainly from the seed-coat. The weight distribution among parts of the maize kernel and their particular chemical composition and nutritive value are of great importance when maize is processed for consumption. In this regard there are two important matters from the nutritive point of view. Germ oil provides relatively high levels of fatty acids (Bressani *et al.*, 1990; Weber, 1987). Where there are high intakes of maize, as in certain populations, those who consume the degermed grain will obtain less fatty acids than those who eat processed whole maize. This difference is probably equally important with respect to protein, since the amino acid content of germ proteins is quite different from that of endosperm protein. This is indicated in Table 6, in which essential amino acids are expressed as mg percent by weight and as mg per g N. As Table 2 shows, the endosperm represents between 70 and 86 percent of the kernel weight and the germ between 7 and 22 percent. It follows that, in considering the whole kernel, the essential amino acid content is a reflection of the amino acid content in the protein of the endosperm, in spite of the fact that the amino acid pattern in the germ protein is higher and better balanced. Germ proteins nevertheless contribute a relatively high amount of certain amino acids, although not enough to provide a higher quality of protein in the whole kernel. The germ provides some lysine and tryptophan, the two limiting essential amino acids in maize protein. Endosperm proteins are low in lysine and tryptophan, as is the whole grain protein (see Table 6, in which the FAO/WHO essential amino acid pattern is also shown). The deficiencies in lysine, tryptophan and isoleucine have been well demonstrated by numerous animal studies (Howe, Janson and Gilfillan, 1965) as well as by a few studies on humans (Bressani, 1971).

TABLE 6

Essential amino acid content of germ protein and endosperm protein

Amino acid	Endosperm[a]		Germ[b]		FAO/WHO pattern
	mg %	mg/g N	mg %	mg/g N	
Tryptophan	48	38	144	62	60
Threonine	315	249	622	268	250
Isoleucine	365	289	578	249	250
Leucine	1 024	810	1 030	444	440
Lysine	228	180	791	341	340
Total sulphur amino acids	249	197	362	156	220
Phenylalanine	359	284	483	208	380
Tyrosine	483	382	343	148	380
Valine	403	319	789	340	310

[a] 1.16 percent N
[b] 2.32 percent N
Source: Orr and Watt, 1957

TABLE 7

Net protein of whole grain, germ and endosperm of Guatemalan maize varieties[a]

Sample	Yellow	Azotea	Cuarenteño	Opaque-2
Whole grain	42.5	44.3	65.4	81.4
Germ	65.7	80.4	90.6	85.0
Endosperm	40.9	42.0	46.4	77.0

[a]Expressed as percentage of casein (100%)
Source: Poey *et al.*, 1979

The superior quality of germ protein to endosperm protein in various samples of maize is shown in Table 7, which compares the quality of the two parts as percentages of the reference protein, casein in this case. The maize varieties include three of common maize and one of quality protein maize (QPM). In all cases the quality of germ proteins is much higher than that of endosperm proteins and is obviously superior to the quality of whole kernel protein. Endosperm protein quality is lower than that of the whole kernel because of the higher contribution of germ protein. The data also show less difference in the quality of germ and endosperm proteins in the QPM variety. Furthermore, the QPM endosperm and whole grain quality are significantly superior to the endosperm and whole grain quality of the other samples. These data, again, are important in regard to how maize is processed for consumption and in its impact on the nutritional status of people. They also clearly show that the quality of QPM is better than that of common maize. The higher quality of QPM endosperm is also of significance for populations that consume maize without the germ.

GROSS CHEMICAL COMPOSITION

Information on the gross chemical composition of maize is abundant. The variability of each major nutrient component is great. Table 8 summarizes data on various types of maize taken from several publications. The

TABLE 8

Gross chemical composition of different types of maize (%)

Maize type	Moisture	Ash	Protein	Crude fibre	Ether extract	Carbohydrate
Salpor	12.2	1.2	5.8	0.8	4.1	75.9
Crystalline	10.5	1.7	10.3	2.2	5.0	70.3
Floury	9.6	1.7	10.7	2.2	5.4	70.4
Starchy	11.2	2.9	9.1	1.8	2.2	72.8
Sweet	9.5	1.5	12.9	2.9	3.9	69.3
Pop	10.4	1.7	13.7	2.5	5.7	66.0
Black	12.3	1.2	5.2	1.0	4.4	75.9

Source: Cortez and Wild-Altamirano, 1972

variability observed is both genetic and environmental. It may influence the weight distribution and individual chemical composition of the endosperm, germ and hull of the kernels.

Starch

The major chemical component of the maize kernel is starch, which provides up to 72 to 73 percent of the kernel weight. Other carbohydrates are simple sugars present as glucose, sucrose and fructose in amounts that vary from 1 to 3 percent of the kernel. The starch in maize is made up of two glucose polymers: amylose, an essentially linear molecule, and amylopectin, a branched form. The composition of maize starch is genetically controlled. In common maize, with either the dent or flint type of endosperm, amylose makes up 25 to 30 percent of the starch and amylopectin makes up 70 to 75 percent. Waxy maize contains a starch that is 100 percent amylopectin. An endosperm mutant called amylose-extender (*ae*) induces an increase in the amylose proportion of the starch to 50 percent and higher. Other genes, alone or in combination, may also modify the amylose-to-amylopectin ratio in maize starch (Boyer and Shannon, 1987).

Protein

After starch, the next largest chemical component of the kernel is protein. Protein content varies in common varieties from about 8 to 11 percent of the kernel weight. Most of it is found in the endosperm. The protein in maize kernels has been studied extensively. It is made up of at least five different fractions, according to Landry and Moureaux (1970, 1982). In their scheme, albumins, globulins and non-protein nitrogen amount to about 18 percent of total nitrogen, in a distribution of 7 percent, 5 percent and 6 percent, respectively. The prolamine fraction soluble in 55 percent isopropanol and isopropanol with mercaptoethanol (ME) contributes 52 percent of the nitrogen in the kernel. Prolamine 1 or zein 1 soluble in 55 percent isopropanol is found in the largest concentration, about 42 percent, with 10 percent provided by prolamine 2 or zein 2. An alkaline solution, pH 10 with 0.6 percent ME, extracts the glutelin fraction 2, in amounts of about 8 percent, while glutelin 3 is extracted with the same buffer as above with 0.5 percent sodium dodecyl sulphate in amounts of 17 percent for a total globulin content of 25 percent of the protein in the kernel. Usually a small amount, about 5 percent, is residual nitrogen.

Table 9 summarizes data by Ortega, Villegas and Vasal (1986) on the protein fractionation of a common maize (Tuxpeño-1) and a QPM (Blanco Dentado-1). Fractions II and III are zein I and zein II, of which zein I (Fraction II) is significantly higher in the Tuxpeño-1 variety than in the QPM. Similar results have been published by other researchers. Amounts of the alcohol-soluble proteins are low in immature maize. They increase as the grain matures. When these fractions were analysed for their amino acid content, the zein fraction was shown to be very low in lysine content and lacking in tryptophan. Since these zein fractions make up more than 50 percent of the kernel protein, it follows that the protein is also low in these two amino acids. The albumin, globulin and glutelin fractions, on the other hand, contain relatively high levels of lysine and tryptophan. Another important feature of the zein fractions is their very high content of leucine, an amino acid implicated in isoleucine deficiency (Patterson *et al.*, 1980).

Quality protein maize differs from common maize in the weight distribution of the five protein fractions mentioned above, as shown in Table 9. The extent of the change is variable and affected by genotype and cultural conditions. It has been found, however, that the opaque-2 gene reduces the concentration of zein by some 30 percent. As a result, lysine and tryptophan content is higher in QPM varieties than in common maize.

TABLE 9

Protein fraction distribution of Tuxpeño-1 and Blanco Dentado-1 QPM (whole grain)

Fraction	Blanco Dentado-1 QPM		Tuxpeño-1	
	Protein (*mg*)	Percent protein	Protein (*mg*)	Percent total protein
I	6.65	31.5	3.21	16.0
II	1.25	5.9	6.18	30.8
III	1.98	9.4	2.74	13.7
IV	3.72	17.6	2.39	12.0
V	5.74	27.2	4.08	20.4
Residue	1.76	8.3	1.44	7.1

Source: Ortega, Villegas and Vasal, 1986

The nutritional quality of maize as a food is determined by the amino acid make-up of its protein. Representative amino acid values are shown in Table 10 for both common maize and QPM. To establish the adequacy of the essential amino acid content the table also includes the FAO/WHO essential amino acid pattern. In common maize, deficiencies in lysine and tryptophan are evident as compared with QPM. An additional important feature is the high leucine content in common maize and the lower value of this amino acid in QPM.

Oil and fatty acids

The oil content of the maize kernel comes mainly from the germ. Oil content is genetically controlled, with values ranging from 3 to 18 percent. The average fatty acid composition of the oil in selected varieties from Guatemala is shown in Table 11. These values differ to some extent; it may

TABLE 10

Amino acid content of maize and teosinte (%)

Amino acid	Guatemalan maize				US maize				Teosinte	Hard QPM	Soft QPM	FAO/ WHO pattern
	Cuyuta (white)	SSD (white)	TGY (yellow)	142-48 (yellow)	4251 (hybrid)	HO (high-oil white)	H5 (high-protein yellow)	HP (high-protein white)				
(Nitrogen)	1.28	1.37	1.57	1.83	1.31	1.99	2.24	2.91	3.81[b]	1.74[c]	1.71[d]	–
Alanine	7.5[a]	12.5	10.3	8.6	8.9	6.4	10.8	9.9	8.5	–	–	–
Arginine	3.5	3.6	4.1	2.9	3.9	4.6	3.6	3.9	2.9	6.3	6.7	–
Aspartic acid	6.5	5.8	6.1	6.0	6.2	6.0	6.8	6.1	5.3	8.7	8.9	–
Cystine	1.4	1.4	1.4	1.6	1.6	1.5	1.2	1.4	1.1	2.2	1.9	–
Glutamic acid	15.4	14.0	13.5	15.0	14.6	13.9	12.4	12.8	19.9	19.8	19.2	–
Glycine	3.1	2.8	2.9	2.6	3.3	3.4	2.6	2.8	2.2	4.6	4.6	–
Histidine	2.1	2.0	2.2	2.1	2.8	2.3	2.0	2.2	1.9	3.7	3.6	–
Isoleucine	2.6	2.7	3.4	3.0	3.3	3.5	3.7	4.0	4.7	3.5	3.5	4.0
Leucine	10.5	12.0	12.2	13.4	12.2	7.8	13.6	15.2	16.8	9.1	8.7	7.0
Lysine	2.8	2.1	2.6	2.3	2.9	3.2	2.1	2.0	1.3	4.5	4.4	5.4
Methionine	1.3	1.2	1.0	1.0	1.6	1.2	1.7	1.1	1.2	1.7	1.8	3.5[c]
Phenylalanine	4.4	4.6	4.4	5.4	4.5	2.9	5.3	5.7	5.7	5.2	4.1	6.0[d]
Proline	8.1	7.4	6.6	9.6	10.3	9.3	8.3	6.8	9.6	8.4	8.1	–
Serine	4.5	4.2	4.6	4.5	4.6	4.8	5.0	5.5	5.2	4.3	4.5	–
Threonine	3.1	2.9	3.1	3.0	3.3	3.2	3.1	3.3	3.0	3.6	3.7	4.0
Tryptophan	0.63	0.47	0.51	0.44	0.49	0.56	0.43	0.44	0.38	0.9	1.0	1.4
Tyrosine	2.9	3.0	3.0	3.3	3.4	3.5	3.6	4.1	4.4	3.7	3.7	–
Valine	4.1	4.1	4.3	4.0	4.6	2.1	4.3	4.6	4.8	5.4	5.3	5.0

[a]Percent of crude protein (N x 6.25), g/16 g N
[b]Dehulled
[c]Total sulphur amino acids
[d]Total aromatic amino acids
Sources: Bressani and Mertz, 1958; Mertz *et al.*, 1975

TABLE 11

Fatty acid content of Guatemalan maize varieties and Nutricta QPM (%)

Maize variety	C16:0 Palmitic	C18:0 Stearic	C18:1 Oleic	C18:2 Linoleic	C18:3 Linolenic
QPM Nutricta	15.71	3.12	36.45	43.83	0.42
Azotea	12.89	2.62	35.63	48.85	—
Xetzoc	11.75	3.54	40.07	44.65	—
Tropical White	15.49	2.40	34.64	47.47	—
Santa Apolonia	11.45	3.12	38.02	47.44	—

Source: Bressani *et al.*, 1990

be expected that oils from different varieties have different compositions. Maize oil has a low level of saturated fatty acids, i.e. on average 11 percent palmitic and 2 percent stearic acid. On the other hand, it contains relatively high levels of polyunsaturated fatty acids, mainly linoleic acid with an average value of about 24 percent. Only very small amounts of linoleic and arachidonic acids have been reported. Furthermore, maize oil is relatively stable since it contains only small amounts of linoleic acid (0.7 percent) and high levels of natural antioxidants. Maize oil is highly regarded because of its fatty acid distribution, mainly oleic and linoleic acids. In this respect, populations that consume degermed maize benefit less in terms of oil and fatty acids than populations that consume whole-kernel products.

Dietary fibre

After carbohydrates, proteins and fats, dietary fibre is the chemical component found in the greatest amounts. The complex carbohydrate content of the maize kernel comes from the pericarp and the tip cap, although it is also provided by the endosperm cell walls and to a smaller extent the germ cell walls. The total soluble and insoluble dietary fibre content of maize kernels is shown in Table 12. Differences in soluble and insoluble dietary fibre are small between samples, even though QPM Nutricta has higher levels of total dietary fibre than common maize, mainly because of

TABLE 12
Soluble and insoluble dietary fibre in common and quality protein maize (%)

Maize type	Dietary fibre		
	Insoluble	Soluble	Total
Highland	10.94 ± 1.26	1.25 ± 0.41	12.19 ± 1.30
Lowland	11.15 ± 1.08	1.64 ± 0.73	12.80 ± 1.47
QPM Nutricta	13.77	1.14	14.91

Source: Bressani, Breuner and Ortiz, 1989

TABLE 13
Neutral and acid detergent fibre, hemicellulose and lignin in five maize varieties (%)

Maize No.	Neutral detergent fibre	Acid detergent fibre	Hemicellulose	Lignin	Cellular walls
1	8.21	3.23	4.98	0.14	9.1
2	10.84	2.79	8.05	0.12	10.8
3	9.33	3.08	6.25	0.13	12.0
4	11.40	2.17	9.23	0.12	13.1
5	14.17	2.68	11.44	0.14	14.2
Average	10.79 ± 2.27	2.79 ± 0.44	8.00 ± 2.54	0.13 ± 0.01	11.8 ± 2.0

Source: Bressani, Breuner and Ortiz, 1989

a higher level of insoluble fibre. Table 13 shows values of fibre expressed as acid and neutral detergent fibre, hemicellulose and lignin in whole maize. The values shown in the table are similar to those reported by Sandstead *et al.* (1978) and Van Soest, Fadel and Sniffen (1979). Sandstead *et al.* found that maize bran was composed of 75 percent hemicellulose, 25 percent

cellulose and 0.1 percent lignin on a dry-weight basis. Dietary fibre content in dehulled kernels would obviously be lower than that of whole kernels.

Other carbohydrates

When mature, the maize kernel contains carbohydrates other than starch in small amounts. Total sugars in the kernel range between 1 and 3 percent, with sucrose, the major component, found mostly in the germ. Higher levels of monosaccharides, disaccharides and trisaccharides are present in maturing kernels. At 12 days after pollination the sugar content is relatively high, while starch is low. As the kernel matures, the sugars decline and starch increases. For example, sugars were found to have reached a level of 9.4 percent of kernel dry weight in 16-day-old kernels, but the level decreased significantly with age. Sucrose concentration at 15 to 18 days after pollination was between 4 and 8 percent of kernel dry weight. These relatively high levels of reducing sugar and sucrose are possibly the reason why immature common maize and, even more, sweet maize are so well liked by people.

Minerals

The concentration of ash in the maize kernel is about 1.3 percent, only slightly lower than the crude fibre content. The average mineral content of some samples from Guatemala is shown in Table 14. Environmental factors probably influence the mineral content. The germ is relatively rich in minerals, with an average value of 11 percent as compared with less than 1 percent in the endosperm. The germ provides about 78 percent of the whole kernel minerals. The most abundant mineral is phosphorus, found as phytate of potassium and magnesium. All of the phosphorus is found in the embryo, with values in common maize of about 0.90 percent and about 0.92 percent in opaque-2 maize. As with most cereal grains, maize is low in calcium content and also low in trace minerals.

Fat-soluble vitamins

The maize kernel contains two fat-soluble vitamins: provitamin A, or carotenoids, and vitamin E. Carotenoids are found mainly in yellow maize,

TABLE 14
Mineral content of maize (Average of five samples)

Mineral	Concentration (*mg/100 g*)
P	299.6 ± 57.8
K	324.8 ± 33.9
Ca	48.3 ± 12.3
Mg	107.9 ± 9.4
Na	59.2 ± 4.1
Fe	4.8 ± 1.9
Cu	1.3 ± 0.2
Mn	1.0 ± 0.2
Zn	4.6 ± 1.2

Source: Bressani, Breuner and Ortiz, 1989

in amounts that may be genetically controlled, while white maize has little or no carotenoid content. Most of the carotenoids are found in the hard endosperm of the kernel and only small amounts in the germ. The beta-carotene content is an important source of vitamin A, but unfortunately yellow maize is not consumed by humans as much as white maize. Squibb, Bressani and Scrimshaw (1957) found beta-carotene to be about 22 percent of total carotenoids (6.4 to 11.3 µg per gram) in three yellow maize samples. Cryptoxanthin accounted for 51 percent of total carotenoids. Vitamin A activity varied from 1.5 to 2.6 µg per gram. The carotenoids in yellow maize are susceptible to destruction after storage. Watson (1962) reported values of 4.8 mg per kg in maize at harvest, which decreased to 1.0 mg per kg after 36 months of storage. The same loss took place with xanthophylls. Recent studies have shown that the conversion of beta-carotene to vitamin A is increased by improving the protein quality of maize.

The other fat-soluble vitamin, vitamin E, which is subject to some genetic control, is found mainly in the germ. The source of vitamin E is four tocopherols, of which alpha-tocopherol is the most biologically active. Gamma-tocopherol is probably more active as an antioxidant than alpha-tocopherol, however.

Water-soluble vitamins

Water-soluble vitamins are found mainly in the aleurone layer of the maize kernel, followed by the germ and endosperm. This distribution is important in processing, which, as will be shown later, induces significant losses of the vitamins. Variable amounts of thiamine and riboflavin have been reported. The content is affected by the environment and cultural practices rather than by genetic make-up. Variability between varieties has, however, been reported for both vitamins. The water-soluble vitamin nicotinic acid has attracted much research because of its association with niacin deficiency or pellagra, which is prevalent in populations consuming high amounts of maize (Christianson *et al.*, 1968). As with other vitamins, niacin content varies among varieties, with average values of about 20 µg per gram. A feature peculiar to niacin is that it is bound and therefore not available to the animal organism. Some processing techniques hydrolyze niacin, thereby making it available. The association of maize intake and pellagra is a result of the low levels of niacin in the grain, although experimental evidence has shown that amino acid imbalances, such as the ratio of leucine to isoleucine, and the availability of tryptophan are also important (Gopalan and Rao, 1975; Patterson *et al.*, 1980).

Maize has no vitamin B_{12}, and the mature kernel contains only small amounts of ascorbic acid, if any. Yen, Jensen and Baker (1976) reported a content of about 2.69 mg per kg of available pyridoxine. Other vitamins such as choline, folic acid and pantothenic acid are found in very low concentrations.

Changes in chemical composition and nutritive value during grain development

In many countries, immature maize is often used as a food, either cooked whole as corn on the cob or ground to remove the seed-coat, with the pulp used to make thick gruels or foods like *tamalitos*. The changes in chemical composition that take place upon maturation are important. All relevant studies have shown a decrease in nitrogen, crude fibre and ash on a dry-weight basis and an increase in starch and ether extract (e.g. Ingle, Bietz and Hageman, 1965). The alcohol-soluble proteins increase rapidly as the kernel matures, while acid- and alkali-soluble proteins decrease. During this biochemical process arginine, isoleucine, leucine and phenylalanine (expressed as mg per g N) increase, while lysine, methionine and tryptophan decrease with maturation. Gómez-Brenes, Elías and Bressani (1968) further showed a decrease in protein quality (expressed as protein efficiency ratio). Thus, immature maize should be promoted during weaning or for infant nutrition.

NUTRITIONAL VALUE OF MAIZE

The importance of cereal grains to the nutrition of millions of people around the world is widely recognized. Because they make up such a large part of diets in developing countries, cereal grains cannot be considered only as a source of energy, as they provide significant amounts of protein as well. It is also recognized that cereal grains have a low protein concentration and that protein quality is limited by deficiencies in some essential amino acids, mainly lysine. Much less appreciated, however, is the fact that some cereal grains contain an excess of certain essential amino acids that influence the efficiency of protein utilization. The classic example is maize. Other cereal grains have the same constraints but less obviously.

A comparison of the nutritional value of maize protein with the protein quality of eight other cereals is given in Table 15, expressed as percentages of casein. The protein quality of common maize is similar to that of the other cereals except rice. Both opaque-2 maize and the hard-endosperm QPM (Nutricta) have a protein quality not only higher than that of common maize, but also significantly higher than that of other cereal grains.

The reasons for the low quality of maize proteins have been extensively studied by numerous investigators. Among the first were Mitchell and Smuts (1932) who obtained a definite improvement in human growth when 8 percent maize protein diets were supplemented with 0.25 percent lysine. These results have been confirmed over the years by several authors (e.g. Howe, Janson and Gilfillan, 1965), while others (e.g. Bressani, Elías and

TABLE 15
Protein quality of maize and other cereal grains

Cereal	Protein quality (*% casein*)
Common maize	32.1
Opaque-2 maize	96.8
QPM	82.1
Rice	79.3
Wheat	38.7
Oats	59.0
Sorghum	32.5
Barley	58.0
Pearl millet	46.4
Finger millet	35.7
Teff	56.2
Rye	64.8

Braham, 1968) have shown that the addition of lysine to maize causes only a small improvement in protein quality. These differing results may be explained by variations in the lysine content of maize varieties. Work in this field led to the discovery by Mertz, Bates and Nelson (1964) of the high-lysine maize called opaque-2.

Some researchers (Hogan *et al.*, 1955) have reported that tryptophan rather than lysine is the first limiting amino acid in maize, which may be true for some varieties with a high lysine concentration or for maize products modified by some kind of processing. All researchers have agreed that the simultaneous addition of both lysine and tryptophan improves the protein quality of maize significantly; this has been demonstrated in experimental work with animals.

The improvement in quality obtained after the addition of lysine and tryptophan has been small in some studies and higher in others when other amino acids have been added. Apparently, the limiting amino acid after lysine and tryptophan is isoleucine, as detected from animal feeding studies (Benton, Harper and Elvehjem, 1955). Most researchers who reported such findings indicated that the effect of isoleucine addition resulted from an excess of leucine which interfered with the absorption and utilization of isoleucine (Harper, Benton and Elvehjem, 1955; Benton *et al.*, 1956). It has been reported that high consumption of leucine along with the protein in maize increases niacin requirements, and this amino acid could be partly responsible for pellagra.

When a response to threonine addition has been observed, it has been attributed to this amino acid's correction of amino acid imbalances caused by the addition of methionine. A similar role can be ascribed to added isoleucine resulting in improved performance. Similarly, the addition of valine, which results in a decrease in protein quality, could be counteracted by the addition of either isoleucine or threonine.

In any case, isoleucine seems to be more effective than threonine, producing more consistent results. A possible explanation for these findings is that maize is not deficient in either isoleucine or threonine. However, some samples of maize may contain larger amounts of leucine, methionine and valine, and these require the addition of isoleucine and threonine besides lysine and tryptophan to improve protein quality. In any case, the addition of 0.30 percent L-lysine and 0.10 percent L-tryptophan easily increases the protein quality of maize by 150 percent (Bressani, Elías and Braham, 1968). Many of the results of the limiting amino acids in maize protein are influenced by the level of protein in the maize. As was indicated previously, protein content in maize is a genetic trait that is affected by nitrogen fertilization. The observed increase in protein content is highly correlated with zein, or the alcohol-soluble protein, which is low in lysine and tryptophan and contains excessive amounts of leucine. Frey (1951) found a high correlation between protein content and zein in maize, a finding that has been confirmed by others. Using different animal species, various authors have concluded that the protein quality of low-protein maize is higher than that of high-protein maize when the protein in the diets used is the same. However, weight for weight, high-protein maize is slightly higher in quality

than low-protein maize. The levels of dietary protein, then, affect the response observed upon amino acid supplementation with lysine and tryptophan in particular but with other amino acids as well, such as isoleucine and threonine.

Chapter 3

Post-harvest technology: pre-processing

The chemical components and nutritive value of maize do not lose their susceptibility to change when the grain is harvested. Subsequent links in the food chain, such as storage and processing, may also cause the nutritional quality of maize to decrease significantly or, even worse, make it unfit for either human and animal consumption or industrial use.

DRYING

Maize harvesting is highly mechanized in developed countries of the world, while it is still done manually in developing countries. The mechanized system removes not only the ear from the plant but also the grain from the cob, while manual harvesting requires initial removal of the ear, which is shelled at a later stage. In both situations, maize is usually harvested when its moisture content is in the range of 18 to 24 percent. Damage to the kernel (usually during the shelling operation) is related to moisture content at harvest; the lower the moisture content, the less the damage.

Changes in the physical quality of the grain are often a result of mechanical harvesting, shelling and drying. The first two processes sometimes result in external damage, such as the breaking of the pericarp and parts around the germ, facilitating attack by insects and fungi. Drying, on the other hand, does not cause marked physical damage. However, if it is carried out too rapidly and at high temperatures, it will induce the formation of stress cracks, puffiness and discoloration, which will affect the efficiency of dry milling and other processes (Paulsen and Hill, 1985).

In tropical countries, drying is sped up by bending down the upper part of the plant holding the ear, a practice that also prevents the kernels from becoming soaked when it rains. In either mechanical or manual harvesting, the shelled kernels contain too much moisture for safe storage, and they must be dried to safe moisture levels of about 12 percent at 30°C and about 14 percent at 10°C (Herum, 1987). Storage stability depends on the relative humidity of the interstitial gases, which is a function of both moisture content in the kernel and temperature. Low moisture content and low storage temperatures reduce the opportunity for deterioration and microbial growth. Aeration therefore becomes an important operation in maize storage as a means of keeping down the relative humidity of interstitial gases.

Significant maize losses have been reported in tropical countries. Losses of up to 10 percent have been found, not including those losses caused by fungi, insects or rodents. If these were included, losses could go up to 30 percent in tropical humid areas or 10 to 15 percent in temperate areas. Schneider (1987) reported post-production losses in Honduras of 6.5 to 8.7 percent in the field and of 7.4 to 13.9 percent in storage. Losses due to fungi (mainly aspergillus and penicillium) are important for both economic and health reasons because of aflatoxins and mycotoxins (de Campos, Crespo-Santos and Olszyna-Marzys, 1980).

In a survey on maize sold in rural markets in Guatemala, Martinez-Herrera (1968) found considerable contamination by several fungi. Among these, some *Aspergillus* species, well known as aflatoxin producers, were frequently present. There is evidence that maximum aflatoxin contamination of maize in Guatemala is during the rainy season. Samples analysed 20 days after maize was harvested had levels of 130 μg aflatoxin per kg of total maize. The same samples analysed 60 days later showed a great increase of up to 1 680 μg per kg. These data as well as data from several other studies strongly indicate the need to dry maize before storage. Diverse drying systems and equipment are available, using various sources of energy including solar energy (Herum, 1987). A number of factors must be considered such as temperature and air velocity, rate of drying, drying efficiencies, kernel quality, air power, fuel source, fixed costs and management. Drying is an important step in ensuring good quality grain that is free of fungi and micro-organisms and that has desirable quality characteristics for marketing and final use.

Drying Methods

Layer drying. In this method, the harvested grain is placed in a bin one layer at a time. Each layer of grain is partially dried, before the next is added, by forcing air through a perforated floor or through a duct in the bottom of the bin. To improve efficiency, the partially dried grain is stirred and mixed with the new layer. An alternative is to remove the partially dried grain and dry it completely in batches. One of the problems with this and other methods of drying is in finding a way to mix low-moisture grain with high-moisture grain to get the desired equilibrium in the final product. Spoilage often occurs in this attempt. Sauer and Burroughs (1980) reported that equilibrium was more than 80 percent complete in 24 hours. Methods have been developed to detect high-moisture maize in mixtures with artificially dried maize.

Portable batch dryers. Since drying installations are costly, few maize producers, particularly small farmers, can afford to have their own. Portable batch dryers are useful since they can be moved from farm to farm. These dryers operate with air heated to 140 to 180°F (60 to 82°C).

Continuous flow dryers. The principle behind these dryers is the continuous flow of grain through heated and unheated sections so that it is discharged dry and cool. The equipment is the central point in grain storage depots.

STORAGE

Biotic and non-biotic factors

The efficient conservation of maize, like that of other cereal grains and food legumes, depends basically on the ecological conditions of storage; the physical, chemical and biological characteristics of the grain; the storage period; and the type and functional characteristics of the storage facility. Two important categories of factors have been identified. First are those of biotic origin, which include all elements or living agents that, under conditions favourable for their development, will use the grain as a source of nutrients and so induce its deterioration. These are mainly insects, micro-organisms, rodents and birds. Second are non-biotic factors, which include relative humidity, temperature and time. The effects of both biotic and non-biotic factors are influenced by the physical and biochemical characteristics of the grain. Changes during storage are influenced by the low thermal conductivity of the grain, its water absorption capacity, its structure, its chemical composition, its rate of respiration and spontaneous heating, the texture and consistency of the pericarp and the method and conditions of drying.

Nutrient losses have been reported in maize stored under unfavourable conditions. Quackenbush (1963) showed carotene losses in maize stored under different temperature and moisture conditions. In other studies common and QPM maize were stored in different types of containers with and without chemicals. After six months samples were examined for damage by insects and fungi and for changes in protein quality. In both types there was some damage to the unprotected maize but not to that stored with chemicals. Protein quality was not affected (Bressani *et al.*, 1982). Other changes subsequent to drying and storage included a decreased solubility of proteins; changes in nutritive value for pigs; changes in sensory properties (Abramson, Sinka and Mills, 1980); and changes in *in vitro* digestibility resulting from heat damage (Onigbinde and Akinyele, 1989).

Although damage caused by insects and birds is of importance, a great deal of attention has been paid to the problems caused by micro-organisms, not only because of the losses they induce in the grain, but more importantly, because of the toxic effects of their metabolic by-products on human and animal health.

Studies on the nutritional effects of insect infestation of maize are not readily available. Daniel *et al.* (1977) and Rajan *et al.* (1975) have reported losses in threonine and in protein quality of maize infested with *Sitophilus oryzae*. In the first study, protein efficiency ratio (PER) decreased after three months from an initial value of 1.30 to 0.91. In the second study, threonine decreased from 3.5 to 2.9 g per 16 g N and PER decreased from 1.49 to 1.16. These researchers also reported that the damaged maize was less efficient in complementing food legumes.

Also of nutritional significance was an increase in uric acid from 3.5 to 90.6 mg per 100 g after three months. Thiamine losses were detected as well.

Bressani *et al.* (1982) evaluated five chemicals and three types of containers for their effectiveness in protecting QPM's nutritional quality against insect damage. About 38 percent of the untreated grain (control) was damaged by insects. This did not, however, affect its protein quality.

Several research studies have identified an association between insect damage and toxin contamination (e.g. Fennell *et al.*, 1978; Pérez, Tuite and Baker, 1982).

Christensen (1967) measured selected changes in United States No. 2 maize stored for two years with moisture contents of 14.5 and 15.2 percent and at temperatures of 12, 20 and 25°C. Changes in condition were evaluated by appearance, fungal invasion, germination percentage and final fat acidity value. Samples stored at 25°C deteriorated rapidly at both levels of moisture content. The samples with 15.2 percent moisture changed slightly after six months at 12°C but appreciably after two years. The maize stored with 14.5 percent moisture content retained its original condition when kept at 12°C for the two-year period and changed only slightly in 18 months at 20°C. However, large variability in the insect-fungi interaction was observed. Some maize-growing regions have experienced extensive insect damage to maturing ears with no occurrence of aflatoxin, while other areas with equivalent insect damage have exhibited relatively broad incidences of the toxin in kernels at harvest.

Many studies have been conducted to assess the nutritional value of mouldy maize. Although some increase in B-vitamin content has been reported, possibly as a result of the metabolites of the micro-organisms, the damage to animal health far exceeds any beneficial change in chemical composition. Several researchers have studied the impairment in nutritive value of mould-damaged maize. For example, Martínez *et al.* (1970a) found significant negative effects in poultry and laboratory rats fed mouldy maize. It is difficult, however, to decide whether these effects were caused by fungi-produced toxins or by a loss in nutrients in the substrate because of their utilization by the micro-organisms.

Christensen and Sauer (1982) reviewed the effects of fungal invasion on cereal grains. They found that it reduced both the quality and grade of the grains through loss of dry matter, discoloration, heating, cooking, mushiness and contamination by mycotoxins. Microbial indices of fungal invasion and seed deterioration include visible damage, seed infection, number of fungal propagules, evolved carbon dioxide and decrease in seed germination and ergosterol content.

Inhibition of aflatoxin contamination

Two ways of preserving maize from being destroyed by aflatoxin contamination have been under investigation. One is to inhibit growth of *Aspergillus flavus* or *Aspergillus parasiticus* and the other is to remove the aflatoxins after they have been produced by the *Aspergillus* infection. Most researchers have concentrated on the inhibition of fungal growth, and some chemicals have already been found effective in storage conditions. This, however, does not solve the problem of field contamination by moulds, since the airborne spores of the organisms are readily available in the environment. The spores can germinate on the cob and infect the inner tissues under optimum temperature and moisture conditions. Therefore, other researchers have pursued the possibility of detoxification.

Roasting has been shown to be effective in reducing aflatoxin levels, depending on the initial level of the toxin as well as on roasting temperatures (Conway and Anderson, 1978). Higher temperatures may cause up to 77 percent aflatoxin destruction; however, it is well known that heat also destroys the nutritive value of the material. Tempering aflatoxin-contaminated maize with aqua ammonia and then roasting it may be a simple and effective way to decontaminate it. Valuable results using ammonia have been reported. It is difficult, however, to remove the smell of ammonia from the treated grain. Other more complex methods have been tried. For example, Chakrabarti (1981) showed that aflatoxin levels could be reduced to less than 20 ppb using separate treatments with 3 percent hydrogen peroxide, 75 percent methanol, 5 percent dimethylamine hydrochloride or 3 percent perchloric acid. These treatments, however, induced losses in weight and also in protein and lipids. Other methods include the use of carbon dioxide plus potassium sorbate and the use of sulphur oxide.

A process that has received some attention is the use of calcium hydroxide, a chemical used for lime-cooking of maize (Bressani, 1990). Studies have

shown a significant reduction in aflatoxin levels, although the extent of reduction is related to the initial levels. Feeding tests with mouldy maize treated with calcium hydroxide have shown a partial restoration of its nutritional value.

Appropriate harvesting and handling can do much to reduce fungal contamination of maize and can thus prevent the need for chemical decontamination measures, which not only increase the cost of the grain but cannot completely restore its original nutritional value. In this respect, Siriacha *et al.* (1989) found that if shelled grain was immediately sun-dried the chance of contamination was reduced as compared with that of undried maize shelled mechanically or by hand. Shelling encourages fungal contamination as it causes damage to the kernel base, which is rough compared with the rest of the grain. Corn on the cob, even with its high levels of moisture, resists fungal contamination relatively well.

CLASSIFICATION OF GRAIN QUALITY

To facilitate marketing and to identify the best uses for the various types of maize produced throughout the world, measures of grain quality have been identified, although they may not be accepted by all maize-producing countries. In the United States maize is classified into five different grades, based on several factors. Minimum test weight is expressed in pounds per bushel, pounds per cubic foot or kilograms per cubic metre. The higher the test weight the higher the grade. The maximum permitted amount of broken maize and foreign material (BCFM) varies from 2 percent for Grade 1 to 7 percent for Grade 5. There is a classification for damaged kernels that includes heat-damaged kernels. Maize is also classified as yellow, white or mixed maize. Yellow maize must have no more than 5 percent white kernels and white maize must not have more than 2 percent yellow grain. The mixed class contains more than 10 percent of the other grain.

Although the moisture content of maize, an important part of its chemical composition, is not considered a quality factor, it has much influence on composition, quality changes during storage and processing and economics. High-moisture maize with a soft texture is easily damaged in storage, while maize with low levels of moisture becomes brittle. The most commonly accepted moisture level for marketing purposes is 15.5 percent. Density of maize — weight per unit volume — is important in storage and transportation since it establishes the size of container for either purpose. Moisture content and density or test weight are related; the higher the moisture level the lower the specific density test weight. This characteristic of maize is also important for milling.

Another important quality characteristic of maize is its hardness, since this influences grinding power requirements, dust formation, nutritional properties, processing for food products and the yield of products from dry and wet milling operations. Hardness of maize is genetically controlled, but it can be modified by both cultural practices and post-harvest handling conditions. Many investigators have proposed methodologies for measuring hardness for a number of different applications (Pomeranz *et al.*, 1984, 1985, 1986). Maize varieties with a horny endosperm, such as flint and popcorn types, have hard kernels, while starchy and opaque maize varieties are soft. Some flint types are intermediate.

Finally, freedom of the kernel from fungi is recognized as a quality characteristic.

Chapter 4

Post-harvest technology: processing

FORMS OF MAIZE CONSUMPTION

Maize is consumed in many forms in different parts of the world, from maize grits, polenta and corn bread to popcorn and products such as maize flakes (Rooney and Serna-Saldivar, 1987). The grain is fermented to give *ogi* in Nigeria (Oke, 1967) and other countries in Africa (Hesseltine, 1979) and is decorticated, degermed and precooked to be made into *arepas* in Colombia and Venezuela (Instituto de Investigaciones Tecnológicas, 1971; Rodriguez, 1972).

In Egypt a maize flat bread, *aish merahra*, is widely produced. Maize flour is used to make a soft dough spiced with 5 percent ground fenugreek seeds, which is believed to increase the protein content, improve digestibility and extend the storage life of the bread. The dough is fermented all night with a sourdough starter. In the morning the dough is shaped into small, soft, round loaves, which are left for 30 minutes to "prove". Before baking the loaves are made into wide, flat discs. *Aish merahra* keeps fresh for seven to ten days if it is stored in airtight containers. A similar product called *markouk* is eaten in Lebanon.

Maize is also widely used to make beer. In Benin, for example, malt is obtained by germinating the grain for about five days. The malt is then exposed to the sun to stop germination. The grains are lightly crushed in a mortar or on a grinding stone. The malt is cooked and the extract is strained off, cooled and allowed to stand. After three days of fermentation it is ready to be drunk as beer (FAO, 1989).

The lime-cooking process for maize is particular to Mexico and Central America (Bressani, 1990), although today the technology has been exported to other countries such as the United States. A dough prepared from lime-cooked maize is the main ingredient for many popular dishes such as *atole*, a beverage with a great variety of flavours, and *tamalitos*, made by wrapping the dough in maize husks and steam-cooking it for 20 to 30 minutes to gelatinize the starch. This form is usually prepared with young *chipilín* leaves (*Crotalaria longirostrata*), the flowers of *loroco* (*Fernaldia pandurata*) or cooked beans mixed with the dough, thus improving the nutritional quality of the product and its flavour (Bressani, 1983). The dough is also used for *tamales*, a more complex preparation because of the number of ingredients it contains, in most cases with chicken or pork meat added to the gelatinized dough. It is also used to provide support for enchiladas, tacos (folded tortillas containing meat, etc.) and *pupusas*, the latter made with fresh cheese placed between two layers of dough and baked like tortillas. When the dough is fried and flavoured, it yields foods such as chips and *chilaquiles*. If the dough is allowed to ferment for two days, wrapped in banana or plantain leaves, it provides a food named *pozol* from which a number of drinks can be made. It has been claimed that this preparation is of high nutritional quality.

There are many ways to convert maize into interesting and acceptable forms which, if presented in attractive and easily prepared products, could to some extent counteract the trend toward greater consumption of wheat-derived foods in *arepa*- and tortilla-eating countries and elsewhere.

PROCESSING OF WHOLE MAIZE: LIME-COOKING

Lime-cooking in rural areas

A number of researchers have described how maize is cooked in rural areas of countries where tortillas are eaten. Illescas (1943) first described the process as carried out in Mexico. It involves the addition of one part whole maize to two parts of approximately 1 percent lime solution. The mixture is heated to 80°C for 20 to 45 minutes and then allowed to stand overnight. The following day the cooking liquor is decanted and the maize, now referred to as *nixtamal*, is washed two or three times with water to remove the seed-coats, the tip caps, excess lime and any impurities in the grain. The addition of lime at the cooking and steeping stages helps to remove the seed-coats.

The by-products are either thrown away or fed to pigs. Originally, the maize was converted into dough by grinding it a number of times with a flat stone until the coarse particles were fine enough. Today the initial grinding is done with a meat grinder or disc mills and the dough is then refined with the stone. A portion of about 50 g of the dough is patted flat and cooked on both sides on a hot iron or clay plate.

In Guatemala a similar process (described by Bressani, Paz y Paz and Scrimshaw, 1958) uses either white or yellow maize, but the lime concentration varies from 0.17 to 0.58 percent based on the weight of maize, with a grain-to-water ratio of 1:1.2, and the maize cooking time varies from 46 to 67 minutes at a temperature of 94°C. The rest of the process is essentially the same, except that the dough is prepared with a disc mill and is cooked for about 5 minutes at a temperature of about 170°C at the edges and 212°C in the centre.

Tamalitos, for which the dough is steamed, are softer and keep longer. For recently harvested maize less lime is used and cooking time is decreased; the procedure is modified conversely when the grain is old and dry. The dry matter losses are about 15 percent , but they can vary between 8.9 and 21.3 percent.

Industrial lime-cooking

Factors such as the migration of people from rural to urban areas created a demand for ready-cooked or pre-cooked tortillas. Equipment for processing raw maize into lime-treated maize and then into a dough and tortillas was developed and industrial production of tortilla flour began in Mexico and other countries. Mechanized production in Mexico became important soon after the Second World War. Two types of industry are found in urban areas. One is the small family-owned home tortilla industry, where the process is as described above but with larger and mechanical equipment used to supply a larger market. This development became possible through the introduction of rotary mills and the tortilla maker designed by Romero in 1908. This equipment was later replaced by a more efficient type in which the dough is passed through a rotating metal drum where it is cut into tortilla shapes. These fall onto a moving belt or continuous cooking griddle, dropping into a receptacle at the end of the belt. This small industry may use whole maize, in which case the dough is cooked in large receptacles, or it may start with industrial tortilla flour.

The second type of industry is the large industrial conversion of maize into an instant precooked tortilla flour. The process has been described by various workers (e.g. Deschamps, 1985). It is based for all practical purposes on the traditional method used in rural areas. More recently, the process of producing the flour has been expanded to produce tortillas.

Maize is bought after the buyer has inspected its quality and sampled it. Batches of maize with a high percentage of defective grains are rejected. Those that are accepted are paid for according to the defects found in the raw material. Maize is also selected according to its moisture content, since very high moisture will result in storage problems. During the cleaning stage, all impurities such as dirt, cobs and leaves are removed. The cleaned maize is sent to silos and warehouses for storage.

From there it is conveyed to treatment units for lime-cooking. There it is converted into *nixtamal*, using either a batch or a continuous process. After cooking and steeping, the lime-treated maize is washed with pressurized water or by spraying. It is ground into a dough (*masa*) which is then transferred to a dryer and made into a rough flour. This flour, consisting of particles of all sizes, is forced through a sifter where the coarse particles are separated from the fine ones. The coarse particles are returned to the mill for regrinding and the fine ones, which constitute the final product, are sent to the packing units. Here the flour is packed into lined paper bags.

One complete unit must have equipment for lime treatment, milling, drying and sifting and a daily production capacity of 30 to 80 tonnes of flour. These figures are the minimum and the maximum; to increase its production capacity, a commercial enterprise must install several parallel units. The use of such units seems to be more a tradition than a technical necessity, since it would be perfectly feasible to design plants with a capacity lower than 30 tonnes or higher than 80 tonnes per day. Plants that are very large or very small are apparently not considered viable.

The industrial yield of alkali-cooked maize flour fluctuates between 86 and 95 percent depending on the type of maize, the quality of the whole

kernels and the lime-treatment conditions. Industrial yields have been reported to be higher than those at the rural and semi-industrial levels, possibly because of the quality of the grain processed.

Tortilla flour is a fine, dry, white or yellowish powder with the characteristic odour of maize dough. This flour when mixed with water gives a suitable dough for the preparation of tortillas, *tamales, atoles* (thick gruels) and other foods. All maize flours made in Mexico must conform to the specifications of the government's Department of Standards and Regulations.

When the flour has a moisture content of 10 to 12 percent it is stable against microbial contamination. If the moisture content is over 12 percent it is easily attacked by moulds and yeast. The problem of bacterial attack is almost nonexistent since the minimum of moisture required for bacterial growth is so high that flour with this moisture content would already be transformed into *masa*. Another matter related to the stability of flour is rancidity, which is normally not a problem unless the flour is packed at high temperatures. The minimum time required for the flour to spoil in Mexico is four to six months during the winter and three months during the summer. Nevertheless, it is usually sold to the consumer within 15 days of being sold to retailers and wholesalers, while its shelf-life is one month (Delvalle, 1972).

Tortillas made from lime-treated maize flour can be made at home or in factories. Such flour has been a great advantage for households and for factories both large and small, although its use in rural areas is not widespread.

In Guatemala, about 3 000 metric tons of maize are produced yearly for tortilla flour production. This amount is significantly lower than that in Mexico; the population is smaller and there are few small tortilla factories. About 90 percent of the production is sold in urban areas and 75 percent goes into tortilla making. Other countries where lime-treated maize flour is produced are Costa Rica and the United States. In Costa Rica tortilla consumption per person is about 25.6 kg per annum. Approximately 62 percent of the production is commercial, 30.6 percent is home-made from commercial flour and 7.4 percent is home-made from grain.

Modifications of lime-cooking

The traditional method of cooking maize with lime to make tortillas at the rural level is both time-consuming (about 14 to 15 hours) and hard work. The cooking and soaking operations take up 70 to 80 percent of the time, which in a sense may be acceptable to the rural housewife. Nevertheless, the availability of an instant tortilla flour offers many advantages such as convenience, less labour and lower use of energy, for a safe, stable and nutritious product. At the industrial or commercial level, grinding and dehydration are large factors in the cost. Lime-cooked maize contains about 56 percent moisture, which must be decreased to 10 to 12 percent in the flour. Therefore, any method that would decrease both time and cost and still yield acceptable tortillas would be advantageous.

Efforts in this respect have been made by a number of workers. Bressani, Castillo and Guzmán (1962) evaluated a process based on pressure cooking at 5 and 15 lb pressure per square inch (0.35 and 1.05 kg per cm^2) under dry and moist conditions for 15, 30 and 60 minutes, without the use of lime. None of the treatments had any effect on chemical composition and true protein digestibility, but all reduced the solubility of the nitrogen. Pressure cooking at 15 lb per square inch (1.05 kg per cm^2) under dry conditions reduced the nutritional quality of the product, particularly when carried out for 60 minutes. The pressure cooking method without lime did not reduce crude fibre content, which is one of the particular effects of lime, and the calcium content was significantly lower than in dry dough (*masa*) prepared by the traditional method.

Khan *et al.* (1982) conducted a comparative study of three lime-cooking methods: the traditional way, a commercial method and a laboratory pressure-cooking procedure. For each process maize was undercooked, optimally cooked and overcooked to measure some of the physical and chemical changes that might occur. Although the traditional method caused the greatest loss of dry matter from the grain, it gave the best tortillas in terms of texture, colour and acceptability. The pressure-cooking procedure yielded a sticky dough and undesirable tortillas. The commercial method was the least desirable. This study allowed the authors to propose a method to evaluate the completeness of cooking.

Bedolla *et al.* (1983) tested various methods of cooking maize and sorghum as well as mixtures of the two grains. The methods tested included the traditional one, steam cooking as tested by Khan *et al.* (1982) and a method using a reflux (condensing) system. They found that the methods of cooking affected the total dry matter lost during processing into tortillas.

Variation of cooking conditions can result in lower processing times. For example, Norad *et al.* (1986) found that a 40 percent reduction in cooking time could be achieved by pre-soaking the grain before alkali cooking. In these studies dry matter losses, water uptake, calcium content and enzyme-susceptible starch increased, whereas amylograph maximum viscosity decreased in both pre-soaked and raw maize upon cooking. The decrease in viscosity and increase in the other parameters was faster in the pre-soaked maize.

Dry-heat processes have also been studied. Johnson, Rooney and Khan (1980) tested the micronizing process to produce sorghum and maize flours. Micronizing is a dry-heat process using gas-fired infrared generators. Rapid internal heating takes place, cooking the product from the inside out. The authors used this process to produce tortilla flour, claiming that it would be quicker and more economical than the traditional method.

Molina, Letona and Bressani (1977) tested production of instant tortilla flour by drum drying at the pilot plant level. Maize flour was mixed with water at a ratio of 3:1 with 0.3 percent lime added on the basis of maize weight. After mixing, the dough was passed through a double-drum dryer heated with steam at 15, 20 or 25 lb per square inch (1.05, 1.40 or 1.75 kg per cm^2), 93, 99 and 104°C surface temperature and 2, 3 or 4 rpm. The process produced an instant tortilla flour with physico-chemical and organoleptic characteristics identical to those of the reference sample prepared by the traditional method but different from those of a commercial product.

Extrusion cooking has also been evaluated as an additional technology for producing tortilla flour. Bazua, Guerra and Sterner (1979), using a Wenger X-5 extruder, processed ground maize mixed with various lime concentrations (0.1 to 1.0 percent). The dough and tortillas made by extrusion were compared with those made by the traditional process for their organoleptic properties as well as lysine, tryptophan and protein content. No appreciable differences were noted at comparable use levels of calcium hydroxide. Both the traditional process and the extrusion modification induced losses of tryptophan related to the amount of lime added. With a 0.2 percent addition 8 percent of the tryptophan was lost, while with 1 percent lime more than 25 percent was lost. Some lysine losses were also observed. The organoleptic results suggested that it is possible to make culturally acceptable tortillas using extrusion as an alternative to the lime-heat treatment.

Maize for tortillas

Grain quality is a concept now growing in importance in breeding programmes aimed at increasing acceptance of genetically improved seeds by farmers as well as by consumers and food processors. The grain quality characteristics include yield, technological properties and, when possible, nutritional elements as well. Technological properties include stability during storage, efficiency of conversion into products under given processing conditions and acceptability to the consumer. The technological aspect of maize quality for tortilla preparation is of little importance to small farmers in the least developed countries, who seldom use seed other than that kept from harvest to harvest. Furthermore, the rural housewife knows how to adjust cooking conditions to the type of maize she will process for consumption. But maize is now being converted into a tortilla flour using industrial processes, where the grain being used may be of different varieties from various producers and different environments. It may have a variety of structures or may have been poorly handled after harvest, factors which influence the yield and physico-chemical and organoleptic as well as culinary properties of the product. This would appear to be of growing importance in countries such as the United States where the maize tortilla is becoming a very popular food.

That physical characteristics of maize are important became clear some time ago, when Bressani, Paz y Paz and Scrimshaw (1958) showed that the yield of dry matter in the form of dried-maize dough or flour was affected by the maize cultivar. In their rural home studies dry matter losses from

white maize averaged 17.2 percent with a variability of 9.5 to 21.3 percent. Dry matter losses from yellow maize averaged 14.1 percent, with a range from 8.9 to 16.7 percent.

Cortez and Wild-Altamirano (1972) conducted a series of measurements on 18 cultivars of maize produced in Mexico. These included kernel weight, colour and lime-cooking time using a standard cooking procedure with 1.5 percent lime at 80°C and a steeping time of 12 hours. Cooking efficiency and time were measured by the ease with which the seed coat could be removed. Evaluations of the cooked maize included measurement of the volume of 1 kg of maize, the yield of dough from 1 kg of grain and the moisture content of the dough. The dough was further evaluated by measuring its strength and water absorption. The dehydrated dough was then ground to 60-mesh size and evaluated for moisture, colour, specific volume and other physical characteristics using a mixograph. The tortillas made from the dough of each maize sample were further evaluated for extensibility, volume, plasticity, softness and roughness of the surface.

From this extensive study, the authors reached several conclusions. Maize varieties or cultivars of higher weight per volume, a harder endosperm, more moisture and a high protein content produced the best tortillas. Two cultivars of popcorn maize were among the best types for tortillas. The Swanson mixograph was useful in establishing differences in maize types. The time required to cook the samples ranged from 30 to 75 minutes, and dry matter losses ranged from 10 to 34 percent. Rooney and Serna-Saldivar (1987) found that maize with hard or corneous endosperm required a longer cooking time. Bedolla and Rooney (1984) stated that the texture of the dough was affected by the endosperm texture and type, drying, storage and soundness of the maize kernel. Martínez-Herrera and Lachance (1979) established a relationship between kernel hardness and the time needed for cooking. They reported that within a maize variety, higher calcium hydroxide concentration slightly decreased cooking time. Furthermore, knowing the initial hardness of a variety made it possible to predict the time required to cook it. Khan *et al.* (1982) and Bedolla and Rooney (1982) measured a parameter termed *nixtamal* shear force (NSF), an indication of kernel hardness. The measurement was related to both cooking time and processing method. These authors showed that the NSF measurement could reveal small differences in maize with similar endosperm texture and could be used to predict optimum cooking time.

Dry matter losses resulting from lime-cooking constitute a good index of maize quality for tortilla preparation. Jackson *et al.* (1988) reported that greater losses resulted from stress-cracked and broken kernels than from sound kernels. Therefore they concluded that any system for assessing maize for alkaline cooking should include measures of broken kernels, the potential for breakage and ease of pericarp removal. Specific studies on the effects of drying and storage on quality of maize for tortilla making are not readily available. Bressani *et al.* (1982) reported on QPM storage as related to tortilla quality. The Nutricta QPM variety was stored under a number of field or rural conditions. Containers made of cloth not treated with insecticides allowed insect infestation and therefore higher dry-matter losses during cooking; but the protein quality was not affected.

Possibly the most interesting feature of the process of converting maize into tortillas is the use of an alkaline medium, and particularly calcium hydroxide. The most obvious effect of adding lime is the facilitation of seed-coat removal during cooking and steeping. According to Trejo-González, Feria-Morales and Wild-Altamirano (1982), added lime maintains an alkaline pH, which is needed to hydrolyse the hemicelluloses of the pericarp. Lime uptake by the kernel follows that of water, but the rate is lower than that of water. Norad *et al.* (1986) showed that soaking the kernels before cooking led to a higher calcium content in the grain. Calcium content of *masa* was affected by lime levels and also by cooking-steeping temperatures. Several other authors (e.g. Pflugfelder, Rooney and Waniska, 1988a) have shown in one way or another that lime uptake during alkaline cooking is affected by physical and chemical characteristics of maize dough.

Martínez-Herrera and Lachance (1979) found that higher calcium hydroxide concentrations slightly decreased cooking time, but the differences were not statistically significant. These authors also reported an interaction between maize variety and calcium hydroxide concentration. However, the coefficient of variation was high (29.1 percent); this was attributed to inherent variability in the kernels of the different varieties.

Bedolla and Rooney (1982) reported that increases in cooking time, cooking temperature, lime concentration and steeping time produced lower viscoamylograph peak viscosities at both 95 and 50°C, which was interpreted to mean a greater degree of starch gelatinization. Trejo-González, Feria-Morales and Wild-Altamirano (1982) showed that calcium was fixed or was bound in some way to the starch of the maize kernel. Other effects included greater solid losses with increasing amounts of lime; changes in colour, aroma and flavour; and a delay in the development of acidity, which extends shelf-life. If added in exceedingly large amounts, lime affects organoleptic properties of the food; this effect is often observed when maize has been stored for a long time.

OGI AND OTHER FERMENTED MAIZE PRODUCTS

Acid porridges prepared from cereals are eaten in many parts of the world, particularly in developing countries, where they may form part of the basic diet. Some examples of acid porridges include *pozol* in Mexico and Guatemala, *ogi* in Nigeria, *uji* in Kenya and *kenkey* in Ghana. These porridges are usually made from fermented raw or heat-treated maize, although sorghum and millet are often used.

Ogi manufacture

The traditional process of making *ogi* has a number of slight variations described by several authors. *Ogi* is traditionally prepared in batches on a small scale two or three times a week, depending on demand. The clean grain is steeped in water for one to three days to soften. Once soft, it is ground with a grinding stone, pounded in a mortar or ground with a power mill. The bran is sieved and washed away from the endosperm with plenty of water. Part of the germ is also separated in this operation. The filtrate is allowed to ferment for 24 to 72 hours to produce a slurry which when boiled gives the *ogi* porridge. *Ogi* is usually marketed as a wet cake wrapped in leaves, or it may be diluted to 8 to 10 percent solids in water and boiled into a pap or cooked to a stiff gel.

Akinrele (1970) reported that the souring of the maize took place spontaneously without the addition of inoculants or enzymes. He identified the organisms involved in this unaided fermentation and investigated their effects on the nutritive value of the food. He identified the moulds as *Epholosporium, Fusarium, Aspergillus* and *Penicillium* species and the aerobic bacteria as *Corynebacterium* and *Aerobacter* species, while the main lactic acid bacterium he found was *Lactobacillus plantarum*. There were also yeasts: *Candida mycoderma, Saccharomyces cerevisiae* and *Rhodotorula* sp.

Although *ogi* is supposed to have an improved B-vitamin content, the results observed are quite variable, at least for thiamine, riboflavin and niacin. Banigo and Muller (1972) identified the carboxylic acids of *ogi* fermentation. They found 11 acids, with lactic, acetic and butyric acids being the most important.

The *ogi*-making process is quite complex, and the porridge can also be prepared from sorghum, rice, millet and maize. Therefore, laboratory procedures have been developed to learn more about the process and introduce changes to convert the grains to food more efficiently. These have been described by Akingbala, Rooney and Faubion (1981) and Akingbala *et al.* (1987), whose studies have been useful also in evaluating varieties of cereal grains for their efficiency in making *ogi*. The authors also reported on the yields of *ogi* from whole maize kernels (79.1 percent) and dry milled flour (79.8 percent).

The commercial manufacture of *ogi* does not differ substantially from the traditional method. Modifications have been introduced, such as the dry milling of maize into a fine meal or flour and subsequent inoculation of the flour-water mixture with a culture of lactobacilli and yeast. In view of the importance of *ogi* in the Nigerian diet, large-scale production is indicated. The material could be dried and packaged in polythene bags for a good shelf-life. There is some problem in achieving a controlled fermentation with pure cultures. Some modifications include spray-drying the slurry or drum-drying.

Other fermented maize products

Ogi has a number of other names such as *akamu* or *ekogbona, agidi* and *eko tutu*. These, with the Kenyan *uji* and Ghanaian *koko*, are substantially the same preparation with changes in the grain used or some modification of the

basic process. For the Mexican *pozol*, maize is processed with lime as for tortillas. The *nixtamal*, or cooked maize without the seed-coat, is ground to a coarse dough which is shaped into balls by hand. The balls are then wrapped in banana leaves to avoid drying and are allowed to ferment for two to three days, or more if necessary. The micro-organisms involved are many.

AREPAS

Another major food made from maize, used daily in Colombia and Venezuela, is *arepa*. Mosqueda Suarez (1954) and Cuevas, Figueroa and Racca (1985) described the traditional preparation method as practised in Venezuela. De Buckle *et al.* (1972) defined the Colombian *arepa* as roasted maize bread without yeast, round in shape, prepared from maize that has been degermed. Whole maize is dehulled and degermed using a wooden bowl called a *pilon* and a double-headed wooden mallet. The moistened maize is pounded until the hulls and part of the germ are released from the endosperm. The hulls and germs are removed by adding water to the mixture containing the endosperm. The endosperm is cooked and then stone-milled to prepare a dough. Small portions of this dough are made into balls, then pressed into flat discs which are cooked rapidly on both sides.

The traditional method of preparing *arepas* has been substantially modified by the introduction of precooked maize flour, which reduces the time from 7 to 12 hours to 30 minutes (Cuevas, Figueroa and Racca, 1985). There are two stages in the industrial process. The first is the preparation of maize grits by cleaning, dehulling and degerming the maize; the second is the processing of the grits to produce precooked flour. Efforts have been made to modify the process even further by extrusion cooking.

OTHER MAIZE PREPARATIONS

In Latin America there are many maize-based foods besides tortillas and *arepas*. Some of these are drinks like *colados, pinol* and *macho*, basically suspensions of cooked maize flour. These three products have a very low protein quality. The production of *humitas*, a *tamale*-like food consumed in Bolivia and Chile, was described by Camacho, Bañados and Fernandez (1989). Made from immature common or opaque-2 maize to which is added a number of other ingredients, *humitas* is produced from precooked maize flour which resembles the lime-treated *masa*. Other products include *mote*, made from cooked maize and cheese, *pupusas*, made from lime-treated maize and cheese, and *patasca*, which is like a lime-treated maize kernel. From immature maize a sweet, tasty *atole* of high nutritive value is made; Khan and Bressani (1987) described the process, which consists of grinding the maize in water followed by filtration and cooking. Immature maize, either common or opaque-2, and sweet maize are also extensively consumed. Chavez and Obregon (1986) reported on the incorporation of the opaque-2 gene into sweet maize to provide a food of high nutritional quality.

Maize has also been used as a substrate for fermented beverages called *chicha*. Cox *et al.* (1987) have reported on the microflora of these fermented products, which are made by basically the same process but using a variety of additives.

MILLING

The maize kernel is transformed into valuable foods and industrial products by two processes, dry milling and wet milling. The first yields grits, meal and flours as primary products. The second yields starch and valuable derived products.

Dry milling

The dry milling of maize as practised today has its origins in the technologies used by the native populations who domesticated the plant. The best example is the method used to make *arepa* flour or hominy grits. The old technology was soon replaced by a grinding stone or stone mill, followed by the grits mill and finally by sophisticated tempering-degerming methods. The products derived are numerous, with their variety depending to a large extent on particle size. They are classified into flaking grits, coarse grits, regular grits, corn meal, cones and corn flour by means of meshes ranging from 3.5 to 60. Their chemical composition has been well established and their uses are extensive, including brewing, manufacturing of snack foods and breakfast cereals and many others.

Wet milling

The largest volume of maize in developed countries such as the United States is processed by wet milling to yield starch and other valuable by-products such as maize gluten meal and feed. The starch is used as a raw material for a wide range of food and non-food products. In this process clean maize is soaked in water under carefully controlled conditions to soften the kernels. This is followed by milling and separation of the components by screening, centrifugation and washing to produce starch from the endosperm, oil from the germ and food products from the residues. The starch has industrial applications as such and is also used to produce alcohol and food sweeteners by either acid or enzymatic hydrolysis. The latter is done with bacterial and fungal alpha-amylase, glucoamylase, beta-amylase and pullulanase. Saccharides of various molecular weights are liberated yielding sweeteners of different functional properties. These include liquid or crystalline dextrose, high-fructose maize syrups, regular maize syrups and maltodextrins, which have many applications in foods.

Chapter 5

Physical and chemical changes in maize during processing

LIME-TREATED MAIZE

Chemical changes

The conversion of maize into tortillas involves a process in which water, heat and calcium hydroxide are used. All three influence the chemical composition of processed maize, causing changes in nutrient content. The changes that take place are caused by both physical losses of the kernel and chemical losses. The latter may result from destruction of some nutrients and chemical transformation of others.

The proximate composition of maize and of home-made and industrially prepared tortillas is shown in Table 16. Changes in fat and crude fibre content are shown, and in some cases an increase in ash content. The values for home-made and industrially produced tortillas are similar for most major chemical components with the exception of fat, which is higher in industrially produced tortillas.

Dry matter losses

From studies on maize cooking by rural housewives using their own traditional method, Bressani, Paz y Paz and Scrimshaw (1958) reported a loss of solids (17.1 percent for white maize and 15.4 percent for yellow maize) when maize was made into dough. Bedolla and Rooney (1982) have reported losses of 13.9 and 10 percent respectively for white and yellow maize using the traditional process and losses of 7 and 5.7 percent in steam cooking. In other studies where variations in the processing technique were evaluated, Khan *et al.* (1982) found losses of 7 to 9 percent in commercial processing, 9 to 11 percent in pressure cooking and 11 to 13 percent using

TABLE 16

Proximate composition of raw maize and home-made and industrially produced tortillas

Product	Moisture (%)	Protein (%)	Fat (%)	Ash (%)	Crude fibre (%)	Carbo-hydrates (%)	Calories per 100 g
Maize							
White	15.9	8.1	4.8	1.3	1.1	70.0	356
Yellow	12.2	8.4	4.5	1.1	1.3	73.9	370
White	13.8	8.3	-	1.2	-	-	-
Tortillas							
White	47.8	5.4	1.0	0.8	0.7	44.5	204
Yellow	47.8	5.6	1.3	0.8	0.6	44.4	212
White	41.9	5.8	-	0.9	-	-	-
Industrial	40.5	5.8	0.9	1.1	1.4	50.3	226
Industrial	44.0	5.3	3.4	1.2	0.7	42.8	215
Industrial	45.2	5.2	3.1	1.4	1.1	41.1	206

Sources: Bressani, Paz y Paz and Scrimshaw, 1958; Cravioto *et al.*, 1945; Ranhotra, 1985; Saldana and Brown, 1984

the traditional method. These workers also reported that dry matter loss increased as cooking time increased.

Likewise, the integrity of the maize kernel influences losses. Jackson *et al.* (1988) reported that dry matter losses in the traditional cooking procedure were higher (10.8 to 12.1 percent) with broken kernels than with undamaged ones (6.3 to 8.9 percent). Besides the integrity of the kernel and the heating process used, other factors such as length of steeping influence dry matter losses. Long steeping caused larger losses than a short steeping time. Dry matter losses of QPM with a hard endosperm are similar to those of common maize. Recently, Bressani *et al.* (1990) reported losses of 17.1 percent for the Nutricta QPM variety as compared with 17.6 percent from a white tropical maize. Sproule *et al.* (1988) found a 9.6 percent dry

matter loss from QPM as compared with a 10.4 percent loss in common maize.

Dry matter losses depend, then, on a number of variables such as the type of maize (hard or soft endosperm), kernel integrity (whole or broken kernels), cooking procedure (traditional, steam cooking, pressure cooking, commercial), the levels of lime used, cooking time and steeping time, as well as other operations such as rubbing to eliminate the seed-coat during washing of the kernels. This process also eliminates other parts of the kernel: the tip cap and possibly the aleurone layer and small amounts of germ. Paredes-López and Saharopulus-Paredes (1983) used scanning electron microscopy to show that the outside surface of lime-treated maize had important structural deterioration. They indicated that the aleurone layer was retained as well as some pericarp layers and that the germ remained attached to the endosperm. Gómez *et al.* (1989) noted that important structural changes took place in maize during "nixtamalization". The alkali weakened the cell walls, facilitating the removal of the pericarp. It solubilized the cell wall in the peripheral endosperm, caused swelling and partial destruction of starch granules and modified the appearance of the protein bodies. The dough contained fragments of germ, pericarp, the aleurone and endosperm, as well as free starch and dissolved lipids. Thus some of the chemical changes that have been observed can be accounted for by the chemical compounds present in these three or four parts of the kernel. The dry matter content has been analysed by Pflugfelder, Rooney and Waniska (1988a), who reported 64 percent non-starch polysaccharides (fibre), 20 percent starch and 1.4 percent protein.

Nutrient losses

Studies on the losses of nutrients during the transformation of maize into tortillas are not abundant, even though significant changes due to processing do take place (Cravioto *et al.*, 1945; Bressani, Paz y Paz and Scrimshaw, 1958). Ether-extractable substances are lost, 33 percent in yellow maize and 43 percent in white maize. This is difficult to explain, although it could be partially accounted for by the loss of the pericarp, the aleurone layer, the tip cap and some of the germ, parts of the kernel containing ether-extractable substances. Losses in crude fibre were reported to be about 46 percent in white maize and 31 percent in yellow maize. Lime treatment at 96°C for about 55 minutes hydrolyses the pericarp, which is removed during washing, pulling the tip cap with it, and this would account to a large extent for fibre loss. Nitrogen losses amount to about 10 and 5 percent for white and yellow maize, respectively. Again, this may be partly due to the physical loss of the pericarp and tip cap. Even though tortillas may have a slightly higher protein content than the original maize on an equal moisture basis, as has been reported by various workers, this may be caused by a concentration effect, since soluble sugars from the kernel are lost. Ash content increases because of the absorption of lime, which significantly increases calcium content (Saldana & Brown, 1984; Ranhotra, 1985). Significant losses take place in thiamine (52 to 72 percent), riboflavin (28 to 54 percent) and niacin (28 to 36 percent). In yellow maize 15 to 28 percent of the carotene was lost (Cravioto *et al.*, 1945; Bressani, Paz y Paz and Scrimshaw, 1958).

Fat and fatty acids. Ether-extractable substances of 33 and 43 percent were reported by Bressani, Paz y Paz and Scrimshaw (1958) from yellow and white maize respectively, as processed in Guatemalan rural homes. Pflugfelder, Rooney and Waniska (1988b) found losses of 11.8 to 18.1 percent and suggested that these could be partly due to the vigorous handling of cooked maize at the industrial plant. Of the total *masa* lipid, 25 to 50 percent was free and partially emulsified. Bedolla *et al.* (1983) found ether extract values of 5.0, 3.1 and 3.6 percent in raw maize, cooked maize and tortillas respectively, or about a 28 percent change. This loss has not been fully explained; however, it may result from the loss of the seed-coat, the tip cap, the aleurone layer and possibly part of the germ, and also from ether-soluble substances, not necessarily fat. Even though ether-extractable substances are lost in the process of converting maize into tortillas, the fatty acid make-up of the fat does not change in common maize or QPM, as shown in Table 17. Differences between maize samples, either raw or processed, are larger than those between raw maize and tortillas, suggesting that the alkaline cooking method does not alter the fatty acid make-up of the fat.

TABLE 17

Fatty acid content of common and quality protein maize and tortillas (%)

Product	C16:0	C18:0	C18:1	C18:2
Common maize	12.89	2.92	37.08	47.10
Opaque-2 maize	15.71	3.12	36.45	43.83
Common maize tortilla	13.63	2.95	37.14	45.76
Opaque-2 tortilla	15.46	3.25	35.84	43.03

Source: Bressani *et al.*, 1990

Fibre content. The crude fibre content of maize – as determined by the Association of Official Analytical Chemists (AOAC) methodology – decreases as the kernel is converted into tortillas. Various investigators (e.g. Saldana and Brown, 1984) have explained how and why such a loss takes place. With newer methodology to determine fibre, Reinhold and Garcia (1979), using the Van Soest method, reported that the neutral detergent fibre (NDF) and acid detergent fibre (ADF) in tortillas (6.60 and 3.75 percent, respectively, on a dry weight basis) were significantly higher than those found in the dough (an average of 5.97 and 2.98 percent, respectively). No difference was reported in hemicellulose, with dough containing 3.18 percent and the tortillas 2.89 percent. Using the same method, Bressani, Breuner and Ortiz (1989) found 10.8 percent NDF in maize and 9 percent in tortillas, as well as ADF of 2.79 and 3 percent respectively. Hemicellulose averaged 8 percent in maize and 6 percent in tortillas, while the values for lignin were 0.13 and 0.15 percent. These values and others are shown in Table 18. Using the method of Asp *et al.* (1983), Acevedo and Bressani (1990) detected a decrease in insoluble fibre from raw maize (13 percent) to the dough (6 percent) and an increase in tortillas (7 percent). Soluble fibre increased from 0.88 percent in raw maize to 1.31 percent in the dough, and further increased to 1.74 percent in tortillas. Fibre decreases from raw maize to dough are due to the losses in seed-coat described previously. Increases

TABLE 18

Dietary fibre in common and quality protein maize and tortillas (%)

Product	Insoluble dietary fibre	Soluble dietary fibre	Total dietary fibre	Neutral detergent fibre	Acid detergent fibre	Hemi-cellulose	Lignin
Raw common maize	11.0	1.4	12.4	10.8	2.8	8.0	0.13
Common maize tortilla	9.5	1.4	10.9	9.0	3.0	6.0	0.15
Raw QPM	13.8	1.1	14.9	–	–	–	–
QPM tortilla	10.3	1.9	12.2	–	–	–	–
Other tortilla	3.4	–	–	6.6	3.7	2.9	–
Other tortilla	4.1	–	–	–	3.8-5.0	–	–

Sources: Acevedo and Bressani, 1990; Bressani, Breuner and Ortiz, 1989; Bressani *et al.*, 1990; Krause, 1988; Ranhotra, 1985; Reinhold and Garcia, 1979

from dough to tortillas, however, may be due to the browning reaction, as has been reported in baked wheat products (Ranhotra and Gelroth, 1988).

Ash. Changes in ash content have not received much attention from researchers. Most findings, however, have shown an increase in total ash content from maize to tortillas, which may be expected because of the lime used for cooking. Along with this increase in ash there is a significant increase in calcium content. According to Pflugfelder, Rooney and Waniska (1988b), calcium content in the dough is influenced by lime levels, cooking and steeping temperatures and maize characteristics. The changes in other minerals are variable and may depend on the purity of the lime used as well as on the type of grinding equipment. In one study (Bressani, Breuner and Ortiz, 1989; Bressani *et al.*, 1990) the magnesium content increased from 8 to 35 percent from maize to tortilla; there was no change in sodium and a small decrease in potassium. Iron content also increased; however, the increases may have resulted from contamination. Phosphorus content also increases from maize to tortilla (Table 19). One aspect of nutritional interest is that the calcium-to-phosphorus ratio, which is about 1:20 in maize, changes to approximately 1:1 in the tortilla.

TABLE 19

Mineral content of raw maize and home and industrial samples of tortillas (mg/100 g)

Product	P	K	Ca	Mg	Na	Fe	Cu	Mn	Zn
Maize	300	325	48	108	54	4.8	1.3	1.0	4.6
Home-made tortilla 1	309	273	217	123	71	7.0	2.0	1.0	5.4
Home-made tortilla 2	–	–	202	–	–	2.7	0.3	–	3.4
Home-made tortilla 3	294	–	104	72	–	3.5	1.3	–	4.6
Industrial tortilla 1	315	–	182	106	–	4.0	2.5	–	3.2
Industrial tortilla 2	240	142	198	60	2	1.2	0.17	0.41	1.2
Industrial tortilla 3	269	185	205	63	9	1.5	0.19	0.40	1.1

Sources: Bressani *et al.*, 1990; Krause, 1988; Ranhotra, 1985; Vargas, Muñoz and Gomez, 1986

Carbohydrates. Maize and tortillas contain significant amounts of soluble carbohydrates, but very little is known on how they change during alkaline processing. Starch losses of about 5 percent have been reported; these are recovered in the solids lost. A decrease in sugar from 2.4 percent in maize to 0.34 percent in tortillas was also found. Robles, Murray and Paredes-Lopez (1988) found that alkali-cooking and soaking of maize caused large increases in viscosity and that cooking time had a significant effect on pasting properties, although there was no extensive gelatinization of the starch. Differential scanning calorimetric studies yielded similar gelatinization endotherms for untreated maize and *nixtamal* flours. In the process enzyme-susceptible starch increases as cooking time lengthens.

Protein and amino acids. Most researchers report a small increase in N content which is attributed to a concentration effect. The solubility of all protein fractions is decreased from raw maize to tortillas, with an increase in the insoluble fraction.

Bressani and Scrimshaw (1958) extracted the nitrogen from raw maize and tortillas using water, sodium chloride, 70 percent alcohol and sodium hydroxide. The solubility of the water, salt and alcohol protein fractions was significantly lower in tortillas, with the alcohol-soluble proteins affected most. Only a small decrease of about 13 percent in the solubility of the alkali-soluble fraction was detected. Because of this, the insoluble nitrogen fraction increased from 9.4 percent in maize to 61.7 percent in tortillas.

Ortega, Villegas and Vasal (1986) observed similar changes in both common and QPM maize using the Landry-Moureaux (1970) protein fractionation technique. The solubility of true zeins decreased 58 percent in the tortillas prepared from common maize and 52 percent in QPM tortillas. The authors indicated that hydrophobic interactions may have been involved in the change in protein solubility observed. Sproule *et al.* (1988) noted a decrease in the albumin plus globulin-nitrogen, expressed as percentage of total nitrogen, from maize to tortillas.

The changes in amino acid content from maize to tortillas are summarized in Table 20. *In vitro* enzymatic studies of amino acids indicated that total nitrogen and alpha-amino nitrogen were released faster from maize than from tortillas. Values for alpha-amino nitrogen released, expressed as a percentage of the total nitrogen release, were higher for tortillas than for raw maize after 12 hours of hydrolysis with pepsin. The percentage of alpha-amino N from the total was similar for maize and tortillas at 60 hours of hydrolysis with trypsin and pancreatin. After 60 hours of hydrolysis with pepsin, trypsin and pancreatin, the percentage of enzymatically released amino acids with respect to the acid-hydrolysed amino acids suggested a faster release from tortillas than from maize. This information was recorded up to 36 hours for most of the amino acids except leucine, phenylalanine, tryptophan and valine, which were released at about the same rate. At 60 hours of hydrolysis the amino acid concentrations of the maize and tortilla hydrolysates reached comparable levels, except for methionine (Bressani and Scrimshaw, 1958). These authors reported losses of arginine (18.7 percent), histidine (11.7 percent), lysine (5.3 percent), leucine (21 percent), cystine (12.5 percent) and small amounts of glutamic acid, proline and serine.

Sanderson *et al.* (1978) found small losses of arginine and cystine from alkaline treatment of common and high-lysine maize. These same authors

TABLE 20

Amino acid changes during the alkaline cooking of maize (g/16 g N)

Amino acid	Maize	Tortilla	Maize	Dough	Tortilla	QPM	Dough
Arginine	5.1	4.2	5.4	4.6	5.5	8.3	7.9
Histidine	2.7	2.4	2.9	2.8	3.5	3.9	3.8
Isoleucine	4.2	4.5	3.7	3.8	3.5	3.4	3.3
Leucine	12.2	9.6	12.6	13.4	12.1	8.3	8.3
Lysine	3.0	2.9	3.0	2.7	2.9	5.1	5.2
Methionine	1.9	1.9	2.8	2.9	2.3	1.9	1.9
Cystine	1.0	0.9	–	–	–	–	–
Cysteine	–	–	2.0	1.7	1.9	2.5	2.2
Phenylalanine	3.7	3.8	5.0	5.2	4.7	4.3	4.2
Tyrosine	3.8	3.8	4.5	4.6	4.4	3.8	3.7
Threonine	3.0	3.0	3.8	3.8	3.4	3.6	3.6
Tryptophan	0.5	0.5	–	–	–	–	–
Valine	4.5	4.8	4.8	5.3	4.9	5.1	5.0
Glutamic acid	20.3	19.0	18.8	19.5	18.9	15.4	15.7
Aspartic acid	6.2	6.2	7.2	6.9	5.8	8.4	8.4
Glycine	4.8	4.8	4.0	4.3	3.5	4.7	4.6
Alanine	8.8	8.8	7.7	8.1	7.6	6.1	6.1
Serine	4.5	4.2	5.0	5.0	4.7	4.4	4.5
Proline	11.0	10.1	9.2	10.7	8.7	7.0	7.6

Sources: Bressani and Scrimshaw, 1958; Sanderson *et al.*, 1978

found 0.059 and 0.049 g of lysino-alanine per 100 g protein from common and high-lysine maize respectively, but none was found in raw maize. In commercial *masa*, they found 0.020 g lysino-alanine per 100 g protein, while in tortillas the level found was 0.081 g per 100 g protein.

Lunven (1968), using his own amino acid column chromatography technique, observed significant losses in both lysine and tryptophan during populations who do not consume diets high in this essential mineral. Furthermore, the finding that better quality in maize protein favours calcium bio-utilization is of nutritional significance and provides an additional reason for the commercial production of QPM for people who depend on maize for their nutrition.

Amino acids. Bressani and Scrimshaw (1958) carried out studies using *in vitro* enzymatic digestions with pepsin, trypsin and pancreatin. At the end of the pepsin digestion, the amount of alpha-amino nitrogen as a percentage of total digested nitrogen was twice as high from tortilla (43.1 percent) as from maize (21.4 percent) and levels of histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine and tryptophan were higher from the tortilla hydrolysate than from maize, suggesting a faster release from the proteins. These authors proposed that the difference in rate of release could derive from the significant decrease in the solubility of the prolamine protein fraction in tortillas, as compared with maize. Serna-Saldivar *et al.* (1987), however, working with ileum-cannulated pigs, found that at this level in the intestinal tract the digestibility of most of the essential amino acids was somewhat higher from water-cooked maize than from lime-cooked maize. Digestibility of the protein decreased slightly, possibly because of the heat treatment involved (Bressani *et al.*, 1990). Other researchers have suggested that during maize processing, hydrophobic interactions, protein denaturation and cross-linking of proteins are probably responsible for changes in the solubility of these components, which could affect amino acid release during enzymatic digestion.

Niacin. The alkaline treatment of maize has been reported to destroy its pellagragenic factor. Evidence from a large number of researchers has suggested that pellagra results from an imbalance of the essential amino acids, increasing the niacin requirement of the animal. This point has been extensively debated between those who claim that niacin in maize is bound and not available to the animal and those who favour the theory of improved amino acid balance induced by the alkaline-cooking process, as lime treatment results in release of the bound niacin. Pearson *et al.* (1957) have

shown that boiling maize in water has the same effect (that is, it increases niacin availability). Bressani, Gómez-Brenes and Scrimshaw (1961) found that *in vitro* enzymatic digestion liberated all the niacin from raw maize as from tortillas and reached the conclusion that differences in amino acid balance rather than in bound niacin were responsible for the differences between raw and lime-processed maize in biological activity and pellagragenic action. Lime treatment of maize improves amino acid balance, as demonstrated by Cravioto *et al.* (1952) and Bressani and Scrimshaw (1958). Other workers have shown that experimental animals grow better when fed lime-treated rather than raw maize. Using cats – which cannot convert tryptophan into niacin – Braham, Villareal and Bressani (1962) showed that niacin from raw and lime-treated maize was utilized to an equal extent, suggesting its availability is not affected by processing.

Dietary Fibre. It has been shown above that when maize was processed into tortillas by lime-cooking, total dietary fibre (TDF) decreased at the dough stage and increased in the tortilla to levels only slightly below those found in raw maize. In these studies the levels of TDF in tortillas averaged 10 percent on a dry weight basis. If a person consumed about 400 g of tortilla (dry-weight), the TDF intake would be 40 g, a value significantly higher than the recommended intake. Even small children can consume relatively large amounts of dietary fibre, which can affect the availability of iron. Hazell and Johnson (1989), however, indicated that maize-based snack foods prepared by extrusion cooking have a higher iron availability than raw maize. These authors indicated that refining of raw maize, product formulation, extrusion cooking and addition of flavourings were responsible to different degrees. Likewise zinc intake could be affected. The other mineral that could be affected would be calcium; however, Braham and Bressani (1966) and Poneros and Erdman (1988) showed that calcium is relatively well available from tortillas and that its availability is increased when the protein quality is improved through addition of the limiting amino acids. An excess of calcium rather than dietary fibre could be responsible for zinc availability, as has been indicated in a number of studies.

Protein quality of maize and nutrient bioavailability

Growing rats retained calcium from tortillas better when it was supplemented with lysine, its limiting amino acid, and with a mixture of amino acids. Protein quality is an important factor in bioavailability of nutrients from maize and its lime-treated products. As already stated, niacin availability also improves when protein quality is improved, and studies with QPM have shown better utilization of niacin. The same observation has been made on the utilization of carotene, which is higher in lysine-supplemented yellow maize than in the unsupplemented product.

Changes in quality. Changes in nutritional value, particularly that of protein, during the transition from raw maize to tortillas have been studied mainly in animals. Even though chemical losses in some nutrients take place upon lime-cooking of maize, protein quality is slightly but consistently better in tortillas than in raw maize. Table 22 summarizes the results of various studies where raw maize and the tortillas made from it have been evaluated. The protein efficiency ratio of the tortillas is in general somewhat higher than that of the raw maize, although some studies have reported otherwise. The difference may be attributed to processing conditions, particularly the concentration of lime added, which is lower in rural home cooking than at the industrial level. The chemically determined amino acid pattern of the tortillas is no better than the pattern in raw maize. The only explanation is that the process increases the availability of key amino acids. This is indicated by the results of feeding studies with young rats (Bressani, Elías and Braham, 1968). Both raw maize and the lime-cooked dough were supplemented with increasing levels of lysine alone (from 0 to 0.47 percent of the diet). Maximum PER for maize was obtained with an addition of 0.31 percent and for the lime-cooked dough with 0.16 percent. At all levels of supplemental lysine the dough gave higher PER values than the raw maize.

Tryptophan supplementation alone was also tested, and in this case 0.025 percent addition gave the highest PER for maize, with no response for the dough. The addition of the two amino acids at the level of 0.41 percent lysine with tryptophan varying from 0.05 to 0.15 percent improved the quality of both materials, although it was higher for the dough.

TABLE 22

Protein quality of maize and tortillas

Type of maize	Protein quality (PER)		
	Raw maize	Tortillas	Casein
Common	1.13 ± 0.26	1.27 ± 0.27	
Common	1.49 ± 0.23	1.55 ± 0.23	2.88 ± 0.20
QPM (Opaque-2)	2.79 ± 0.24	2.66 ± 0.14	2.88 ± 0.20
Common	1.38	1.13	2.50
Common Tropical	0.99 ± 0.25	1.41 ± 0.11	2.63 ± 0.17
Common Highland Xetzoc	0.96 ± 0.19	1.41 ± 0.20	2.63 ± 0.17
Common Highland Azotea	1.02 ± 0.19	1.41 ± 0.17	2.63 ± 0.17
Common Highland Sta. Apolonia	0.71 ± 0.20	0.98 ± 0.17	2.63 ± 0.17
QPM Nutricta	1.91 ± 0.23	2.12 ± 0.12	2.63 ± 0.17
Biological value of common maize	59.5	59.1	69.4
Net protein utilization of common maize	51.2	49.4	64.5

These results were interpreted to mean that the quality of lime-treated maize is superior to that of raw maize. This explanation is supported by *in vitro* studies showing a greater release of essential amino acids (EAA) from tortillas than from maize, even though Ortega, Villegas and Vasal (1986) reported *in vitro* protein digestibility in maize, dough and tortillas to be 88, 91 and 79 percent respectively. For QPM the respective values were 82, 80 and 68 percent. Recently, Serna-Saldivar *et al.* (1987) reported on dry matter, gross energy and nitrogen digestibilities of maize cooked with and without lime. No differences in dry matter or gross energy digestibility values were found between the different processing treatments. Cooking maize with lime, however, reduced nitrogen digestibility from 76.5 to 72.8

percent. These values were measured near the end of the small intestine in pigs. Values for dry matter, gross energy and nitrogen digestibility increased when measured over the pigs' total digestive tract. From nitrogen balance studies, the same authors reported a retention of intake nitrogen of 45.8 percent for maize cooked without lime and 41.2 percent for lime-cooked maize. Retention of absorbed nitrogen was 48.2 percent for the lime-cooked maize and 52.9 percent for the maize cooked with water alone. Digestible and metabolizable energy were similar in maize processed with and without lime. The authors concluded that the lime-cooking process decreased the nutritive value of maize.

In another study by Serna-Saldivar *et al.* (1988b), this time conducted with rats, the authors noted an increase in percentage of dry matter and gross energy digestibilities from maize to *nixtamal* (dough) and to tortillas; however, protein digestibility decreased. *In vitro* studies correlated with *in vivo* values. Braham, Bressani and Guzmán (1966) showed better weight gain in Duroc-Jersey pigs fed lime-treated maize than in those fed raw maize, with better feed efficiency. In studies with dogs, lysine and tryptophan added to lime-cooked maize improved nitrogen balance to the value obtained with skim milk (Bressani and de Villareal, 1963; Bressani and Marenco, 1963). It was further shown that after these two amino acids, isoleucine, threonine, methionine and valine increased nitrogen retention above values measured with lysine and tryptophan. Lime-treated maize has also been evaluated in children (see Chapter 6). Nitrogen balance results have shown a high response to lysine and tryptophan addition, which in turn is dependent on the level of protein intake. At low levels, only lysine improved quality, but as nitrogen intake increased, the addition of tryptophan with lysine became important. All studies suggest that in lime-treated maize, lysine is slightly more deficient than tryptophan, and the contrary seems to be the case for raw maize. Nevertheless, for a significant improvement in protein nutritional quality of lime-treated maize, both of these amino acids are required.

Use of QPM. Nutritionally improved (QPM) maize shows the same changes in protein quality and bioavailability after lime-cooking and conversion to

tortillas as observed in normal maize. The difference is that QPM tortillas and products are nutritionally superior to those made from common maize. They are as acceptable to consumers.

Other effects of lime-cooking

Lysinoalanine formation. In 1969, De Groot and Slump demonstrated that alkali treatment of proteins gave rise to peptides such as lysinoalanine (LAL), lanthionine and ornithine which had negative effects on animals. They were not biologically available and had detrimental effects on protein quality. Consequently, the effect of the alkaline-cooking process to convert maize into tortillas has received attention from various researchers. Sternberg, Kim and Schwende (1975) reported that commercial samples of *masa* flour, tortillas and taco shells contained 480, 200 and 170 µg LAL per gram. Sanderson *et al.* (1978) also found that lanthionine and ornithine were formed during alkaline cooking of maize. These authors found no LAL in common or in high-lysine raw maize; however, these products contained 0.059 and 0.049 g percent protein respectively after alkali treatment. A commercial *masa* contained 0.020 percent, and tortillas 0.081 percent protein. These authors also reported lanthionine and ornithine values in the *masa* prepared from the two types of maize. Chu, Pellet and Nawar (1976) reported values of 133.2 µg of LAL per gram protein when maize was processed with 4.1 mol per kg of lime for 30 minutes at 170°F (76.6°C). The use of sodium hydroxide under equal conditions yielded higher levels of LAL. Since higher levels of LAL were obtained with NaOH and KOH, the authors suggested that calcium ions may in some way interfere with the mechanism of LAL formation. It is difficult to evaluate the significance of LAL formation during tortilla-making for people who eat relatively large amounts of this food daily. Since this has been practised for a long time, the small amounts may not interfere with nutritive value or cause any pathological effects. Studies on the effect of lime level on the protein quality of maize have shown, however, that levels above 0.5 percent of grain weight reduce protein quality. The type of maize used and its size are of importance in this respect. Softer types of grains are more affected than hard grains cooked under similar conditions (Bressani *et al.*, unpublished data).

Mycotoxins and alkaline-cooking of maize. The presence of mycotoxins in a variety of cereal grains and other foods and feedstuffs is today widely recognized, and maize is no exception. In Central America, where maize is such an important food, the grain is harvested twice a year in the tropical areas. One harvest is in August, when rain and temperature conditions are ideal for the growth of fungi. Martínez *et al.* (1970b) reported the presence of six different fungi in maize samples obtained from different markets throughout Guatemala. The frequency of *Aspergillus versicolor* was 57.1 percent; of *Aspergillus wentii*, 32.1 percent; of *Aspergillus ruber*, 26.8 percent; of *Aspergillus echinulatus*, 25.0 percent; of *Aspergillus flavus*, 25.0 percent; and of *Chaedosporium* spp., 26.8 percent.

Because of the significance of the presence of mycotoxins in cereal grains, a number of studies have been conducted to assess the degree of retention of mycotoxins during grain processing. The effect of calcium hydroxide cooking of maize has received some attention. Martínez-Herrera (1968) fed infected maize, raw and alkali-processed, to chickens and rats. The maize was infected with *Fusarium* sp., *Penicillium* spp., *Aspergillus niger* and *A. flavus*. The author found high mortality among birds fed on the raw infected maize, but none in the group of chickens fed the same maize processed with calcium hydroxide. In young rats, the raw and infected grain reduced weight gain and caused some mortality. Infected grain processed with lime induced no mortality, however, and weight gain as well as feed efficiency were like those in the control. Adult rats were also affected by the infected maize, but not by infected maize processed with lime. The study did not report levels of mycotoxins before and after processing.

Martínez (1979) reported on studies of tortilla samples collected in Mexico City in different seasons. He found that 15 to 20 percent of the samples collected in spring 1978 and in the rainy season of 1977-1978 contained aflatoxins. Furthermore, he found that concentrations of aflatoxins B1 varied from 50 to 200 ppb. He also indicated that lime-cooking of maize reduced aflatoxin concentrations by 50 to 75 percent. Martínez and also de Campos, Crespo-Santos and Olszyna-Marzys (1980) reported that lime concentrations of up to 10 percent were no more effective in reducing aflatoxins than a 2 percent concentration.

Ulloa-Sosa and Schroeder (1969) reported that the tortilla-making process was not effective in removing aflatoxins from contaminated maize. Nevertheless, others have obtained different results. For example, Solorzano-Mendizabal (1985) found that maize inoculated with *A. flavus* and *Aspergillus parasiticus* produced high levels of aflatoxins which were reduced by lime-cooking, completely in some cases, but most often by up to 80 percent. Lime concentration varied from 0.6 to 8 percent, and analyses were done on maize, *masa*, tortillas and cooking waters. In another study, de Arriola *et al.* (1987, 1988), using QPM Nutricta, found that the lime levels at which *nixtamal* is normally prepared in Guatemala do not reduce aflatoxin in contaminated grain sufficiently to make it safe for human consumption.

Lime levels of 2 percent and above gave high aflatoxin reduction, but the tortillas were not acceptable. Aflatoxin B1 was reported to be reduced the most. Torreblanca, Bourges and Morales (1987) found relatively high aflatoxin levels in both maize and tortillas in a study conducted in Mexico City. Aflatoxin B1 was found in 72 percent of the maize tortilla samples tested; furthermore, 24 percent of the samples gave positive reactions for zearalenone. Carvajal *et al.* (1987) found mycotoxins in maize and tortillas in Mexican samples and indicated that aflatoxins, zearalenone and deoxynivalenol (DON) were not destroyed by the lime treatment or by temperatures of 110°C.

Price and Jorgensen (1985) found that the alkaline cooking process reduced aflatoxin levels from 127 µg per kg in raw maize to 68.6 µg per kg in tortillas. The authors concluded that the process was poorly effective, since the lower value obtained was still much above the value established as acceptable (about 20 mg per kg). These authors found that acidification – as it occurs in the intestinal tract – increased aflatoxin levels. Abbas *et al.* (1988) reported on the effect of 2 percent lime-cooking of maize on the decomposition of zearalenone and DON. They found significant reductions, i.e. 58 to 100 percent for zearalenone and 72 to 82 percent for DON. Furthermore, 15-acetyl-DON was completely destroyed.

Results obtained by various authors are somewhat conflicting, since some of them report partial reduction in some mycotoxins while others note total reduction. In many studies the mycotoxin levels were relatively high, necessitating stronger processing conditions in terms of lime concentration and cooking time. The problem warrants further study. Grain quality is probably the best means of ensuring the absence of mycotoxins rather than dependence on lime to reduce them partly or eliminate them in the final product.

Microbiological aspects of tortillas and tortilla flour. Studies on the microflora in lime-cooked maize tortillas are very limited. Capparelli and Mata (1975) showed that the main contaminants of tortillas as made in the highlands of Guatemala were coliforms, *Bacillus cereus,* two *Staphylococcus* species and many types of yeasts. When tortillas are first cooked, bacterial counts are about 10^3 or fewer organisms per gram, which is a safe level for consumption. After they are cooked for about five minutes on a hotplate they are placed hot in a basket, often covered with a cloth. This captures the vapour from the tortillas, creating an environment appropriate for microbial growth. After some ten hours under these conditions the surfaces of stacked tortillas become slimy and they are not acceptable for consumption.

Although there are many opportunities in rural areas for contamination during processing from maize to tortillas, the factors that possibly contribute the most are the water used during conversion of cooked maize to dough and the mill used to grind the cooked maize. Molina, Baten and Bressani (1978) reported a greater increase in bacteria counts in tortillas fortified with soybean flour and vitamins than in unfortified tortillas. In this case the mill used to grind the cooked maize to make the dough was chlorinated, which helped in lowering the bacteria count in the soy-supplemented maize. The tortillas made from it also had a lower bacteria count. The rate of increase in bacterial number decreased as well. Higher bacteria counts were reported by Valverde *et al.* (1983) in the dough and tortillas made from QPM Nutricta than in those from common maize, showing the effect of nutritional quality on bacterial growth.

The relatively high moisture content which is responsible for a very short shelf-life has limited marketing of tortillas. Nevertheless, there is a demand

for them in urban areas, where they are marketed under refrigerated conditions. A number of attempts have been made to lengthen their shelf-life. Rubio (1972a, 1972b, 1973, 1974a, 1974b, 1975) patented a number of methods which included various additives: epichlorohydrin and polycarboxylic acid and their anhydrides; hydrophilic inorganic gels; sorbic acid and its salts as well as the methyl, ethyl, butyl and propyl esters of *para*-hydroxy benzoic acid; and acetic and propionic acids. Pelaez and Karel (1980) developed an intermediate-moisture tortilla with a stable shelf-life. It was free from microbial growth, including *Staphylococcus aureus*, yeasts, moulds and enterotoxin. This was achieved through the use of glycerol, corn solids DE-42 and salt, as well as the mycostatic agent potassium sorbate. Protection with appropriate packaging was claimed for at least 30 days and the appearance, texture and other characteristics were similar to those of regular tortillas with a water activity of 0.97. Hickey, Stephens and Flowers (1982) reported relatively good protection of tortillas with low levels of sorbates or propionates added to the dough, and with a spray of sorbate on the surface (both sides) after cooking on the hot plate. More recently, Islam, Lirio and Delvalle (1984) claimed that using calcium propionate extended the shelf-life of tortillas at room temperature to 2 to 5 days; with dimethyl fumarate shelf-life was 2 to 11 days under the same storage conditions and using polythene bags. Although advances have been made in extending shelf-life, it still constitutes a problem for people who buy food in supermarkets.

Reports on the microbiology of tortilla flour and the tortillas made from it are not available. Lower total bacteria counts would be expected, however, because of the process employed to prepare the flour and use it at home.

OGI AND OTHER FERMENTED MAIZE PRODUCTS

Chemical changes

The process of fermenting maize, sorghum, millet or rice to produce *ogi* not only removes parts of the maize kernel such as the seed-coat and the germ, but also involves washing, sieving and decanting, all of which induce changes in the chemical composition and nutritive value of the final product. Akinrele (1970) reported on specific nutrients of a number of *ogi* samples produced in different ways: unfermented and fermented with *Aerobacter cloacae*, *Lactobacillus plantarum* and a mixture of the two bacteria. He compared the values found with those from the traditionally fermented product. Judging from the ratio of amino nitrogen to total nitrogen, the author reported that protein was degraded to a very small amount by any bacterial species. When compared with the unfermented *ogi*, *A. cloacae* appeared to synthesize more riboflavin and niacin, which did not take place with *L. plantarum*. Traditionally produced *ogi* had more thiamine and slightly lower values of riboflavin and niacin than that made with maize and *A. cloacae*. In any case the changes were small, and smaller if compared with whole maize, whereas in comparison with degermed maize, the *ogi* products contained more riboflavin and niacin. Akinrele (1970) and Banigo and Muller (1972) reported on the carboxylic acids in *ogi* and found lactic acid in greatest concentration (0.55 percent) followed by acetic acid (0.09 percent) and smaller amounts of butyric acid. The latter investigators suggested levels of 0.65 percent for lactic acid and 0.11 percent for acetic acid, responsible for the sour taste, as goals for flavour evaluations. Banigo, de Man and Duitschaever (1974) reported on the proximate composition of *ogi* made from common whole maize which was uncooked and freeze-dried or cooked and freeze-dried after fermentation. Changes were relatively small in all major nutrients, with a slight increase in fibre and a decrease in ash content when compared with whole maize.

These authors also reported on amino acid content; they found no differences between maize flour and *ogi* for all amino acids including the essential ones. The *ogi* samples, however, had about twice the amount of serine and somewhat higher values for glutamic acid. Adeniji and Potter (1978) reported that *ogi* processing did not decrease the protein content of maize, but total and available lysine were significantly reduced. On the other hand, tryptophan levels were more stable and in two samples increased, probably because of fermentation. These authors also found an increase in neutral detergent fibre and ash but no change in lignin. Akingbala *et al.* (1987) found a decrease in protein, ether extract, ash and crude fibre in *ogi* as compared with maize that was processed as a whole grain or dry milled.

Nutritional changes

Nutritional evaluations of *ogi* and other maize-fermented products are not readily available. Adeniji and Potter (1978) found a substantial decrease in protein quality of drum-dried common maize *ogi*, which they ascribed to the drying process. These same authors reported significant losses in lysine. Several authors have more recently tested maize and sorghum and reported that fermentation improved the nutritional quality of the product. Akinrele and Bassir (1967) found net protein utilization, protein efficiency ratio and biological value of *ogi* inferior to those values in whole maize, even though some increase in thiamine and niacin was obtained. It has been indicated that some of the micro-organisms responsible for *ogi* fermentation, such as *Enterobacter cloacae* and *L. plantarum*, use some of the amino acids for growth. This together with the elimination of the germ from kernels explains the very low protein quality of *ogi* and similarly produced maize products. However, there are some exceptions, such as *kenkey* and *pozol*, both products in which the maize is fermented with the germ. Although protein quality values are not available for *kenkey*, Cravioto *et al.* (1955) found higher levels of tryptophan and available lysine, which suggested higher protein quality than in raw maize or lime-treated maize. More recently, Bressani (unpublished) found the fermented product to be higher in protein quality than raw maize, but not different in quality from lime-cooked dough.

Use of QPM

Adeniji and Potter (1978) used high quality protein maize to make *ogi* and found similar results to those from common maize, except that the protein quality was higher (although lower than that of the original raw maize). *Pozol* made from QPM has significantly higher protein quality than raw QPM (Bressani, unpublished data).

AREPAS

Chemical changes

Arepa flour is made in a dry milling process which removes the pericarp and the germ from maize. Therefore, *arepa* flour may be expected to differ from whole maize flour, and this was in fact reported by Cuevas *et al.* (1985). The protein, ether extract, fibre and ash content of *arepa* flour from both white and yellow maize were lower than in whole maize. The same is true for thiamine, riboflavin and niacin as well as for calcium, phosphorus and iron. These changes evidently result from the removal of the germ and seed-coat.

Nutritive value

Arepa flour has been subjected to biological assay for protein quality by Chavez (1972a). He reported a decrease of about 50 percent in protein quality from maize (0.74) to *arepa* (0.33), although there was some increase in protein digestibility.

Use of QPM

High protein quality maize has been used to make *arepas*. Chavez (1972b) found the process to reduce nitrogen, lysine and tryptophan, thiamine and niacin and attributed this to germ removal. Protein quality was also significantly less than in whole maize, but was nonetheless superior to that of maize and *arepas* from normal maize. All products – *tortillas*, *ogi*, *pozol*, *kenkey* and *arepas* – made from QPM are of better protein quality and energy value than the products made from common maize.

OTHER DRY MILLING PRODUCTS

Chemical changes

The main maize products for food use derived from dry milling include flaking grits, coarse or fine grits, maize cones and maize flour. They are products from which the pericarp and germ have been eliminated and they differ from each other in granulation, with flaking grits having the largest particle size and flour the smallest. Basically, their chemical compositions based on food composition data are very similar.

Nutritive value

The protein quality of these products, as with most dry-milled maize products, is inferior to that of the original whole grain. If there are any changes, these come about from the processes used to turn such products into the different forms in which they are consumed. For example, the

protein digestibility of maize meal was reported by Wolzak, Bressani and Gómez-Brenes (1981) to be 86.5 percent and that of corn flakes 72.0 percent. A significant diminution of protein quality also takes place since available lysine decreases.

QPM products

Studies on dry milling of QPM, particularly the hard-endosperm types, are not readily available. Wichser (1966) found yields of 8.8 percent grits from milled QPM, while the yield of grits from maize hybrids was about 17 percent. The yields of meal and flour were essentially the same from QPM and hybrid maize. However, the fat, protein, fibre and ash contents in QPM grits, breakfast cereal and flour were higher than those in similar products from hybrid maize.

Not much information on the nutritional value of QPM dry-milled products is available; however, Wichser (1966) showed the endosperm of QPM to have a net protein ratio (NPR) of 76 percent of the value of casein (100 percent), while the endosperm from hybrid maize had an NPR of 47 percent of the value of casein. These results are very similar to those for maize flour made for *arepa* production from QPM and common maize as shown by Chavez (1972a).

Chapter 6

Comparison of nutritive value of common maize and quality protein maize

CONSUMPTION OF MAIZE

Maize in its different processed forms is an important food for large numbers of people in the developing world, providing significant amounts of nutrients, in particular calories and protein. Its nutritional quality is particularly important for small children. Table 23 shows the consumption of maize as tortillas or lime-treated maize by children in Guatemala. Amounts varied from 64 to 120 g per day, providing about 30 percent of the daily protein intake and close to 40 percent of the daily energy intake. García and Urrutia (1978) reported an intake of 226 g of tortillas by weaned three-year-old children, providing about 47 percent of their calories.

Although these findings are not basically bad, adequate supplementary foods are often not provided or are given only in insignificant amounts. Food legumes are the most readily available supplementary food in developing countries; however, the amounts are generally very small (Flores, Bressani and Elías, 1973). The average intake of beans per age group for the six countries in Central America was 7, 12, 21 and 27 g per child per day at 1, 2, 3 and 4/5 years, respectively. On the basis of 22 percent crude protein in beans, the amounts of protein provided by this food were 1.5, 2.6, 4.6 and 5.9 g, respectively; however, amounts of digestible protein on the basis of a true digestibility of 70 percent were only 1.0, 1.8, 3.2 and 4.1 g. Thus beans provided about 14, 18, 22 and 30 percent of the dietary protein in the total intake from maize and beans. These amounts and their supplementary effects were very small, particularly for the one- and two-year-old children.

TABLE 23
Maize consumption and its contribution to daily calorie and protein intake of children in a rural area of Guatemala

Age (*years*)	Maize intake (*g/day*)	Protein intake			Calorie intake		
		Maize (*g/day*)	Total (*g/day*)	Percent of total from maize	Maize (*cal/day*)	Total (*cal/day*)	Percent of total from maize
1-2	64	5.4	20.0	27	231	699	33
2-3	86	7.3	21.7	34	310	787	39
3-4	120	10.2	27.9	36	433	981	44
4-5	89	7.6	23.3	33	321	819	39

Source: M. Flores (cited in Bressani, 1972)

Data for 1979-1981 from FAO (1984) showed that 22 of 145 countries had a maize consumption of more than 100 g per person per day as indicated in Table 24, which also gives the calories and protein that maize provides. It should be pointed out, however, that 1960-1962 figures from FAO food balance sheets (FAO, 1966) were higher for some countries than the 1979-1981 figures. The figures confirm the importance of maize as a staple food in some Latin American countries, particularly Mexico and Central America, as well as in some African countries. It follows that if the maize intake is high, maize contributes significant amounts of calories and protein to the daily intake of people in these countries.

Table 25 summarizes maize intake, calories per day and protein per day among the rural and urban populations of the six countries of Central America. Two general trends are evident. The first is that maize intake decreases from north to south. The cereal grain that replaces it is rice. The second trend is that intake of maize is higher in rural than in urban areas. In at least three countries maize makes up the greatest proportion of all the ingested food in the rural sector and is therefore an important source of nutrients in the diet. The table shows that maize provides up to 45 and 59 percent of the daily intake of calories and protein respectively.

TABLE 24
Maize intake and its calorie and protein contribution to the daily diet

Country	Intake (*g/person/day*)	Calories (*per person/day*)	Protein (*g/person/day*)
Benin	160.5	481	12.7
Botswana	209.3	665	17.5
Cape Verde	334.1	1 052	28.0
Egypt	149.7	508	13.4
El Salvador*	245.0	871	23.3
Guatemala	276.2	977	15.4
Honduras	255.9	878	22.8
Kenya	286.1	808	21.3
Lesotho	315.4	1 002	26.4
Malawi	468.8	1 422	37.6
Mexico	328.9	1 061	27.1
Nepal	116.4	379	9.4
Nicaragua*	131.0	472	11.1
Paraguay	131.2	445	11.6
Philippines	152.1	399	8.7
Romania	128.6	373	8.6
Singapore	122.2	345	8.6
South Africa, Rep.	314.7	961	24.6
Swaziland	381.4	1 279	33.7
Tanzania, United Rep.	129.1	421	10.0
Togo	136.9	411	10.8
Venezuela	118.3	339	7.4
Zambia	418.6	1 226	31.3
Zimbabwe	330.9	958	25.2

Sources: FAO, 1984; *FAO, 1966

TABLE 25
Importance of maize consumption in rural areas

Country	Urban maize intake (*g/day*)	Rural maize intake (*g/day*)	Rural calorie intake (*per day*)		Rural protein intake (*g/day*)	
			From maize	Total	From maize	Total
Guatemala	102	318	1 148	1 994	27.0	60
El Salvador	166	352	1 271	2 146	29.9	68
Honduras	135	225	812	1 832	19.1	58
Nicaragua	56	131	472	1 986	11.1	64
Costa Rica	14	41	148	1 894	3.5	54
Panama	4	4	14	2 089	0.3	60

Source: INCAP, Guatemala, 1969

Although this information was compiled from dietary surveys conducted in 1969, figures have not changed significantly in recent years. For example, in 1976 average consumption in El Salvador varied from 146 to 321 g per person per day; in Honduras in 1983 consumption in different regions varied from 111 to 246 g per person per day; and in Costa Rica in 1986, intake varied from 14 to 31 g per person per day. Chavez (1973) indicated that about 45 percent of the national calorie intake is provided by maize in Mexico. In poor rural areas men may consume about 600 g of maize and women about 400 g. On this basis the importance of the nutritional quality of maize is obviously great. Although all nutrients are of interest, the quality of protein has received more attention from researchers.

COMMON MAIZE

Protein quality for children

The protein quality of maize evaluated for children recovering from protein-energy malnutrition has been reported by various researchers. Table 26 shows the results when lime-cooked maize was supplemented with maize gluten to obtain a product with higher protein content and to facilitate higher

TABLE 26
Nitrogen balance in children fed lime-treated maize as the sole protein source

Protein intake (*g/kg/day*)	Nitrogen (*mg/kg/day*)			% of intake	
	Intake	Absorbed	Retained	Absorbed	Retained
3	470 (435 to 479)	339 (327 to 369)	9 (-8 to 174)	72 (61 to 77)	2 (-2 to 36)
2	331 (308 to 367)	260 (207 to 284)	22 (-41 to 59)	78 (65 to 82)	7 (-13 to 17)
1.5	238 (235 to 241)	180 (168 to 193)	-11 (-22 to -2)	76 (70 to 82)	-4 (-9 to -1)

Note: Diet consisted of 95% lime-treated maize and 5% maize gluten
Source: Viteri, Martínez and Bressani, 1972

protein intakes with lower intakes of solids. The amino acid deficiencies in maize protein were thus magnified and this facilitated their detection using the nitrogen balance technique (Scrimshaw *et al.*, 1958; Bressani *et al.*, 1958, 1963). The results showed decreasing nitrogen retention as nitrogen intake decreased, which was to be expected; however, even at a high nitrogen intake of 469 mg per kg body weight per day, retention was significantly lower than nitrogen retention from milk given in amounts providing the same level of protein. Apparent protein digestibility indicated as nitrogen availability was fairly similar for different nitrogen intakes, varying from 72 to 78 percent. Table 27 refers to nitrogen balance studies in children fed water-cooked maize. Nitrogen retention from maize was significantly lower than from milk at the same level of protein intake. Protein digestibility was 80 percent for milk and 75 percent for maize (Viteri, Martínez and Bressani, 1972). Similar data were obtained with cooked maize endosperm and whole normal maize (Graham, Placko and MacLean, 1980), as shown in Table 28. In this case nitrogen balance was lower for common maize endosperm than for the whole kernel and lower than the results from the reference protein, casein. Graham *et al.* (1980) calculated that in order to match nitrogen retention from casein, the

TABLE 27

Nitrogen balance in children fed common maize and milk

Food	Intake (g/kg/day)	Protein intake (g/kg/day)	Nitrogen absorbed (mg/kg/day)	Nitrogen retained (mg/kg/day)	% N intake absorbed	% N intake retained
Milk	195 (175-210)	1.25	157 (114-181)	75 (40-106)	80 (61-47)	38 (22-50)
Common maize	192 (183-198)	1.25	144 (129-157)	30 (10-59)	75 (66-20)	16 (5-30)

Note: Average values, with dispersion in parentheses
Source: Viteri, Martínez and Bressani, 1972

TABLE 28

Nitrogen balance in children fed whole common maize kernels and maize endosperm flour

Food fed	Nitrogen absorbed (% of intake)	Nitrogen retained (% of intake)
Endosperm	64.1 ± 11.4	15.1 ± 8.9
Casein	81.8 ± 5.2	37.0 ± 14.2
Whole kernel	73.1 ± 1.9	26.8 ± 4.6
Casein	83.5 ± 2.5	39.6 ± 9.1

Source: Graham, Placko and MacLean, 1980

children would have to obtain 203.9 percent of their energy requirements from maize, which is obviously impossible.

As was discussed earlier, germ proteins do contribute significantly to essential amino acids (EAA), so maize food products without the germ, including QPM endosperm, are always lower in protein quality than the whole kernel. Similarly, maize with a high zein content is of a lower quality than maize with lower prolamine content, because of a higher relative lysine deficiency and a higher imbalance of essential amino acids such as leucine relative to isoleucine.

TABLE 29

Effects on nitrogen retention of additions of lysine, tryptophan and methionine to lime-treated maize (nitrogen values in mg/kg/day)

Diet	Nitrogen intake	Faecal nitrogen	Urinary nitrogen	Nitrogen absorbed	Nitrogen retained
Basal (B)	461	117	334	344	10
B + tryptophan	457	115	289	342	53
B + tryptophan + lysine	464	135	243	329	86
B + tryptophan + lysine + methionine	459	135	272	324	52

Note: Amino acids used: DL-tryptophan: 0.34%; L-lysine/HCl: 0.56%; DL-methionine: 0.34%
Source: Scrimshaw *et al*., 1958

Amino acid supplementation

It is widely accepted that maize proteins are deficient in both lysine and tryptophan, as documented from studies with animals. In tests with children, however, the EAA contents of lime-treated maize supplemented with 5 percent maize gluten to raise the protein content (Scrimshaw *et al*., 1958; Bressani *et al*., 1958, 1963) were compared with the amino acid contents of the 1957 FAO reference protein. This comparison suggested the following order of amino acid deficiency: tryptophan, lysine, methionine, valine, isoleucine and threonine. It also suggested the amounts of amino acids needed to reach the reference level. Representative results from two children fed 3 g of protein per kg body weight per day are shown in Table 29. There was an apparent response to the addition of 148 mg DL-tryptophan per g N which was much improved by the simultaneous addition of tryptophan and lysine, the latter in the amount of 243 mg per g N. Addition of methionine decreased nitrogen retention.

TABLE 30

Response to lysine and tryptophan added alone
(nitrogen values in mg/kg/day)

Diet	Nitrogen intake	Faecal nitrogen	Urinary nitrogen	Nitrogen absorbed	Nitrogen retained
Subject No. 1					
Milk	586	93	320	393	73
Basal (B)	474	185	349	289	-60
B + tryptophan	474	108	352	366	14
B	479	111	346	368	22
B + lysine	482	120	324	362	38
Subject No. 2					
Milk	392	45	295	347	52
Basal (B)	320	56	273	264	-9
B + lysine	335	54	257	285	24
B	346	63	287	283	-4
B + tryptophan	337	52	308	285	-23

Note: Levels added to give 75-90 mg tryptophan/g N and 180-270 mg L-lysine HCl/g N
Source: Bressani *et al.*, 1958

In other studies, nitrogen balance tests were carried out to learn about the response previously obtained by tryptophan addition alone. The results from two subjects (Table 30) clearly show that tryptophan had no effect on improving protein quality. The addition of lysine, on the contrary, appeared to give a response, suggesting lysine to be more limiting than tryptophan.

Similar studies were carried out by feeding children 2 g of protein per kg body weight per day. The results in two children are summarized in Table 31. Tryptophan addition did not induce a positive nitrogen retention, but the addition of tryptophan and lysine with and without isoleucine improved nitrogen balance. Methionine addition decreased retention of nitrogen, as previously demonstrated.

TABLE 31

Effects on nitrogen retention of additions of lysine, tryptophan, isoleucine and methionine to lime-treated maize
(nitrogen values in mg/kg/day)

Diet	Nitrogen intake	Faecal nitrogen	Urinary nitrogen	Nitrogen absorbed	Nitrogen retained
Basal (B)	320	68	270	252	-18
B + tryptophan	320	91	241	229	-12
B + tryptophan + lysine	321	105	201	216	15
B + tryptophan + lysine + isoleucine	321	90	207	231	24
B + tryptophan + lysine + isoleucine + methionine	314	84	217	230	13
B	319	98	242	221	-21

Note: Amino acid levels added: DL-isoleucine 0.45%; other amino acids added in amounts shown in Table 29
Source: Bressani *et al.*, 1958

Nitrogen balance tests were performed with protein intake of 1.5 g per kg body weight per day. The results for one child are shown in Table 32. Although lysine addition did not induce a positive balance, it did tend to cause a decrease in nitrogen losses. The improvement from lysine and tryptophan, with and without isoleucine, is evident. The addition of methionine, even at this level of protein intake, decreased the nitrogen balance as previously indicated for higher intakes of protein.

Because of the consistency of the results, the data obtained for different protein levels under the various dietary treatments were pooled. The results are shown in Table 33. There was a response to tryptophan addition only at the highest level of dietary protein, but the response to lysine was consistent at all protein intake levels, suggesting that this amino acid is more deficient than tryptophan. The response to addition of lysine alone, however, was small and without much nutritional significance, which implies the need to add both amino acids at the same time, as can be done with supplementary foods.

TABLE 32

Effects of amino acid supplementation of maize at intakes of 1.5 g protein per kg body weight per day (nitrogen values in mg/kg/day)

Treatment	Nitrogen intake	Faecal nitrogen	Urinary nitrogen	Nitrogen absorbed	Nitrogen retained
Basal (B)	241	71	187	170	-17
B + lysine[a]	239	59	184	180	-4
B + lysine[b]	239	48	193	191	-2
B + lysine + tryptophan	239	47	162	192	-30
B + lysine + tryptophan + isoleucine	240	44	150	196	46
N + lysine + tryptophan + isoleucine + methionine	240	55	162	185	23
B	235	45	193	190	-3

[a]0.56% L-lysine HCl
[b]0.30% L-lysine HCl
Other amino acids added in the amounts shown in Table 29
Source: Bressani *et al.*, 1958

A nitrogen intake level of 239 mg per kg body weight per day is equivalent to 20 g of maize per kg per day, or about the 200 g of maize normally ingested by children. Supplementation with lysine alone would have little effect. When tryptophan is also added, however, the nitrogen retention is significantly higher and even surpasses that of milk at the highest level of dietary protein. The overall conclusion that can be reached from the results obtained by amino acid supplementation of maize is that both lysine and tryptophan must be added to obtain a significant response in protein quality as measured by nitrogen retention. It also appears that the two amino acids are equally limiting in spite of the fact that the addition of lysine alone tended to improve protein quality slightly, while the results from the addition of tryptophan were inconsistent.

The effect of methionine deserves further comment. It was attributed to an amino acid imbalance, because maize already contains enough of this amino acid to meet nutritional requirements.

TABLE 33

Nitrogen balance in children fed lime-treated maize at various levels of protein intake with and without amino acid supplementation
(nitrogen levels in mg/kg/day)

Dietary treatment	2.9 g protein/kg/day		2.1 g protein/kg/day		1.5 g protein/kg/day	
	N intake	N balance	N intake	N balance	N intake	N balance
Basal diet (B)	469	14	326	-5	238	-10
B + tryptophan	465	33	327	-17	–	–
B + lysine	482	38	335	24	239	-4
B + lysine +tryptophan	461	83	328	36	239	30
B + tryptophan + lysine + isoleucine	475	108	335	40	240	46
Milk	458	70	364	73	–	–

Note: Amino acid levels used: DL-tryptophan, 0.34%; L-lysine/HCl, 0.56%; DL-isoleucine, 0.45%
Sources: Flynn *et al.*, 1954; Frey, 1951; Mitchell, Hamilton and Beadles, 1952

The results shown in Table 34 indicate that valine also decreases nitrogen retention and that its effect can be reversed by the addition of isoleucine and threonine. A more detailed study in dogs led to the conclusion that there is also a close interrelationship among all four of these amino acids – methionine, valine, isoleucine and threonine – as supplements to maize proteins (Bressani, 1962, 1963).

It is a point of major importance that children are so sensitive to small changes in amino acid proportions that they are readily detectable in a short period by testing the nitrogen balance. The data presented here emphasize the importance of establishing a proper balance among the essential amino acids if a maximum retention of nitrogen is to be obtained. This is the principle of amino acid supplementation. The results obtained on the amino acid supplementation of maize confirm data derived from studies with rats, pigs and other animals. Results of studies on adult human subjects are shown in the next section.

TABLE 34

Effect of multiple amino acid supplementation of maize (nitrogen values in mg/kg/day)

Diet	Nitrogen intake	Faecal nitrogen	Urinary nitrogen	Nitrogen absorbed	Nitrogen retained
Basal (B)	471	117	315	354	39
B + lysine + tryptophan + methionine	451	223	244	228	-16
B + lysine + tryptophan + methionine + valine	454	241	242	213	-29
B + lysine + tryptophan + methionine + valine + isoleucine	460	128	265	332	67
B + lysine + tryptophan + methionine + valine + isoleucine + threonine	447	190	218	257	39
B + lysine + tryptophan + methionine[a] + valine + isoleucine + threonine	450	129	238	321	83

[a]0.14% DL-methionine in this diet only; all others 0.34% DL-methionine. DL-valine: 0.90%; DL-threonine: 0.22%. Other amino acids added in amounts shown in Table 29

Source: Scrimshaw *et al.*, 1958

QUALITY PROTEIN MAIZE

Children

The high consumption of maize by the human population in a number of countries in Latin America and Africa and the well-established lysine and tryptophan deficiencies in maize protein motivated the search for a maize kernel with higher concentrations of these essential amino acids in its protein. The possibility of finding better varieties of maize appeared feasible on the basis of three facts. One was that by selection, oil content in the maize kernel could be increased from about 4 to 15 percent (Dudley and Lambert, 1969). This increase was obtained by increasing the size of the germ, the part of the kernel where the oil is concentrated. The same researchers showed that it was possible to increase total protein content from about 6 to 18 percent by increasing the prolamine (zein) fraction in maize endosperm. The third finding was the wide variability in lysine content reported among varieties and selections of maize.

The search for a high quality protein maize succeeded when Mertz, Bates and Nelson (1964) announced their discovery that the opaque-2 gene used as a marker in maize breeding significantly increased lysine and tryptophan in the cereal protein.

Results from initial alkaline processing studies of opaque-2 maize (cultivated in Indiana, United States, in 1965) showed that the process did not induce significant nutritional changes in the dough or in the tortillas, as concluded from chemical data and biological trials with rats.

The protein quality of alkali-processed opaque-2 maize was evaluated in children using the nitrogen balance index (the relationship between nitrogen absorption and retention). Six healthy children were used in two studies. The average nitrogen balances, at protein intake levels of 1.8 and 1.5 g per kg body weight per day, are presented in Table 35 (Bressani, Alvarado and Viteri, 1969). As can be observed, there were no significant differences in nitrogen retention among the children fed the diets based on milk and on

TABLE 35

Summary of the nitrogen balances of children fed whole milk and opaque-2 maize (nitrogen values in mg/kg/day)

Treatment	Nitrogen intake	Faecal nitrogen	Urinary nitrogen	Nitrogen absorbed	Nitrogen retained	% N intake absorbed	% N intake retained
1.8 g protein/kg/day							
Milk	277	52	157	225	68	81.2	24.5
Opaque-2	295	72	140	223	83	75.6	28.1
Milk	271	42	152	229	77	84.5	28.4
1.5 g protein/kg/day							
Milk	187	31	88	156	68	83.4	36.4
Opaque-2	238	68	108	170	62	71.4	26.0
Milk	190	34	108	156	48	82.1	25.3

Source: Bressani, Alvarado and Viteri, 1969 alkali-processed opaque-2 maize when the level of protein intake was 1.8 g per kg per day. The data demonstrate differences in nitrogen absorption.

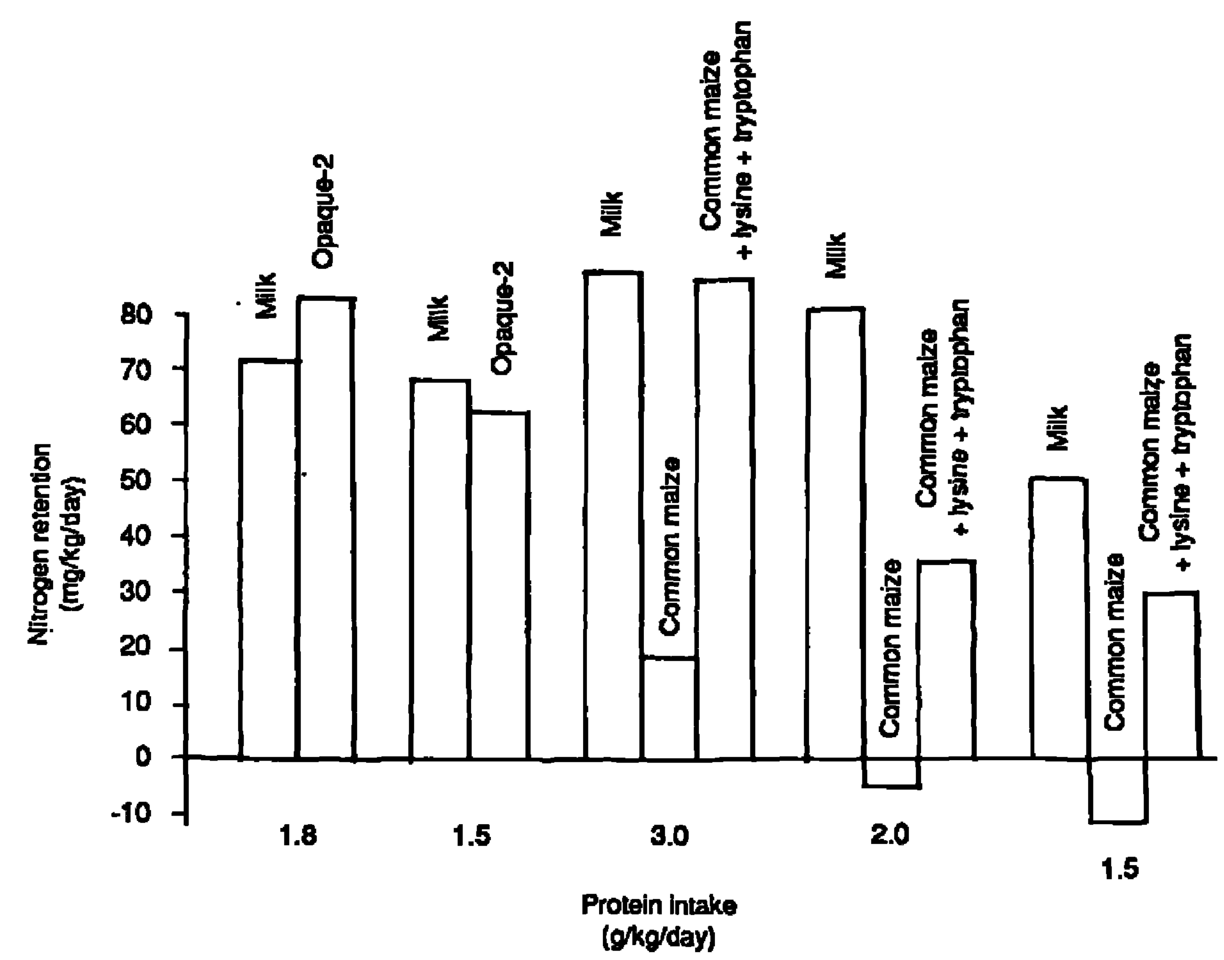
The apparent protein digestibility for processed opaque-2 maize averaged 73.5 percent in these studies. Based on faecal metabolic nitrogen determined in the children, true protein digestibility was 83.8 percent. From these results, it was concluded that the quantities of opaque-2 maize ingested by the children were 16.3 to 16.7 g and 12.9 to 14.5 g per kg body weight to take in 1.8 and 1.5 g protein per kg per day, respectively. These figures are equivalent to a total maize intake of 140 to 227 g per day, amounts similar to those commonly ingested by children in Guatemala.

With the data obtained in this study and data on urinary endogenous nitrogen, the relationship between nitrogen absorption and retention from milk and from opaque-2 maize was calculated. This nitrogen balance index constitutes a good measure for the biological value of proteins. The index was 0.80 for milk and 0.72 for opaque-2 maize, establishing then that the protein value of opaque-2 maize is equivalent to 90 percent of the biological value of milk. When the figure for true digestibility was used, the biological value of opaque-2 maize protein was calculated to be 87.1 percent. The figures also indicated that 90 mg of nitrogen must be absorbed from opaque-2 maize to obtain a nitrogen equilibrium.

For comparative purposes the same type of analysis was carried out for common maize in children (Scrimshaw *et al.*, 1958; Bressani *et al.*, 1958, 1963). Data on the nitrogen balance index were obtained from various studies in which children were fed with maize proteins as the only protein source in their diet. The biological value calculated was 32 percent. These data demonstrated again the low quality of common maize protein.

The difference between the nutritive value of opaque-2 maize protein and that of common maize is clearly shown in Figure 2, obtained from data in the studies described above. This figure shows the nitrogen retention in groups of children fed exclusively with opaque-2 and in others fed with common maize, in both cases at different protein intake levels. The supplementation effect of lysine and tryptophan on common maize is also shown. Even at intakes of 400 or 500 g of common maize, nitrogen retention is quite low, and this decreases to lower levels when the intake is reduced to

FIGURE 2
Nitrogen retention of children fed milk, common maize (alone and supplemented) and opaque-2 maize



200 or 300 g per day. With opaque-2, on the contrary, intakes of 140 or 230 g per child per day induced positive retentions that exceeded even those obtained with common maize supplemented with lysine and tryptophan. This suggests that it may be necessary to supplement common maize with other amino acids to make it comparable in protein value to opaque-2 maize.

The difference between opaque-2, common maize and the latter supplemented with lysine and tryptophan can be attributed to the better essential amino acid pattern found in opaque-2 maize, since the digestibility of the three is essentially the same. QPM maize also has a lower leucine content, implicated in the low nutritional value of maize.

The information presented clearly indicates the superiority of opaque-2 maize protein to that of common maize, a fact that is of great importance for populations consuming large quantities of maize as part of their habitual diet.

In a study by Luna-Jaspe, Parra and Serrano (1971) the nitrogen retention of common maize, Colombian opaque-2 maize (ICA H-208) and milk was compared in three children aged 24 to 29 months and weighing 5.9 to 10.1 kg. The protein and calorie intakes were approximately 1 g and 100 calories per kg body weight daily. Nitrogen retention was negative when the children received the opaque-2 maize. Common maize, however, gave an even lower or more negative figure. When milk was given, one child showed a negative balance and the other two a positive one, with the average balance on the positive side.

The authors indicated that the apparent protein digestibility of common maize was 61.5 percent, opaque-2 maize 57.9 percent and milk 66.4 percent. They also concluded that opaque-2 maize is of a higher nutritional value than common maize. They pointed out, however, that its use for young children with a rapid growth rate should be carefully controlled, and they could not recommend it as the main source of daily protein intake.

The results of these investigators are in agreement with those reported by other workers (Bressani, Alvarado and Viteri, 1969). The latter found that with 90 mg N absorbed per kg body weight per day, nitrogen equilibrium was obtained. The investigators in Colombia found that 90 mg absorbed nitrogen yielded a relatively low negative retention, while 100 mg absorbed nitrogen yielded nitrogen equilibrium. The differences between the results were not significant, and they could be explained by the age of the children, who were younger in the Colombian study and had lower body weight than the subjects used in the 1969 study. A more important factor was the lower protein intake. In any case, the data suggest that a minimum daily intake of approximately 125 g of opaque-2 maize might guarantee nitrogen equilibrium. This could not be obtained by using even twice the amount of common maize.

Similar studies were conducted by Pradilla *et al.* (1973) using the same variety of maize but with the opaque-2 gene (H-208 opaque). A crystalline endosperm containing the opaque-2 gene was also tested. The results are shown in Table 36, in which similar values may be observed for digestibility, biological value and nitrogen retention for the two maize selections containing the opaque-2 gene. These values were slightly lower than those for casein but significantly higher than values for common maize. In more recent studies Graham *et al.* (1989) evaluated QPM Nutricta, a maize variety containing the opaque-2 gene. This maize is high yielding, has a hard endosperm and contains high levels of lysine and tryptophan, although not as high as those in the original opaque-2 maize first studied. These authors used six male children aged 7.9 to 18.5 months who were recovering from malnutrition. Common maize and QPM as well as a casein diet were fed to provide 6.4 percent of the calories as protein. Total energy intake was approximately 125 kcal per kg per day, which was calculated to support weight and growth at previously established rates. The nitrogen balance results are shown in Table 37. Nitrogen absorption from QPM and common maize was 70 and 69 percent respectively, and 82 percent from casein. Nitrogen retention as a percentage of intake was 32 percent for QPM as compared with 41 percent for casein and 22 percent for common maize. These results, like others previously reported, confirm the great superiority of opaque-2 maize to common maize as food for children.

TABLE 36

Comparative nitrogen balances in children fed QPM and common maize

Protein	Protein digestibility (%)	Net protein utilization (%)	Biological value (%)	Nitrogen source retention (*g/day*)
Casein	98	75	77	1.81
H-208 opaque	91	89	76	1.52
H-208 crystalline[a]	87	65	75	1.50
H-208 common	78	36	47	0.93

[a]High lysine and tryptophan
Source: Pradilla *et al.*, 1973

TABLE 37

Digestibility and energy and protein use from common maize, quality protein maize and casein, measured in six infants

Products	Faecal nitrogen	Wet weight (*g/day*)	Energy (*kcal/day*) [% of intake]	Fat (*g/day*) [% of intake]	Carbohydrate (*g/day*) [% of intake]	Nitrogen intake (*mg/day*) [% Absorbed % Retained]	Body weight gain		Serum albumin (*g/100 ml*)	
							(*g/day*)	(*g/kg/day*)	Change	Final
Casein	80 ± 33[a]	14 ± 4[a]	48 ± 11[a] [6 ± 1[a]]	1.3 ± 0.4 [14 ± 4]	5 ± 2[a] [3 ± 1][a]	2 181 ± 292 [82 ± 4[a] 41 ± 9[a]]	33 ± 6	5.4 ± 1.0	0.0 ± 0.4	3.8 ± 0.3
QPM	108 ± 28[a]	29 ± 7[b]	121 ± 27[b] [13 ± 2[b]]	0.9 ± 0.3 [9.0 ± 4]	22 ± 7[b] [11 ± 3[b]]	2 273 ± 295 [70 ± 5[b] 32 ± 4[b]]	25 ± 16	3.9 ± 2.7	0.0 ± 0.4	3.7 ± 0.5
Common maize	129 ± 19[b]	34 ± 5[a]	137 ± 17[a] [16 ± 1[a]]	1.3 ± 0.3 [13 ± 4]	24 ± 4[b] [13 ± 1[b]]	2 256 ± 299 [69 ± 7[b] 22 ± 10[a]]	18 ± 16	2.6 ± 2.3	-0.3 ± 0.3	3.6 ± 0.4

Note: Like parameters with different letters are significantly different (P>0.05). Values are given as means ± SD

Source: Graham *et al*., 1989

Graham *et al.* (1980) and Graham, Placko and MacLean (1980) also reported on studies of eight convalescent malnourished children, 10 to 25 months of age, who were fed opaque-2 and sugary-2 opaque-2 endosperm and the whole kernel. Protein was fed so as to provide 6.4 percent of total calories, and the diets provided 100 to 125 kcal per kg body weight per day. The results showed an apparent N retention from the endosperm meal lower than that from the whole kernel meals, and both were lower than from casein. The difference between whole kernel and endosperm nitrogen retentions can probably be attributed to the amino acids contributed by the germ. The same researchers reported on plasma-free amino acids in the studies described above and concluded that the types of maize tested were possibly limiting in lysine, tryptophan and isoleucine.

These authors also reported that for the children to match N retention from casein, presumably equal to the requirement, they would have to consume 203.9, 148 or 122.5 percent of their energy requirements as common, opaque-2 or sugary-2 opaque-2 endosperm meals, which is impossible. For whole meals, they would have to consume 108.2, 90.3 or 84.2 percent of their energy as common, opaque-2 or sugary-2 opaque-2 maize.

Growth studies of children fed QPM have been conducted by various workers, among them Amorin (1972) and Valverde *et al.* (1981). In all reports, QPM was significantly superior to common maize and only slightly below the growth response observed when milk was fed.

Graham *et al.* (1989) stated: "To anyone familiar with the nutritional problems of weaned infants and small children in the developing countries of the world, and with the fact that millions of them depend on maize for most of their dietary energy, nitrogen and essential amino acids, the potential advantages of quality protein maize are enormous. To assume that these children will always be given a complementary source of nitrogen and amino acids is a cruel delusion."

Human adults

Two studies evaluating the protein quality of opaque-2 maize for human adults have been published. In the first, Clark *et al.* (1967) used ten university students as subjects in two experiments. The maize utilized was

TABLE 38

Average daily nitrogen balance in adult human subjects fed at different intake levels of opaque-2 maize

Maize kernels	Human weight *(kg)*	Nitrogen[a] *(g)*		
		Faeces	Urine	Balance
300	64.4	1.38	4.33	0.29
250	64.6	1.23	4.63	0.07
200	64.9	1.17	4.93	-0.09
150	65.0	0.97	5.37	-0.34

[a]Total nitrogen intake: 6.00 g
Source: Clark *et al.*, 1967

finely ground and included the whole grain. It contained 11 to 12 percent protein, 4.65 g lysine per 16 g N and 1.38 g tryptophan per 16 g N, values similar to those of the opaque-2 maize used in the study of children by Bressani, Alvarado and Viteri (1969). The maize was given in quantities of 300, 250, 200 and 150 g per day, which provided 5.58, 4.65, 3.72 and 2.79 g nitrogen per individual per day. The results of one experiment are shown in Table 38. All the individuals were in positive balance with an intake of 300 g of the maize and all of them were in equilibrium when they were administered 250 g. The 200 and 150 g levels resulted in a negative balance. With these data the regression equation between nitrogen balance and maize consumed was calculated. On the average, nitrogen equilibrium was obtained with an intake of 230 g.

The same authors studied the effect of lysine or tryptophan supplementation alone. Only one subject showed improved nitrogen retention. The addition of methionine did not induce any change. This indicated that the protein of opaque-2 maize was not deficient in these three amino acids for adult human subjects. Similar results were reported by Clark *et al.* (1977) for adult human subjects fed QPM and sugary-2 opaque-2 maize.

Unfortunately no studies have been done on adult human subjects comparing opaque-2 and common maize in the same study. The protein

quality of common maize has, however, been evaluated in human adults by Kies, Williams and Fox (1965). In one study ten subjects were fed degermed maize to provide nitrogen intakes of 4, 6 and 8 g per day. The results clearly indicated that when the degermed maize provided 4 and 6 g of nitrogen, the average nitrogen balance was negative. When the intake increased to 8 g of nitrogen per day, the balance became positive. The regression between nitrogen intake and nitrogen retained was calculated. From the equation it was calculated that 6.9 g of degermed maize nitrogen was necessary to give nitrogen equilibrium. The regression coefficient, multiplied by 100 and divided by the protein digestibility, gives the biological value of the protein. In the present case this value was 46.5 percent.

Based on 8.0 g protein per 100 g degermed maize, an intake of 6.9 g nitrogen is equivalent to 539 g maize. This figure is close to levels of maize consumed by adults in Mexico, Guatemala and El Salvador.

In the study described above lysine and tryptophan added alone did not produce changes in average nitrogen retention. When both amino acids were added, however, nitrogen retention increased – not necessarily because of the higher amount of nitrogen being administered with the addition of these two amino acids. This possibility may be discarded in view of the response obtained when non-specific nitrogen was added. These data demonstrate that the common maize protein is deficient in lysine and tryptophan for adult humans, as it is for children (see above in this chapter).

The results of these studies of amino acid intake from QPM and common maize (Clark *et al.*, 1967; Kies, Williams and Fox, 1965) are compared in Table 39. As shown earlier in this chapter, twice as much common maize is necessary to obtain nitrogen equilibrium in adults. This is equivalent to a protein intake of approximately 1.6 times more from common maize than from opaque-2. EAA intake follows the same trend as total nitrogen intake.

Using a biological value of 82 percent for opaque-2, of the 28 g ingested about 23 g are retained, which is the approximate amount (21 g) retained from common maize, which has a biological value of 46.5 percent. These data indicate the great losses of nitrogen occurring with common maize. With the exception of lysine and tryptophan, common maize provides a

TABLE 39

Protein and amino acid intake of opaque-2 and common maize needed to obtain nitrogen balance (g/day)

	Opaque-2	Common
Maize	250	547
Protein[a]	27.9	43.8
Isoleucine	1.01	2.00
Leucine	2.70	5.60
Lysine	1.34	1.25
Methionine	0.60	0.80
Cystine	0.55	0.56
Phenylalanine	1.33	1.96
Tyrosine	1.14	1.64
Threonine	1.10	1.72
Tryptophan	0.39	0.26
Valine	1.54	2.20
Total amino acids	11.70	18.99

[a]Protein digestibility of opaque-2 maize, 76.5%; biological value of common maize protein, 46.5%
Sources: Clark *et al.*, 1967; Kies, Williams and Fox, 1965

greater quantity of essential amino acids. They are, however, a load the body has to discard, a load that is greater in the cases of leucine, tyrosine and valine. The physiological cost of metabolizing these unnecessary amino acids is unknown, but it should be estimated.

Furthermore, the amino acid intake pattern is unbalanced, which may be an additional reason for the poor biological value of the common maize protein. Another method of analysing intake of individual amino acids is to express it as a percentage of the total amino acid intake, a calculation which magnifies the deficiency in lysine and tryptophan in common maize and also indicates the excess of other amino acids. This information, in reference to adults as well as children, demonstrates once more the excellent quality of opaque-2 maize protein and the poor quality of common maize protein.

BIOLOGICAL VALUE OF PROTEIN OF COMMON MAIZE AND QPM

No direct comparative studies are available on the digestibility and biological value of common and opaque-2 maize proteins, so to make a comparison between them the studies of common maize by Truswell and Brock (1961, 1962) and of opaque-2 maize by Young *et al.* (1971) will be used. In one of the studies conducted by Truswell and Brock (1962), the experimental subjects received 90 percent of their nitrogen intake from maize and 10 percent from other foods. A positive nitrogen balance was obtained when the nitrogen intake was more than 7 g per day, although great variability was found as in other studies. The authors calculated the biological value, which averaged 45 percent at a high intake level and 57 percent at a lower level of nitrogen intake. The difference was to be expected, since the biological value of a protein depends on the level of protein intake. Since all the experimental subjects showed a positive nitrogen balance when the intake was high, the authors concluded that the biological value of maize is close to the 57 percent figure. Similar results were found by Young *et al.* (1971). Truswell and Brock (1961) also found that in adult human subjects fed maize, the addition of lysine, tryptophan and isoleucine increased nitrogen balance from 0.475 to 0.953 g N per day in one study and from 0.538 to 1.035 g N per day in a second study. The flour fed was degermed maize flour, in which deficiencies are more apparent.

The biological value of opaque-2 maize protein was studied by Young *et al.* (1971). Egg protein was used as reference, fed at intakes of 2.64 to 3.95 g nitrogen per day. The authors calculated true protein digestibility and biological value from the faecal metabolic nitrogen and urinary endogenous nitrogen. The protein digestibility of opaque-2 maize protein varied from 67 to 106 percent, with an average for the eight individuals in the study of 92 percent, while the variability for egg protein was from 78 to 103 percent with an average of 96 percent. The average biological value for opaque-2 maize was 80 percent, and for egg the average was 96 percent.

Practical significance of protein evaluation of opaque-2 maize

The evidence presented from studies in both children and adults clearly indicates the superiority of opaque-2 maize over common maize. In spite of this, of the maize-consuming countries only Colombia and Guatemala have made efforts during the last few years to introduce this superior maize into agricultural production systems. The reasons are not clear, since agronomic studies conducted in a number of locations have shown that there are no differences between QPM and common maize in cultural practices, yield per unit of land and physical quality of grain. Furthermore, the plants look alike; QPM kernels are crystalline and grain yields are comparable to those of common maize. These factors are perhaps more important to growers than the nutritional advantages offered by QPM.

Energy content is alike in both types of maize, but the protein content of QPM is higher and is better utilized because of its better essential amino acid balance. The protein value of opaque-2 maize, however, can be analysed from other points as well. The information in Table 39 could be used to decide whether to introduce the opaque-2 maize varieties in grain-consuming countries.

It has been established that the intake of both types of maize as well as their nitrogen content (protein) are alike, but their digestibility percentages are very different: of 48 g of nitrogen intake from common maize, only 39.4 g are absorbed and 8.6 g are lost in the faeces. In the opaque-2 maize, of the 48 g of nitrogen intake, 44.2 g are absorbed and 3.8 g are lost in faeces.

The factor that should be considered, then, is the biological value, which is defined as the amount of absorbed nitrogen needed to provide the necessary amino acids for the different metabolic functions. The biological value of common maize is 45 percent; from the 39.4 g absorbed, 17.7 g are retained and 21.7 g are excreted. In opaque-2 maize the biological value of the protein is 80 percent; of the 44.2 g of absorbed nitrogen, 35.4 g are retained and 8.8 g are excreted. The total amount of nitrogen lost when common maize is consumed equals 30.3 g, while only 12.6 g are lost when the same amount of opaque-2 maize protein is consumed. In other words, only 37 percent of the common maize intake is utilized, compared to 74 percent from opaque-2 maize. The production and consumption of QPM in maize-eating countries would therefore have a significant beneficial effect on the nutritional state of populations, with important economic implications from the better use of what is produced and consumed.

Chapter 7

Approaches to improving the nutritive value of maize

Because of the great importance of maize as a basic staple food for large population groups, particularly in developing countries, and its low nutritional value, mainly with respect to protein, many efforts have been made to improve the biological utilization of the nutrients it contains. Three approaches have been tried: genetic manipulation, processing and fortification. Abundant data show great variability in the chemical composition of maize. Although environment and cultural practices may be partly responsible, the variability of various chemical compounds is of genetic origin; thus composition can be changed through appropriate manipulation. Efforts in this direction have concentrated on carbohydrate composition and on quantity and quality of oil and protein. Some efforts have also been made to manipulate other chemical compounds such as nicotinic acid and carotenoids. Processing is not widely recognized as a means of improving nutritive value; however, examples are presented to show its effects and potential. Finally, there have been many efforts to fortify maize, with outstanding results, but unfortunately fortification has not been implemented to a large extent. This approach, however, may become important in the future as more people consume industrially processed foods, which can be more easily and efficiently fortified.

GENETIC APPROACHES

Carbohydrates

The component with the greatest concentration in the maize kernel is starch. Since the plant accumulates starch in the endosperm, which is subject to genetic influence, starch can become a good source of energy. The quantity and quality of the carbohydrate fraction can be modified by breeding as described in recent reviews by Boyer and Shannon (1983) and Shannon and Garwood (1984). The waxy gene (*Wx*) in waxy maize has been shown to control amylopectin starch in the endosperm up to 100 percent with very low amounts of amylose (Creech, 1965). Other genes and gene combinations have been shown to be responsible for the composition of the starch in the endosperm. The amylose-extender gene (*Ae*) increases the amylose fraction of the starch from 27 to 50 percent (Vineyard *et al.*, 1958). Other genes cause an increase in reducing sugars and sucrose. Sugary (*Su*) genes produce relatively high amounts of water-soluble polysaccharides and amylose. Maize kernels containing this gene are sweet and are important for canning. Their starch content and quality also have nutritional implications, since some starch granules have low digestibility while others have high digestibility, as demonstrated by Sandstead, Hites and Schroeder (1968). These researchers suggested that maize varieties with waxy or sugary genes could be of better nutritional value for monogastric animals because of the greater digestibility of the type of starch they produce.

Protein quantity

Classical studies at the University of Illinois demonstrated the feasibility of changing the protein content of the maize kernel by selection. In these studies it was shown that protein content could be increased from 10.9 to 26.6 percent in the high-protein (HP) strain after 65 generations of selection. The low-protein strain contained about 5.2 percent. Dudley, Lambert and Alexander (1974) and Dudley, Lambert and de la Roche (1977) demonstrated that the protein content of standard inbred lines could be increased by crossing with the HP strain from Illinois and then backcrossing to the inbred line. Woodworth and Jugenheimer (1948) concluded that total protein content could be increased by selection in an open pollinated variety or by crossing standard inbred lines with an HP strain followed by back-crossing and selection in segregating populations.

The full expression of the protein genes in maize can be attained with appropriate levels of nitrogen fertilizers. Tsai, Huber and Warren (1978, 1980) and Tsai *et al.* (1983) showed that nitrogen fertilization of maize increased total protein because of an increase in prolamine content. Studies conducted by others showed, however, that the protein quality of the HP strains was lower than that of common maize since the increase in protein was due to an increase in the prolamine fraction. Eggert, Brinegar and Anderson (1953), from studies of pigs, showed that HP maize had lower biological value than common maize, which they attributed to the higher prolamine content in HP than in normal protein maize. The value of an HP maize kernel will depend on how it behaves agronomically and economically compared with maize with about 10 percent protein. The data available show that these types of maize not only require more soil nitrogen but also yield less than normal protein maize.

Protein quality

The low protein quality of maize stems mainly from the deficiency in the protein of the essential amino acids lysine and tryptophan. Still, variability in both amino acids has been shown (Bressani, Arroyave and Scrimshaw, 1953; Bressani *et al.*, 1960). As early as 1949, Frey, Brimhall and Sprague were able to show the genetic variability in tryptophan content in a cross between the Illinois HP and LP strains as well as in hybrids. Biological testing in which maize strains furnished the same level of protein in the diet also showed variability. All of these data suggest the feasibility of improving the quality of maize varieties. Mertz, Bates and Nelson (1964) found that the opaque-2 gene significantly increased the lysine and tryptophan content in maize endosperm. This gene also reduced the leucine level, giving a better leucine-to-isoleucine ratio. In 1965, Nelson, Mertz and Bates showed that the floury-2 gene when homozygous could also increase the lysine and tryptophan levels in maize. Research conducted at the Centro Internacional de Mejoramiento de Maíz y Trigo (CIMMYT) eventually yielded maize lines of QPM, which agronomically behave like common maize. As shown elsewhere in this book, the protein quality of these materials is significantly higher than that of common maize as shown by tests in humans.

Although such types of maize are available, it has been difficult to grow them commercially, even though the benefits to be derived from them by large maize-consuming populations would be high.

Oil

Genetic studies have also revealed that oil content in maize is subject to genetic influence, with diversity often found, although environment and agronomic practices can influence fatty acid composition (Jellum and Marion, 1966; Leibovits and Ruckenstein, 1983). As with protein content, mass selection over 65 years increased oil content from 4.7 to 16.5 percent. This increase was obtained through increases in the size of the germ. The problem with high-oil varieties is a low yield, although it has been reported that varieties with 7 to 8 percent oil yield as well as varieties with lower oil content. Besides total oil content, some studies have shown that the fatty acid content may also be subject to genetic control, as seen by changes in linoleic acid content in maize oil. Poneleit and Alexander (1965) suggested a single-gene or single-gene-plus-modifier effect. A multi-gene system of inheritance has been proposed by other investigators. QPM oil fatty acid composition was found to be similar to that reported for normal maize.

Other nutrients

Because of the association of maize consumption and pellagra and the low availability of nicotinic acid in maize, efforts have been made to increase niacin in maize by genetic means. Variability in 22 varieties planted in one location ranged from 1.25 to 2.6 mg per 100 g (Aguirre, Bressani and Scrimshaw, 1953). The problem with niacin in maize and in other cereal grains is that it is unavailable to the animal organism.

The other nutrient that has received some attention is carotene, a precursor of vitamin A. Results from some investigators have shown yellow maize to vary in vitamin A activity from 1.52 to 2.58 µg per gram. Cryptoxanthein contributed 38.3 to 57.3 percent of the total activity and beta-carotene the difference (Squibb, Bressani and Scrimshaw, 1957). Other researchers have indicated that provitamin A activity is under genetic control in the maize kernel.

PROCESSING

Often the processing of foodstuffs stabilizes nutrients in the food, but losses may take place when optimum conditions are exceeded. There are cases, however, in which processing induces beneficial changes in the food; a classic case is the elimination of antiphysiological factors in beans.

Lime-cooking

Lime-cooking of maize as described in Chapter 4 causes some losses in nutrient content, but it also induces some important nutritional changes. Its effects on calcium, amino acids and niacin content have already been described in Chapter 4.

Other processes

Besides the lime cooking process, other processes have been reported to improve the quality of maize. One such process is natural fermentation of cooked maize, which results in higher B-vitamin concentration and protein quality (Wang and Fields, 1978). *Pozol*, a food made from lime-treated maize allowed to ferment naturally, has been shown to be of higher quality than raw maize or tortillas. Germination of the grain has also been reported to improve the nutritional value of maize by increasing lysine and to some extent tryptophan (Tsai, Dalby and Jones, 1975; Martínez, Gómez-Brenes and Bressani, 1980) and decreasing zein content. A similar result was found with QPM.

FORTIFICATION

A third approach often used to improve the nutritive value of foods, mainly cereal grains, is fortification. Because of the great nutritional limitations in maize, many efforts have been made to improve its quality, and particularly that of its protein, through addition of amino acids or protein sources rich in the limiting amino acids.

Supplementation with amino acids

Raw maize proteins have been shown to be of a low nutritive value because of deficiencies in the essential amino acids lysine and tryptophan. Many studies conducted with animals have demonstrated that the addition of both amino acids improves the quality of the protein. Some workers have even found that besides lysine and tryptophan, isoleucine is also deficient, possibly because of an excess of leucine in maize proteins (Rosenberg, Rohdenburg and Eckert, 1960). Similar data have been obtained from studies with animals when lime-treated maize was supplemented with lysine and tryptophan (Bressani, Elías and Braham, 1968). These results have been confirmed in nitrogen balance studies conducted with children as shown in Chapter 6. (Selected results are shown in Table 32.) The finding that the addition of lysine and tryptophan at the lower levels of protein intake gave a nitrogen retention significantly higher than at the higher level of protein intake has often been overlooked, and the importance of protein quality has been overshadowed by that of energy intake.

Supplementation with protein sources

The results from animal and human studies in which limiting amino acids have been added to lime-treated maize have served as the basis for evaluating the ability of different types of protein supplements to improve its protein quality. Studies on protein supplementation of lime-treated maize flour have been published by many researchers using different food sources including milk, sorghum, cottonseed flour, fish flour, torula yeast and casein. Table 40 summarizes the results of adding small recommended amounts of various protein sources. The quality increase is at least 200 percent of the protein quality value of maize. In tests with young dogs, the nitrogen balances when maize was supplemented with 5 percent skim milk, 3 percent torula yeast and 4 percent fish flour were significantly higher than those measured when maize was given alone. Most of the supplements that have been tested have several characteristics in common. They all have a relatively high protein content and are good sources of lysine, with the exception of cottonseed protein and sesame oil meal. The latter is a good source of methionine. With the exception of casein and/or milk and fish protein concentrate, they are of vegetable origin.

The improvement in quality of protein in tortilla flour is in most cases a synergistic response to lysine and tryptophan enhancement and to a higher level of protein, both provided by the supplement. Since soybean protein in different forms is the supplement to tortilla flour most often tested by different investigators and because it is almost the only one also tested in

TABLE 40

Recommended levels of protein concentrates to improve the protein quality of lime-treated maize

Protein source	Recommended level (%)	PER
None	–	1.00
Casein	4.0	2.24
Fish protein concentrate	2.5	2.44
Soy protein isolate	5.0	2.30
Soybean flour	8.0	2.25
Torula yeast	2.5	1.97
Egg protein	3.0	2.24
Meat flour	4.0	2.34
Cottonseed flour	8.0	1.83

Source: Bressani and Marenco, 1963

children, with results comparable to those in studies with animals, its importance and effects are reviewed in this section. Figure 3 depicts the PER for combinations of common maize and opaque-2 maize with soybean flour in different ratios.

Studies show that maximum PER is achieved upon addition of 4 to 6 g percent soybean protein, whether from whole soy, soy flour (50 percent), soy protein concentrate or soy protein isolate (Bressani, Elías and Braham, 1978; Bressani *et al.*, 1981). For reasons of availability, cost and practical applications in developing countries, the results with whole soybean are discussed here. The 4 to 6 g percent level of supplementary protein can be provided by either 15 percent whole soybean or 8 percent soybean flour, which have resulted in comparable protein quality improvement. The advantage of using 15 percent whole soybeans is that supplementation can be carried out in the home with soybeans produced by the family; soybeans are very economical, and besides providing higher protein quantity and quality, they give some additional energy from the oil they contain.

TABLE 41

Nitrogen balance in preschool children fed milk, normal maize and soybean/lysine supplemented maize

Diet	No. of children	No. of balances	Chronologic age (*months*)	Height age (*months*)	Weight for height (%)	Protein intake (*g/kg/day*)	Nitrogen (*mg/kg/day*)			% Nitrogen	
							Intake	Absorbed	Retained	Absorbed	Retained
Milk	7	11	24	14	103	1.25	195 (173-210)	157 (114-181)	75 (40-106)	80 (61-87)	38 (22-50)
Normal maize	6	12	30	16	104	1.25	192 (183-198)	144 (129-157)	30 (10-55)	75 (66-80)	16 (5-30)
Maize + soybean + lysine	6	12	30	16	104	1.25	197 (189-203)	154 (144-169)	63 (52-77)	78 (73-85)	32 (25-38)

Source: Viteri, Martínez and Bressani, 1972

Tamalitos are often eaten instead of tortillas and have the advantage of remaining soft for a longer period. There are various ways to prepare them, some of which include the young leaves of native vegetables such as crotalaria and amaranthus. Chemical and nutritional studies have demonstrated that about a 5 percent contribution of these leaves improves the protein quality of the dough (Bressani, 1983). The reason is that they have relatively high levels of protein rich in lysine and tryptophan. They also provide minerals and vitamins, particularly provitamin A. Leaf protein concentrates have also been shown to improve the protein quality of cereal grains (Maciejewicz-Rys and Hanczakowski, 1989).

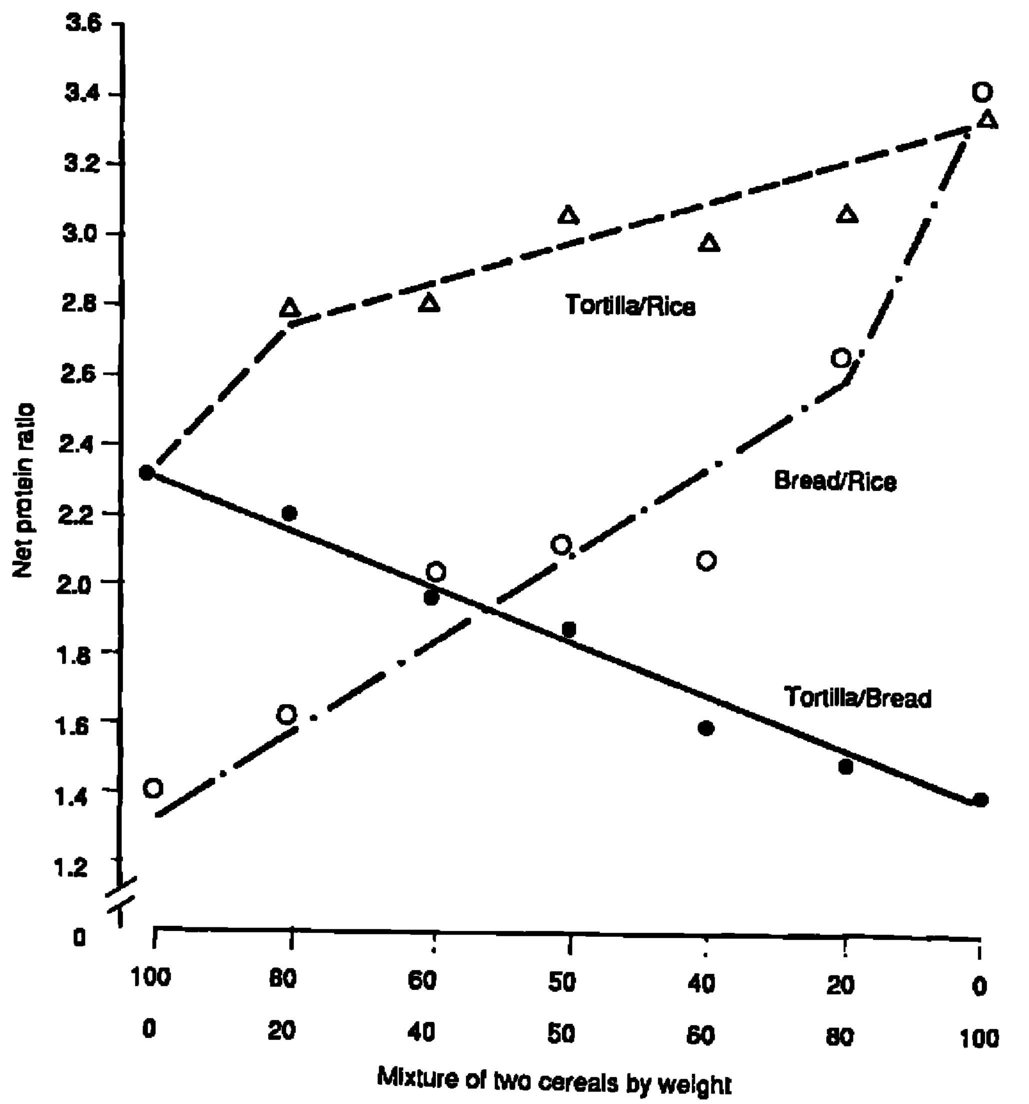
Supplementation with other grains

Sorghum is another grain that has been processed by lime-cooking in Mexico and Central America, particularly in areas where maize does not grow well. Sorghum tortillas, however, are not of the same organoleptic or nutritional quality as maize tortillas. Many successful efforts have been made to use blends of both cereal grains, among others by Vivas, Waniska and Rooney (1987) and Serna-Saldivar *et al.* (1987, 1988a, 1988b). Other approaches include the use of blends of common maize, since germination has been reported to increase lysine. Mixtures of tortilla flour and rice and of tortilla flour and wheat flour have also been studied. The rice/maize products have higher nutritive value than the wheat/maize tortillas, as shown in Figure 4. These results show the superiority of rice over whole maize flour and of the latter over wheat flour. More recently, blends of amaranth grain with lime-cooked maize flour have been shown to have an improved protein quality because of the much higher lysine and tryptophan content of amaranth as compared with maize. The product has been reported to be of an acceptable organoleptic quality. Other products added include potato, rice and pinto beans, providing foods with acceptable sensory attributes.

High-quality protein foods

The nutritional value of maize, particularly maize protein, can also be improved by protein complementation. In this approach, the objective is to

FIGURE 4
Protein value of mixtures of two cereals



8.1	9.2	10.4	11.1	11.5	12.6	13.8	T/B
8.1	7.9	7.0	7.3	7.2	7.0	6.8	T/R
13.8	11.8	11.1	10.2	9.4	7.9	6.8	B/R

Protein (%)

T= Tortilla R= Rice B= Bread

Source: Bressani and Elías (1981)

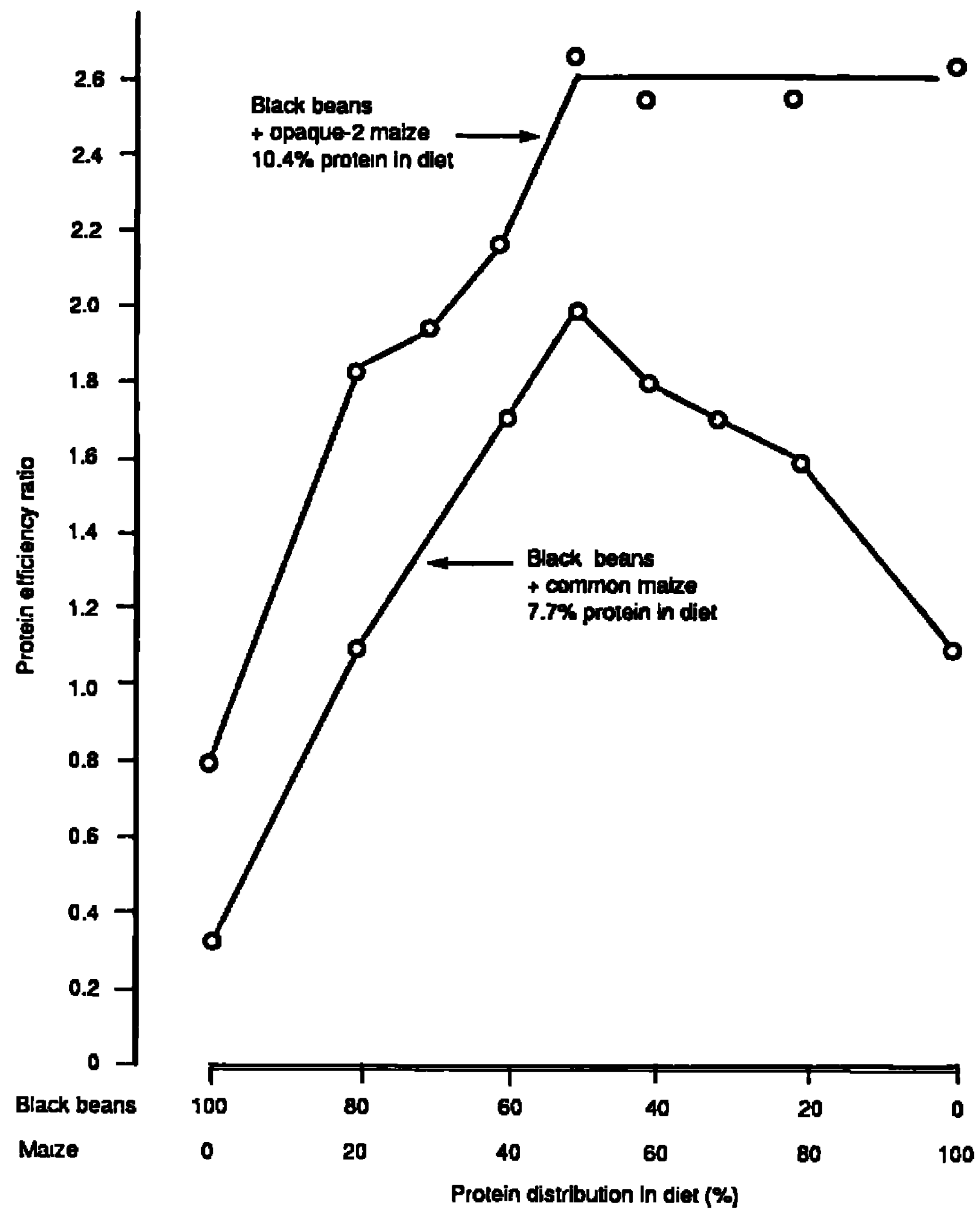
combine two or more protein sources with maize to maximize the quality of the product by achieving a good balance of the essential amino acids. Using this approach a number of high-quality foods have been developed. (Similar results can be obtained with other cereal grains.)

An example, complementation of both common and QPM maize with common black beans, is shown in Figure 5. Here the isonitrogenous replacement of the bean nitrogen by QPM nitrogen resulted in a constant PER increase up to a level corresponding to 50 percent of the protein from each component, with no further change as the nitrogen of the mixture was provided increasingly from the QPM. A similar result is observed with mixtures of the beans and common maize, except that as more of the dietary nitrogen is provided by maize, the protein quality drops. Further studies indicated that on the left side of the peak response the limiting amino acid was methionine, while on the right side it was lysine. The peak was obtained through the contribution of lysine from beans to maize and the contribution of methionine from maize to beans. This response has served as the basis for formulating high-quality protein food mixtures containing 70 percent maize and 30 percent common beans.

A similar type of response is observed with mixtures of normal and QPM maize and soybean flour. The peak mixture is equivalent to 77 percent maize and 23 percent soybean flour on a weight basis. When whole soybean flour is used, however, the mixture by weight is 70 percent maize and 30 percent whole soyflour. This product is called *maisoy* and is commercially produced in Bolivia. It is used to improve lime-treated maize for tortillas or as a wheat flour extender for bakery products. Other oil seed flours have been used in a similar fashion, for example cottonseed flour (CSF) and maize. In this case there is no synergistic effect of complementation. Optimum quality mixtures can be obtained when CSF provides about 78 percent of the protein and maize 22 percent. This distribution by weight is equal to 40 percent CSF and 60 percent maize flour, which is the ratio for *incaparina* produced in Guatemala since 1960.

Many other mixtures of maize and other foods have been developed. The United States Department of Agriculture has been involved since 1957 in product and process development, and products such as instant and

FIGURE 5
Protein efficiency ratio of combinations of common or opaque-2 maize and black beans



	Lysine	Tryptophan mg/g N	TSAA
Common maize	180	38	197
Opaque-2 maize	306	94	234
Black bean	484	58	125

Source: Bressani (1988)

sweetened corn-soya milk and corn-soya bread are well known throughout the developing world. Many other mixtures have been developed with common maize or QPM and other protein sources, giving products of high nutritional value and acceptability.

Chapter 8

Improvement of maize diets

In nutritive value maize is quite similar to other cereal grains. In fact, it is somewhat superior to wheat flour and only to a small extent below rice. These are the three cereal grains most consumed by people throughout the world. The problem with maize lies in the diet of which it is a component, a diet mostly deficient in the kind of supplementary foods necessary to upgrade the nutrients ingested in relatively large amounts of maize. Maize-consuming populations would be nutritionally better off if the maize consumed had the lysine and tryptophan genes of QPM or if it were consumed with a sufficient amount of protein foods such as legumes, milk, soybeans and amaranth seeds and leaves. This, however, will not occur in the near future, and therefore other measures should be taken. In this section, a number of possibilities, the results of studies to improve the nutritional quality of maize-based diets, are presented.

MAIZE/LEGUME CONSUMPTION

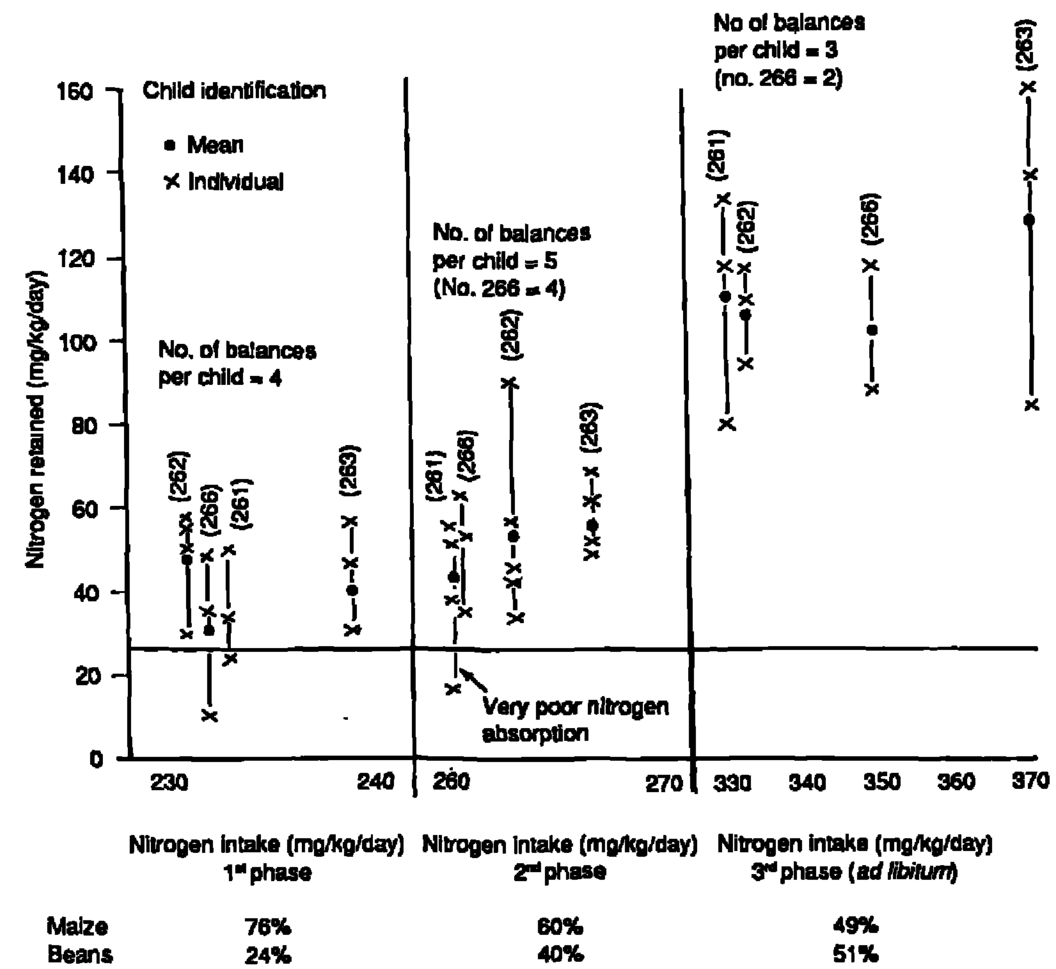
Throughout the world, particularly in developing countries, the diet of populations is based on the consumption of a cereal grain, usually maize, sorghum or rice, and a food legume, either common beans or any of the others known. Results of many studies have shown that these two types of basic food nutritionally complement each other. A complementary effect was observed, for example, when animals were fed diets in which the protein was derived from maize and common beans in various proportions from 100:0 to 0:100. When each component provided close to 50 percent of the protein in the diet a high quality was obtained, higher than the individual qualities of the components alone. The reason for this is the essential amino acid make-up of each component. Maize protein is deficient in lysine and tryptophan but has fair amounts of sulphur-containing amino acids (methionine and cystine). On the other hand, the protein of food legumes is a relatively rich source of lysine and tryptophan but is low in sulphur amino acids (Bressani and Elías, 1974). From these studies it was concluded that the protein of beans or food legumes complements the protein of maize best in the proportion of 30 parts beans to 70 parts maize.

This complementarity is also found in maize with cowpeas, with mung beans, with soybeans and with other food legumes. The response is the same even if the protein level in the diet is not fixed as in the example above but varies depending on the protein content of each component. Beneficial results have been obtained with oil added to the diet in amounts from zero to 10 percent. It is also of significance that food intake was highest at the maximum level of complementation. That is, a higher energy intake was also observed.

The great importance of protein quality has been missed by those who claim that energy is more limiting in diets than protein. The complementary effect described above has also been shown to occur in humans. Nitrogen balance was tested in studies of children fed lime-treated maize and beans in two fixed ratios and *ad libitum*, as selected by the children (Figure 6). The nitrogen balance at the fixed ratio in Phase 1 (76:24) was lower than when they were fed a ratio of 60:40 maize to beans (Phase 2). Nitrogen balance improved when the children were allowed to select, and the selection was close to seven parts maize and three parts beans by weight. Equally important is the fact that total food intake also increased. The usual intake of maize and beans established by dietary surveys in the 1960s varied between 11:1 and 18:1; therefore the supplementation of beans was relatively small. More recent data (Garcia and Urrutia, 1978) for three-year-old children gave an 8:4 maize-to-bean ratio, and the ratio was even poorer for children 6 to 11 months of age.

Combinations of maize and bean protein, although of relatively high protein value in tests in animals, are not adequate for the treatment of children with protein malnutrition. Furthermore, the increase found by Arroyave *et al.* (1961) in plasma amino acid levels following a test meal of milk was much higher after a period of treatment with a 1:1 maize-bean combination than the response observed when milk protein was given after

FIGURE 6
Nitrogen retention of children receiving maize/bean diets



treatment with either milk or a vegetable mixture made of maize, cottonseed flour, torula yeast and minerals (Bressani and Scrimshaw, 1961). These results confirmed the inadequacy of the maize and bean diet. Likewise, nitrogen balance results in children fed maize and bean mixtures as compared with milk and other vegetable proteins have been relatively low. Gomez *et al.* (1957) reported on nitrogen balance experiments made on eight children, one to five years old, with chronic severe malnutrition, who were kept on a diet of maize meal and beans during the experiments. Both nitrogen absorption and retention were extremely variable from child to child; four children had a positive nitrogen balance, and four negative. The addition of tryptophan and lysine to the maize-bean diet greatly improved nitrogen absorption and nitrogen retention in four cases. In these studies, no indication was given on the amounts of maize and beans mixed, and protein intake varied from 1.53 to 8.50 g per day. Frenk (1961) also found poor performance in children fed maize and beans. A significant improvement was obtained upon supplementation with fish meal.

In common with other investigators, Hansen (1961) found that milk initiated the cure of kwashiorkor without difficulty; a two-component mixture of 66 percent maize meal and 33 percent cowpea meal, however, did not initiate a cure in the three cases treated with it. A three-component mixture made up of equal parts of maize meal, maize germ and cowpea (*Vigna sinensis*) brought about satisfactory recovery in the one case in which it was employed.

It would require 238 g of the dry three-component mixture and 267 g of the two-component mixture to supply the essential amino acids contained in 100 g of skim milk. Since the vegetable formulas also require greater dilution, it is difficult to supply enough of them to meet protein needs.

Scrimshaw *et al.* (1961) considered that excessive bulk relative to protein content was a major reason for the lack of success in initiating cure of kwashiorkor with mixtures of maize and beans. Hansen *et al.* (1960) stated that the differences in biological value of the proteins tested were clearly reflected in nitrogen retention, which averaged 13 to 14 percent for milk, 8.8 percent for the two-component mixture and only 5.7 percent for the three-component mixture. It was concluded that the two- and three-component mixtures were each adequate to prevent kwashiorkor after initial recovery from the disease but that only the three-component mixture had proteins sufficient in concentration and quality to be satisfactory for use in treatment.

It should be noted that the two-component mixture of 66 percent maize meal and 33 percent cowpea meal is not the best combination of these two sources of protein. Bressani and Scrimshaw (1961) reported that in the best mixtures of these two foods, cowpea provided 50 to 75 percent of the protein, and maize 50 to 25 percent.

In other studies by Hansen *et al.* (1960) and Brock (1961), the nutritive value of maize alone and of maize supplemented with lysine and tryptophan, with pea flour and fish flour and with pea flour and milk was measured by means of nitrogen balance. Nitrogen retention was increased significantly by each form of supplementation, but at protein intakes of less than 2.5 g per kg body weight per day N retention was significantly less with the lysine and tryptophan supplement or the pea-flour supplement than with a milk diet. These differences disappeared at higher intakes of protein. The maize-pea mixture supplemented with 12 percent milk or with 10 percent fish flour resulted in nitrogen retentions comparable to those of a milk diet at all levels of protein intake. These variable results for bean and other legume seed proteins may be due to the type of legume seed used, to amino acid deficiencies or to some unknown factor. They deserve further investigation, because legume seeds have good potential for helping to solve the nutritional problems of the world.

Baptist and de Mel (1955) obtained a highly satisfactory response in 23 Ceylonese children one to six years old fed a mixed diet of three cereals and four legumes supplemented with skim milk. On the other hand, Navarrete and Bressani (1981) reported from nitrogen balance studies in adults that a bean diet produced nitrogen equilibrium at an intake of 114 mg N per kg per day; however, an 87:13 maize/bean mixture induced nitrogen equilibrium with an intake of 98 mg N per kg per day.

All these studies suggest that even though maize protein is improved in nutritive value upon addition of beans, its quality is still not fully adequate to feed infants and preschool children. This was evident when high-quality protein supplements were also tested with the maize/bean diet. Bulkiness limiting intake and nutritional quality are two factors of importance in maize/bean mixtures or diets.

LIMITING NUTRIENTS IN A MAIZE/BEAN DIET

Amino acids

It has been shown that adding 0.3 percent L-lysine HCl and 0.10 percent DL-tryptophan to a diet of 90 percent maize and 10 percent beans resulted in significant increases in weight gain and protein quality. These did not

TABLE 42

Effect on the nutritive value of a 90/10 maize/bean diet of the addition of lysine and tryptophan to maize or methionine to beans

Dietary treatment	Ave. weight gain (*g/28 days*)	PER
Maize Beans	69	2.11
Maize + lysine + tryptophan Beans	103	2.64
Maize Beans + methionine	66	1.93
Maize + lysine + tryptophan Beans + methionine	108	2.64

Note: Lysine 0.3% (L-lysine HCl); tryptophan 0.1%; methionine 0.3%
Source: Gómez-Brenes, Elías and Bressani, 1972

increase further when methionine was also added (see Table 42). The significance of protein quality in a system based on maize and beans was observed when diets of mixtures of maize and beans plus methionine were offered. The results confirmed the limitation of this amino acid in beans, since a response was observed when more beans were included in the diet. Likewise, those diets with maize and beans and methionine also induced the subjects to consume greater amounts of food or of energy, demonstrating thus the value of protein quality in stimulating food intake (Contreras, Elías and Bressani, 1980, 1981). The results also served to demonstrate that even with the best combination — that is, a 7:3 maize-to-beans ratio — the diet is still short of an adequate quality for small children, and it is even more so when the proportion of beans is lower.

Vitamins and minerals

A diet of maize and beans in the ratio of 7:3 responds to the single addition of a complete B-vitamin and fat-soluble mixture and more so to a complete mineral supplement, but not to calories or to lysine and tryptophan. The best results from double combinations have been obtained from minerals plus

amino acids, minerals plus vitamins, minerals plus calories, vitamins plus amino acids and vitamins plus calories. The addition of calories plus amino acids did not significantly improve either the weight gain of the subjects or the PER of the diet. For triple combinations an adequate intake of vitamins and minerals is needed before an effect from the amino acids can be obtained, since animals fed with amino acid enriched diets probably develop a vitamin and mineral deficiency. Although this may be obvious, it is usually not acted on in practice.

It was observed that animals on a diet enriched with amino acids developed vitamin and mineral deficiencies, and many died. This was attributed to a depletion of these nutrients caused by the catalytic effect of the improved protein quality on the potential of the animal to respond to this stimulus.

Provision of additional calories in the diet resulted in a slight decrease in the quality of the diet. This suggests that the addition of calories lowered the protein intake of the diet, which in turn reduced its quality by enhancing essential amino acid deficiencies in the mixture of maize and beans. Similar results were found by Contreras, Elías and Bressani (1980, 1981) using young growing rats and pigs fed maize/bean mixtures in either an 87:13 or a 70:30 weight ratio. These authors confirmed the results previously reported and indicated that one of the main constraints in maize/bean diets is their bulkiness, which does not permit greater intakes. Results of some of these supplementations in rats are summarized in Table 43.

A number of studies have been carried out to learn whether an increase in the protein content of the diet from an increase in maize and bean proteins would improve animal performance. These showed that the use in the maize/bean diet of a maize with 13 percent protein to replace one with 8.3 percent protein resulted in some increase in weight gain and in utilizable protein in spite of the fact that protein quality decreased as shown by PER and relative nitrogen value (RNV) figures. This was expected, since utilizable protein is the result of protein quantity and quality. When the two maize samples (low and high protein content) were supplemented in this maize/bean diet with lysine and tryptophan, a greater improvement in weight gain and utilizable protein was obtained than from the diet with the high-protein maize.

TABLE 43
Nutritive value of a 90/10 maize/bean diet supplemented with vitamins, minerals, calories and amino acids

Supplement	Ave. weight gain (g/28 days)	PER
None (basal diet)	26 ± 2.3	1.11 ± 0.07
+ Vitamin mixture	49 ± 4.0	1.55 ± 0.06
+ Mineral mixture	65 ± 4.3	1.94 ± 0.06
+ Calories (5% oil)	23 ± 1.2	0.95 ± 0.05
+ Amino acids[a]	26 ± 2.5	1.13 ± 0.08

[a]Lysine (0.3%); DL-tryptophan (0.10%)
Source: Bressani, 1990

Increases in weight gain and utilizable protein compared with the basal diet also resulted when the proportion of beans in the diet was increased from 10 to 20 percent, but these were lower when compared with the respective amino acid supplemented diets. These data were interpreted to mean that diets of maize and beans in a 90:10 ratio are limiting first in protein quality and to a lower extent in protein quantity (Gómez-Brenes, Elías and Bressani, 1972; Elías and Bressani, 1971; Bressani, Elías and de España, 1981). This is in agreement with the conclusions of Arroyave (1974), who indicated that for one- to two-year-old children to obtain an adequate nitrogen retention from maize and beans, similar to that from 1.27 g milk protein per kg body weight per day, 1.7 g protein per kg per day were required. These results show that the protein of common maize in the diet is improved by the addition of lysine and tryptophan.

IMPROVEMENT OF THE MAIZE/LEGUME DIET

Animal supplements

Various studies conducted with animals demonstrated that methionine is the limiting amino acid in diets containing more than 30 parts of beans, while those diets containing more than 70 parts of maize are limiting in lysine. The

diet giving the highest quality is deficient in both amino acids (Bressani, Valiente and Tejada, 1962). At the same time such diets are low in total protein content. Therefore, in order to improve the quality of maize/bean mixtures, it is necessary to add protein sources rich in both amino acids. Studies with animals fed diets based on maize, beans and various animal protein sources such as chicken or beef indicated that a 20 to 30 percent addition of animal protein would result in significant increases in nutritive value (Bressani, 1987). In experiments by other researchers, animals were fed *ad libitum* with 1, 2, 3 and 4 g of milk as a daily supplement to a diet of maize and beans. The results demonstrated that approximately 1 to 2 g milk per day added to a basal diet intake of 15 g per day was enough to increase the nutritional quality of the diet, evaluated from the protein quality point of view. In these studies 12 percent milk was found to be the minimum necessary to induce a relatively high improvement in the quality of the maize/bean diet. Furthermore, the effect of the supplement was more consistent when it was given on a daily basis. With growing dogs as experimental animals, Murillo, Cabezas and Bressani (1974) found 20 percent milk as the minimum complement to give the highest nitrogen balance to a maize/bean diet. This was not obtained when the basal maize/bean diet was supplemented with lysine, methionine and tryptophan as found in milk proteins. Torún and Viteri (1981) and Torún *et al.* (1984) showed in metabolic studies with children that a diet of maize and beans in a weight ratio of 85:15 with 18 percent animal protein (milk) would induce good and consistent biological responses. These authors concluded from the diet used in the study that protein intakes were adequate when energy intakes corresponded to the estimates of energy requirements.

QPM

Replacement of common maize by QPM is another alternative that could improve the quality of maize/bean diets. The results obtained by feeding animals with mixtures of QPM and beans showed that, as with common maize, optimum complementation takes place at approximately a 50:50 diet protein ratio, equivalent to 70:30 maize/beans by weight (Bressani and Elías, 1969). However, there are two differences that should be noted. One is that both weight gain of the animals and protein quality were higher with the QPM/bean blends than with the common maize/bean mixtures. The second point, possibly even more important, is that the weight gain and protein quality of mixtures with more than 70 parts of maize were no different from the values found for the best mixture, a 70:30 diet. Likewise, diet intake in a 28-day experimental period increased from 224 g per animal to 388 g at the maximum point and remained constant in all other diets with higher levels of QPM in the mixture.

In other series of studies the protein quality of QPM as a component of a maize/bean diet of 82.8 percent maize and 10.5 percent cooked beans was evaluated in young and adult dogs fed at two levels of protein (Bressani & Elías, 1972; Murillo, Cabezas and Bressani, 1974). The QPM/bean diet was compared with similar diets of common maize and beans and common maize supplemented with lysine and tryptophan and beans. Nitrogen balance data showed that nitrogen retention levels for young or adult dogs fed QPM/bean diets were as high as or higher than those in which common maize in the diet was supplemented with lysine and tryptophan, and the levels were significantly higher with both diets than with maize and beans alone.

These studies, as well as studies conducted with growing pigs, indicated also that maize/bean diets are bulky, which limits the amount that can be ingested to meet nutritional needs fully (Contreras, Elías and Bressani, 1980, 1981).

High-quality food mixtures

In many developing countries and for quite a long time, many efforts have been made to develop high-quality food mixtures that would supply the nutrients, particularly protein, provided by animal food products. Most of these foods have a relatively high protein content with a good essential amino acid pattern which can to some extent correct deficiencies of amino acids and of other nutrients in maize/bean diets, if consumed in the appropriate amounts. Studies have shown this supplementary effect to be present. Young growing animals were fed a basal diet of about 85 percent lime-treated maize and 15 percent cooked black beans. This diet was

properly supplemented with minerals, vitamins and energy. Groups of animals were fed daily 1, 2, 3 and 4 g of a high protein food based on maize, soybeans and skim milk. The results demonstrated that these levels, particularly the highest, effectively supplemented the basal diet, as judged by weight gain, protein utilization and biochemical parameters (de Souza, Elías and Bressani, 1970).

These diets with animal foods and with high-quality foods are effective because they are able to provide nutrients still deficient in diets based on maize and beans. Therefore, any food of animal origin and some foods of vegetable origin, such as soybeans and green leafy vegetables, would improve the quality of such diets.

Green vegetables

An examination of a maize/bean diet shows that besides protein quality, other nutrients are deficient. The effect of adding vitamins and/or minerals to such a diet has already been described. Other studies were conducted in which the basal maize/bean diet was supplemented with small amounts of leafy vegetables such as amaranth, spinach and *chipilín* (crotalaria). These leafy vegetables provide not only essential amino acids and protein, but vitamins and carotenes which supply to some extent the vitamin A needs of the animal.

Various vegetables as supplements to maize/bean diets have been reported on and results are shown in Table 44. Two sets of diets were tested, one with vitamins added and the other without. The level of addition was 5 percent dry weight. All vegetables in both sets of diets improved weight gain and increased diet intake. Utilizable protein was also higher in the maize/bean diets with vegetables than in the control, and it was highest with the leafy vegetables. Nutritional values were higher with added vitamins than without. These studies clearly indicate that nutritional improvement of 87:13 maize/bean diets is possible by providing vitamins, some additional protein and essential amino acids.

TABLE 44

Effect of various vegetables added to improve the nutritive value of a common (87/13) maize/bean diet

Vegetable	Without vitamins					With vitamins				
	Ave. wt. gain *(g/28 days)*	Food intake *(g)*	PER	RNV	Utilizable protein *(%)*	Ave wt. gain *(g/28 days)*	Food intake *(g)*	PER	RNV	Utilizable protein *(%)*
Potatoes	42	274	1.49	59.6	5.6	68	357	2.08	83.2	7.6
Carrots	50	287	1.83	73.2	6.9	65	349	2.04	81.6	7.4
Green peas	52	311	1.66	66.4	6.7	80	370	2.28	91.2	8.7
String beans	55	313	1.75	70.0	7.1	79	378	2.15	86.0	8.3
Spinach	56	282	1.82	72.8	7.9	103	417	2.36	94.4	9.9
Amaranth	67	327	1.96	78.4	8.2	100	420	2.32	92.8	9.5
Crotalaria	63	313	1.92	76.8	8.1	92	329	2.28	91.2	9.7
None	37	268	1.48	50.2	5.4	58	337	1.84	73.6	6.8

Bibliography

Abbas, H.K., Mirocha, J.K., Rosiles, R. & Carvajal, M. 1988. Decomposition of zearalenone and deoxynivalenol in the process of making tortillas from corn. *Cereal Chem.*, 65:15-19.

Abramson, D., Sinka, R.N. & Mills, J.T. 1980. Mycotoxin and odor formation in moist cereal grain during granary storage. *Cereal Chem.*, 57: 346-351.

Acevedo, E. & Bressani, R. 1990. Contenido de fibra dietética de alimentos Centroamericanos. Guatemala. *Arch. Latinoam. Nutr.*, 40: 439-451.

Adeniji, A.O. & Potter, N.N. 1978. Properties of ogi powders made from normal, fortified and opaque-2 corn. *J. Food Sci.*, 43: 1571-1574.

Aguirre, F., Bressani, R. & Scrimshaw, N.S. 1953. The nutritive value of Central American corns. III. Tryptophan, niacin, thiamine and riboflavin content of twenty-three varieties in Guatemala. *Food Res.*, 18: 273-279.

Akingbala, J.O., Onochie, E.V., Adeyemi, I.A. & Oguntimein, G.B. 1987. Steeping of whole and dry milled maize kernels in ogi preparation. *J. Food Proc. Preserv.*, 11: 1-11.

Akingbala, J.O., Rooney, L.W. & Faubion, J.M. 1981. A laboratory procedure for the preparation of ogi, a Nigerian fermented food. *J. Food Sci.*, 46: 1523-1526.

Akinrele, I.A. 1970. Fermentation studies on maize during the preparation of a traditional African starch-cake food. *J. Sci. Food Agric.*, 21: 619-625.

Akinrele, I.A. & Bassir, O. 1967. The nutritive value of ogi, a Nigerian infant food. *J. Trop. Med. Hyg.*, 70: 279-280.

Amorin, P.J. de. 1972. Opaque-2 maize for feeding infants. *Pediatr. Prat.* 43: 235-244.

Arroyave, G. 1974. Amino acid requirements by age and sex. *In* P.L. White and D.C. Fletcher, eds. *Nutrients in processed food. Proteins*, p. 15-28. Acton, Mass., USA, Publishing Sciences Group Inc.

Arroyave, G., Wilson, D., Behar, M. & Viteri, F. 1961. The development of INCAP vegetable mixtures II. Biochemical testing. In *Progress in meeting protein needs of infants and preschool children.* NRC Pub. 843, p. 49-55. Washington, D.C., Natl. Acad. Sci., Natl. Res. Counc.

Asp, N.G., Johansson, C.G., Hallmer, H. & Siljestrom, M. 1983. Rapid enzymatic assay of insoluble and soluble dietary fiber. *J. Agric. Food Chem.*, 33: 476-482.

Banigo, E.O.I., de Man, J.M. & Duitschaever, C.L. 1974. Utilization of high-lysine corn for the manufacture of ogi using a new, improved processing system. *Cereal Chem.*, 51: 559-572.

Banigo, E.O.I. & Muller, H.G. 1972. Carboxylic acid patterns in ogi fermentation. *J. Sci. Food Agric.*, 23: 101-111.

Barber, S.A. 1979. Corn residue management and soil organic matter. *Agron. J.*, 71: 625-627.

Baptist, N.G. & de Mel, B.V. 1955. Growth and amino acid intakes of children on a cereal-legume vegetable diet. *Br. J. Nutr.*, 9: 156-170.

Bazua, C.D., Guerra, R. & Sterner, H. 1979. Extruded corn flour as an alternative to lime-treated corn flour for tortilla preparation. *J. Food Sci.*, 44: 940-941.

Bedolla, S., de Palacios, M.G., Rooney, L.W., Ciehl, K.C. & Khan, M.N. 1983. Cooking characteristics of sorghum and corn for tortilla preparation by several cooking methods. *Cereal Chem.*, 60: 263-268.

Bedolla, S. & Rooney, L.W. 1982. Cooking maize for masa production. *Cereal Foods World*, 27: 219-221.

Bedolla, S. & Rooney, L.W. 1984. Characteristics of US and Mexican instant maize flours for tortilla and snack preparations. *Cereal Foods World*, 29: 732-735.

Benton, D.A., Harper, A.E. & Elvehjem, C.A. 1955. Effect of isoleucine supplementation on the growth of rats fed zein or corn diets. *Arch. Biochem. Biophys.*, 57: 13-19.

Benton, D.A., Harper, A.E., Spivey, H.E. & Elvehjem, C.A. 1956. Leucine, isoleucine and valine relationships in the rat. *Arch. Biochem. Biophys.*, 60: 147-155.

Boyer, C.D. & Shannon, J.C. 1983. The use of endosperm genes for sweet corn improvement. *In* J. Janick, ed. *Plant breeding reviews*, Vol. 1, p. 139-161. Westport, Conn., USA, AVI Publishing Co.

Boyer, C.D. & Shannon, J.C. 1987. Carbohydrates of the kernel. *In* S.A. Watson & P.E. Ramstad, eds. *Corn: chemistry and technology*, p. 253-272. St Paul, Minn., USA, Am. Assoc. Cereal Chem.

Braham, J.E. & Bressani, R. 1966. Utilization of calcium from home treated maize. *Nutr. Bromatol. Toxicol.*, 5: 14-19.

Braham, J.E., Bressani, R. & Guzmán, M.A. 1966. Effect of lysine and tryptophan supplementation on nicotinic acid metabolism in pigs given raw or lime-treated corn diets. *Metabolism*, 15: 548-556.

Braham, J.E., Villareal, A. & Bressani, R. 1962. Effect of lime treatment of corn on the availability of niacin for cats. *J. Nutr.*, 76: 183-186.

Bressani, R. 1962. Effect of amino acid imbalance on nitrogen retention. I. Effect of a relative deficiency of tryptophan in dogs. *J. Nutr.*, 78: 365-370.

Bressani, R. 1963. Effect of amino acid imbalance on nitrogen retention. II. Interrelationships between methionine, valine, isoleucine and threonine as supplements to corn proteins for dogs. *J. Nutr.*, 79: 389-394.

Bressani, R. 1971. Amino acid supplementation of cereal grain flours tested in children. *In* N.S. Schrimshaw & A.M. Altschul, eds. *Amino acid fortification of protein foods*, p. 184-204. Cambridge, Mass., USA, MIT Press.

Bressani, R. 1972. The importance of maize for human nutrition in Latin America and other countries. In *Nutritional improvement of maize.* INCAP publication L-4, p. 5-29. Guatemala, INCAP.

Bressani, R. 1983. *World needs for improved nutrition and the role of vegetables and legumes.* Asian Vegetable Research and Development Center, 10th Anniversary Monograph Series. AVRDC Pub. 83-185. Shanhua, Taiwan, AVRDC.

Bressani, R. 1987. The incorporation of nutritional goals in agricultural research in developing countries. *Benson Inst. Rev.*, 10: 18-28.

Bressani, R. 1988. Protein complementation of foods. *In* E. Karmas & R.S. Harris, eds. *Nutritional evaluation of food processing*, 3rd ed., p. 627-657. New York, Van Nostrand Reinhold Company.

Bressani, R. 1990. Chemistry, technology and nutritive value of maize tortillas. *Food Rev. Int.*, 6: 225-264.

Bressani, R., Alvarado, J. & Viteri, F. 1969. Evaluación en niños de la calidad de la proteína del maíz opaco-2. *Arch. Latinoam. Nutr.*, 19: 129-140.

Bressani, R., Arroyave, G. & Scrimshaw, N.S. 1953. The nutritive value of Central American corns. I. Nitrogen, ether extract, crude fiber and minerals of twenty-four varieties in Guatemala. *Food Res.*, 18: 261-267.

Bressani, R., Benavides, V., Acevedo, E. & Ortiz, M.A. 1990. Changes in selected nutrient content and in protein quality of common and quality protein maize during tortilla preparation. *Cereal Chem.*, 67(6): 515-518.

Bressani, R., Braham, J.E., Elías, L.G. & Rubio, M. 1979. Further studies on the enrichment of lime-treated corn with whole soybeans. *J. Food Sci.*, 44: 1707-1710.

Bressani, R., Breuner, M. & Ortiz, M.A. 1989. Contenido de fibra ácido- y neutro-detergente y de minerales menores en maíz y su tortilla. *Arch. Latinoam. Nutr.*, 39: 382-391.

Bressani, R., Castillo, S.V. & Guzmán, M.A. 1962. The nutritional evaluation of processed whole corn flour. *J. Agric. Food Chem.*, 10: 308-312.

Bressani, R. & de Villareal, E.M. 1963. Nitrogen balance of dogs fed lime-treated corn supplemented with proteins and amino acids. *J. Food Sci.*, 28: 611-615.

Bressani, R. & Elías, L.G. 1969. Studies on the use of opaque-2 corn in vegetable protein-rich foods. *J. Agric. Food Chem.*, 17: 659-662.

Bressani, R. & Elías, L.G. 1972. La calidad proteínica del maíz opaco-2 como ingrediente de dietas rurales de Guatemala. *Arch. Latinoam. Nutr.*, 22: 577-594.

Bressani, R. & Elías, L.G. 1974. Legume foods. *In* A.M. Altschul, ed. *New protein foods*, Vol. 1A, p. 231-297. New York, Academic Press Inc.

Bressani, R. & Elías, L.G. 1981. Estudios nutricionales sobre dietas. Valor proteínico de mezclas de cereales. *INCAP Informe Anual 1980*, p. 10-11. Managua, Nicaragua, INCAP.

Bressani, R., Elías, L.G. & Braham, J.E. 1968. Suplementación con aminoácidos del maíz y de la tortilla. *Arch. Latinoam. Nutr.*, 18: 123-134.

Bressani, R., Elías, L.G. & Braham, J.E. 1978. Improvement of the protein quality of corn with soybean protein. *Adv. Exp. Med. Biol.*, 105: 29-65.

Bressani, R., Elías, L.G. & de España, M.E. 1981. Posibles relaciones entre medidas físicas, químicas y nutricionales en frijol común (*Phaseolus vulgaris*). *Arch. Latinoam. Nutr.*, 31: 550-570.

Bressani R., Elías, L.G., Santos, M., Navarrete, D. & Scrimshaw, N.S. 1960. El contenido de nitrógeno y de aminoácidos esenciales de diversas selecciones de maíz. *Arch. Venez. Nutr.*, 10: 85-100.

Bressani, R., Gómez-Brenes, R. & Scrimshaw, N.S. 1961. Effect of processing on distribution and in vitro availability of niacin of corn (*Zea mays*). *Food Technol.*, 15: 450-454.

Bressani, R., Hernandez, E., Colon, A., Wolzak, A. & Gómez-Brenes, R. 1981. Efecto suplementario de 3 fuentes de proteína de soya sobre diferentes selecciones o productos de maíz. *Arch. Latinoam. Nutr.*, 31: 52-62.

Bressani, R. & Marenco, E. 1963. Corn flour supplementation: the enrichment of lime-treated corn flour with proteins, lysine and tryptophan and vitamins. *J. Agric. Food Chem.*, 11: 517-522.

Bressani, R., Medrano, J.F., Elías, L.G., Gómez-Brenes, R., González, J.M., Navarrete, D. & Klein, R.E. 1982. Estudios de control de insectos para la preservación del maíz opaco-2 almacenado y efectos sobre su valor nutritivo. *Turrialba*, 32: 51-58.

Bressani, R. & Mertz, E.T. 1958. Studies on corn protein. IV. Protein and amino acid content of different corn varieties. *Cereal Chem.*, 35: 227-235.

Bressani, R., Murillo, B. & Elías, L.G. 1974. Whole soybeans as a means of increasing protein and calories in maize-based diets. *J. Food Sci.*, 39: 577-580.

Bressani, R., Paz y Paz, R. & Scrimshaw, N.S. 1958. Chemical changes in corn during preparation of tortillas. *J. Agric. Food Chem.*, 6: 770-774.

Bressani, R. & Scrimshaw, N.S. 1958. Effect of lime treatment on in-vitro availability of essential amino acids and solubility of protein fractions in corn. *J. Agric. Food Chem.*, 6: 774-778.

Bressani, R. & Scrimshaw, N.S. 1961. The development of INCAP vegetable mixtures. 1. Basic animal studies. In *Progress in meeting protein needs of infants and preschool children*. NRC Pub. 843, p. 35-48. Washington, D.C., Natl. Acad. Sci., Natl. Res. Counc.

Bressani, R., Scrimshaw, N.S., Béhar, M. & Viteri, F. 1958. Supplementation of cereal proteins with amino acids. II. Effect of amino acid supplementation of corn-masa at intermediate levels of protein intake on the nitrogen retention of young children. *J. Nutr.*, 66: 501-513.

Bressani, R., Valiente, A.T. & Tejada, C. 1962. All vegetable protein mixtures for human feeding. VI. The value of combinations of lime-treated corn and cooked black beans. *J. Food Sci.*, 27: 394-400.

Bressani, R., Wilson, D., Chung, M., Béhar, M. & Viteri, F. 1963. Supplementation of cereal proteins with amino acids. V. Effect of supplementing lime-treated corn with different levels of lysine, tryptophan and isoleucine on the nitrogen retention of young children. *J. Nutr.*, 80: 80-84.

Brock, J.F. 1961. Protein malnutrition, requirements and supplementation. In *Progress in meeting protein needs of infants and preschool children.* NRC Pub. 843, p. 103-118. Washington D.C., Natl. Acad. Sci., Natl. Res. Counc.

Burge, R.M. & Duensing, W.J. 1989. Processing and dietary fiber ingredient applications of corn bran. *Cereal Foods World*, 34: 535-538.

Camacho, L., Bañados, E. & Fernandez, E. 1989. Enlatado de humitas preparados de maíz opaco-2 complementando con lupino dulce (*Lupinus albus* var. *Multolupa*), cambios nutricionales y de calidad. *Arch. Latinoam. Nutr.*, 39(2): 185-199.

Capparelli, E. & Mata, L. 1975. Microflora of maize prepared as tortillas. *Appl. Microbiol.*, 29: 802-806.

Carvajal, M., Rosiles, R., Abbas, H.K. & Moricha, C.J. 1987. Mycotoxin carryover from grain to tortillas in Mexico. *In* M.S. Zuber, E.B. Lillehoj & B.L. Renpro, eds. *Aflatoxin in maize, proceedings of the workshop,* p. 318-319. El Batan, Mexico, CIMMYT.

Chakrabarti, A.G. 1981. Detoxification of corn. *J. Food Prot.*, 44: 591-592.

Chavez, A. 1973. El maíz en la nutrición de México. In *Simposio sobre desarrollo y utilización de maíces de alto valor nutritivo, junio 29-30, 1972, Colegio de Postgraduados, ENA*, p. 9-11. Chapingo, México, SAG.

Chavez, J.F. 1972a. Composición del maíz opaco-2 venezolano. Análisis y calidad biológica de la arepa de opaco-2 y de maíz corriente. *Arch. Latinoam. Nutr.*, 22: 147-160.

Chavez, J.F. 1972b. Calidad nutritiva de la proteína de la harina de arepa y su mejoramiento por medio de la fortificación en Venezuela. *In* R. Bressani, J.E. Braham & M. Béhar, eds. *Mejoramiento nutricional del maíz.* INCAP Pub. L-3, p. 116-125. Guatemala, INCAP.

Chavez, J.F. & Obregon, G.P. 1986. Composición y valor nutritivo del maíz dulce Pajimaca y del Pajimaca opaco-2 cultivados en Venezuela. *Arch. Latinoam. Nutr.*, 36: 312-318.

Christensen, C.M. 1967. Some changes in No. 2 corn stored two years at moisture contents of 14.5% and 15.2%, and temperatures of 12°C, 20°C, and 25°C. *Cereal Chem.*, 44: 95-99.

Christensen, C.M. & Sauer, D.B. 1982. Microflora. *In* C.M. Christensen, ed. *Storage of cereal grains and their products*, 2nd ed., p. 219-240. St Paul, Minn., USA, Am. Assoc. Cereal Chem.

Christianson, D.D., Wall, J.S., Dimler, R.J. and Booth, A.N. 1968. Nutritionally available niacin in corn. Isolation and biological activity. *J. Agric. Food Chem.*, 16: 100-104.

Chu, N.T., Pellet, P.L. & Nawar, W.W. 1976. Effect of alkali treatment on the formation of lysinoalanine in corn. *J. Agric. Chem.*, 24: 1084-1085.

Clark, H.E., Allen, P.E., Meyers, S.M., Tuckett, S.E. & Yamamura, Y. 1967. Nitrogen balances of adults consuming opaque-2 maize protein. *Am. J. Clin. Nutr.*, 20: 825-833.

Clark, H.E., Glover, D.V., Betz, J.L. & Bailey, L.B. 1977. Nitrogen retention of young men who consumed isoitrogenous diets containing normal, opaque-2 or sugary-2 opaque-2 corn. *J. Nutr.*, 107: 404-411.

Contreras, G., Elías, L.G. & Bressani, R. 1980. Limitations of corn (*Zea mays*) and common bean (*Phaseolus vulgaris*) diets as protein and calorie sources. *Plant Foods Human Nutr.*, 30: 145-153.

Contreras, G., Elías, L.G. & Bressani, R. 1981. Efecto de la suplementación con vitaminas y minerales sobre la utilización de la proteína de mezclas de maíz y frijol. *Arch. Latinoam. Nutr.*, 31: 808-826.

Conway, H.F. & Anderson, R.A. 1978. Detoxification of aflatoxin contaminated corn by roasting. *Cereal Chem.*, 55: 115-117.

Cortez, A. & Wild-Altamirano, C. 1972. Contributions to the lime-treated corn flour technology. *In* R. Bressani, J.E. Braham & M. Béhar, eds. *Nutritional improvement of maize.* INCAP Pub. L-4, p. 99-106. Guatemala, INCAP.

Cox, L.J., Caicedo, B., Vanos, V., Heck, E., Hofstaelter, S. & Cordier, J.L. 1987. A catalogue of some Ecuadorean fermented beverages, with notes on their microflora. *Mircen J.*, 3: 143-153.

Cravioto, R.O., Anderson, R.K., Lockhart, E.E., De Miranda, F. de P. & Harris, R.S. 1945. Nutritive value of the Mexican tortilla.

Science, 102: 91.

Cravioto, R.O., Cravioto, O.Y., Massieu, G. & Guzman, G.J. 1955. El "pozol", forma indígena de consumir el maíz en el Sur-Este de México y su aporte de nutrientes a la dieta. *Ciencia (Mex.)*, 15: 27-30.

Cravioto, R.O., Massieu, G.H., Cravioto, O.Y. & Figueroa, M. 1952. Effect of untreated corn and Mexican tortilla upon the growth of rats on a niacin-tryptophan deficient diet. *J. Nutr.*, 48: 453-459.

Creech, R.G. 1965. Genetic control of carbohydrate synthesis in maize endosperm. *Genetics*, 52: 1175-1186

Cuevas, R., de Padua, M.R., Paredes, G. & Ortega, C. 1985. Precooked corn flour for Venezuelan arepa preparation: characterization of particle size fractions. *J. Food Sci.*, 50: 1668-1671.

Cuevas, R., Figueroa, E. & Racca, E. 1985. The technology for industrial production of precooked corn flours in Venezuela. *Cereal Foods World*, 30: 707-712.

Daniel, V.A., Rajan, P., Sanjeevarayappa, K.V., Srinivasan, K.S. & Swaminathan, M. 1977. Effect of insect infestation on the chemical composition and protein efficiency ratio of the proteins of kaffir corn and green gram. *Indian J. Nutr. Diet*, 14: 38-42.

de Arriolo, M.C., de Porres, E., de Cabrera, S., de Zepeda, M. & Rolz, C. 1987. Aflatoxin and tortilla preparation in Guatemala. *In* M.S. Zuber, E.B. Lillehoj & B.L. Renpro, eds. *Aflatoxin in maize, proceedings of the workshop*, p. 298-307. El Batan, Mexico, CIMMYT.

de Arriola, M.C., de Porres, E., de Cabrera, S., de Zepeda, M. & Rolz, C. 1988. Aflatoxin fate during alkaline cooking of corn for tortilla preparation. *J. Agric. Food Chem.*, 36: 530-533.

De Buckle, T.S., Pardo, C.A., de Sandoval, A.M. & Silva, A. 1972. Propiedades funcionales de harina de maíz y fortificada para la preparación de arepa. *In* R. Bressani, J.E. Braham & M. Béhar, eds. *Nutritional improvement of maize*. INCAP Pub. L-4, p. 131-146. Guatemala, INCAP.

de Campos, M., Crespo-Santos, J. & Olszyna-Marzys, A.E. 1980. Aflatoxin contamination in grains from the Pacific coast in Guatemala and the effect of storage upon contamination. *Bull. Environ. Contam. Toxicol.*, 24: 789-795.

De Groot, A.P. & Slump, P. 1969. Effect of severe alkali treatment of proteins on amino acid composition and nutritive value. *J. Nutr.*, 98: 45-46.

Del Valle, F.R. 1972. Industrial production, distribution and marketing of maize flour for tortilla in Mexico. *In* R. Bressani, J.E. Braham & M. Béhar, eds. *Nutritional improvement of maize*. INCAP Pub. L-4, p. 57-83. Guatemala, INCAP.

Del Valle, F.R., Montemayor, E. & Bourges, H. 1976. Industrial fortification of corn tortillas with oilseed flour by lime cooking of whole raw corn/soybean mixtures. *J. Food Sci.*, 41: 349-351.

Del Valle, F.R. & Pérez-Villasenor, J. 1974. Enrichment of tortillas with soy proteins by lime cooking of whole raw soybean mixtures. *J. Food Sci.*, 39: 244-247.

Deschamps, A.I. 1985. Aprovechamiento industrial del maíz en la manufactura de productos alternos a los de panificación originados en el trigo. *Congreso Tecnología Alimentos*, Viña del Mar, Chile.

de Souza, N., Elías, L.G. & Bressani, R. 1970. Estudios en ratas del efecto de una dieta básica del medio rural de Guatemala suplementada con leche de vaca y una mezcla de proteínas. *Arch. Latinoam. Nutr.*, 20: 293-307.

Dudley, J.W. & Lambert, R.J. 1969. Genetic variability after 65 generations of selection in Illinois high oil, low oil, high protein and low protein strains of *Zea mays* L. *Crop Sci.*, 9: 179-181.

Dudley, J.W., Lambert, R.J. & Alexander, D.E. 1974. Seventy generations of selections for oil and protein concentration in the maize kernel. *In* J.W. Dudley, ed. *Seventy generations of selection for oil and protein in maize*, p. 181-211. Madison, Wis., USA, Crop. Sci. Soc. Am.

Dudley, J.W., Lambert, R.J. & de la Roche, I.A. 1977. Genetic analysis of crosses among corn strains divergently selected for percent oil and protein. *Crop. Sci.*, 17: 111-117.

Earll, L., Earll, J.M., Navjokaitis, S., Pyle, S., McFalls, K. & Altschul, A.M. 1988. Feasibility and metabolic effects of a purified corn fiber food supplement. *J. Am. Diet. Assoc.*, 88: 950-952.

Eggert, R.G., Brinegar, M.J. & Anderson, C.R. 1953. The quality of protein of normal and high protein corn for growing swine. *J. Anim. Sci.*, 12: 282-290.

Elías, L.G. & Bressani, R. 1971. Improvement of the protein quality of corn-bean diets by the use of fortified corn or opaque-2 corn. In *Third Western Hemisphere Nutrition Congress*, Bal Harbour, Fla., USA.

FAO. 1966. *Food balance sheets, 1960-62 average.* Rome, FAO.

FAO. 1984. *Food balance sheets, 1979-81 average.* Rome, FAO.

FAO. 1988. *FAO Production Yearbook (1987).* Vol. 41. Rome, FAO.

FAO. 1989. *Utilization of tropical foods: cereals.* Food and Nutrition Paper 47/1. Rome, FAO.

Fennell, D.I., Lillehoj, E.B., Kwolk, W.F., Guthrie, W.D., Sheeley, R., Sparks, A.N., Widstrom, N.W. & Adams, G.L. 1978. Insect larval activity on developing corn ears and subsequent aflatoxin contamination of seed. *Econ. Entomol.*, 71: 624.

Flores, M., Bressani, R. & Elías, L.G. 1973. Factors and tactics influencing consumer food habits and patterns. In *Potential of field beans and other food legumes in Latin America*, p. 88-114. Cali, Colombia, CIAT.

Flynn, L.M., Zuber, M.S., Leweke, D.H., Grainger, R.B. & Hogan, A.G. 1954. Relationship between protein content of corn and concentration of amino acids and nicotinic acid. *Cereal Chem.*, 31: 217-228.

Frenk, S. 1961. Biological value of some new sources of protein in Mexican malnourished children. In *Progress in meeting protein needs of infants and preschool children.* NRC Pub. 843, p. 21-38. Washington, D.C., Natl. Acad. Sci., Natl. Res. Counc.

Frey, K.J. 1951. The interrelationships of proteins and amino acids in corn. *Cereal Chem.*, 28: 123-132.

Frey, K.J., Brimhall, B. & Sprague, G.F. 1949. The effects of selection upon protein quality in the corn kernel. *Agron. J.*, 41: 399-403.

Galinat, W.C. 1977. The origin of corn. *In* G.F. Sprague, ed. *Corn and corn improvement*, p. 1-47. Madison, Wis., USA, Am. Soc. Agron.

García, B. & Urrutia, J.J. 1978. Descripciones de las condiciones socioeconómicas de la comunidad de Santa María Cauqué. In *Tecnología de Alimentos y Nutrición. Memorias Conferencia Interacción entre Producción Agrícola*, p. 116-132. Guatemala, INCAP.

Gómez, M.H., McDonough, C.M., Rooney, L.W. & Waniska, R.D. 1989. Changes in corn and sorghum during nixtamalization and tortilla baking. *J. Food Sci.*, 54: 330-336.

Gómez, F., Ramos-Galvan, R., Cravioto, J., Frenk, S., de la Peña, C., Moreno, M.E. & Villa, M.E. 1957. Protein metabolism in chronic severe malnutrition (kwashiorkor). Absorption and retention of nitrogen from a typical poor Mexican diet. *Br. J. Nutr.*, 11: 229-233.

Gómez-Brenes, R.A., Elías, L.G. & Bressani, R. 1968. Efecto del proceso de maduración del maíz sobre su valor nutritivo. *Arch. Latinoam. Nutr.*, 18: 65-79.

Gómez-Brenes, R.A., Elías, L.G. & Bressani, R. 1972. Improvement of the protein quality of diets of low nutritive value through the use of fortified maize and opaque-2 maize. *In* R. Bressani, J.E. Braham & M. Béhar, eds. *Nutritional improvement of maize.* INCAP Pub. L-4, p. 256-272. Guatemala, INCAP.

Gopalan, C. & Rao, K.S.J. 1975. Pellagra and amino acid imbalance. *Vitam. Horm.*, 33: 505-528.

Graham, G.G., Glover, D.V., de Romana, G.L., Morales, E. & MacLean, W.C. Jr. 1980. Nutritional value of normal, opaque-2 and sugary-2 opaque-2 maize hybrids for infants and children. I. Digestibility and utilization. *J. Nutr.*, 110: 1061-1069.

Graham, G.G., Lembcke, J., Lancho, E. & Morales, E. 1989. Quality protein maize: digestibility and utilization by recovering malnourished infants. *Pediatrics*, 83: 416-421.

Graham, G.G., Placko, R.P. & MacLean, W.C. Jr. 1980. Nutritional value of normal, opaque-2 and sugary-2 opaque-2 maize hybrids for infants and children. 2. Plasma-free amino acids. *J. Nutr.*, 110: 1070-1075.

Hansen, J.D.L. 1961. The effects of various forms of supplementation on the nutritive value of maize for children. In *Progress in meeting protein needs of infants and preschool children.* NRC Pub. 843, p. 89-102. Washington, D.C., USA, Natl. Acad. Sci., Natl. Res. Counc.

Hansen, J.D.L., Schendel, H.E., Wilkins, A. & Brock, J.F. 1960. Nitrogen metabolism in children with kwashiorkor receiving milk and vegetable diets. *Pediatrics*, 25: 258-282.

Harper, A.E., Benton, D.A. & Elvehjem, C.A. 1955. L-leucine, an isoleucine antagonist in the rat. *Arch. Biochem. Biophys.*, 57: 1-12.

Hazell, T. & Johnson, I.T. 1989. Influence of food processing on iron availability in vitro from extruded maize-based snack foods. *J. Sci. Food Agric.*, 46: 365-374.

Herum, F.L. 1987. Harvesting and postharvest management. *In* S.A. Watson & P.E. Ramstad, eds. *Corn, chemistry and technology*, p. 83-123. St Paul, Minn., USA, Am. Assoc. Cereal Chem.

Hesseltine, C.W. 1979. Some important fermented foods of mid-Asia, the Middle East and Africa. *J. Am. Oil*

Chem. Soc., 56: 367-374.

Hickey, C.S., Stephens, D.O. & Flowers, R.S. 1982. Preservation of flour tortillas. Paper No. 221, presented at the 42nd Annual Meeting, Inst. Food Technol., Las Vegas, Nev., USA.

Hogan, A.G., Gillespie, G.T., Kocturk, O., O'Dell, B.L. & Flynn, L.M. 1955. The percentage of protein in corn and its nutritional properties. *J. Nutr.*, 57: 225-239.

Howe, E.E., Janson, G.R. & Gilfillan, E.W. 1965. Amino acid supplementation of cereal grains as related to the world food supply. *Am. J. Clin. Nutr.*, 16: 315-320.

Illescas, R. 1943. La teoría química de la formación del nixtamal. *Rev. Soc. Mex. Hist. Nat.*, 4: 129.

Ingle, J., Bietz, D. & Hageman, R.H. 1965. Changes in composition during development and maturation of maize seed. *Plant Physiol.*, 40: 835-839.

Institute of Nutrition of Central America and Panama (INCAP). 1969. *Evaluación nutricional de la población de Centro América y Panamá.* Guatemala, INCAP; Oficina de investigaciones internacionales de los institutos nacionales de salud; Ministerio de salubridad pública. 144 pp.

Instituto de investigaciones tecnológicas. 1971. Harinas precocidas de cereales. Primera fase. *Informe preparado para la OEA (Abril)*, p. 27-39. Bogotá, Colombia, IIT.

Islam, M.N., Lirio, M.E. & Delvalle, F.R. 1984. Mold inhibition in tortilla by dimethyl fumarate. *J. Food Proc. Preserv.*, 8: 41-45.

Jackson, D.S., Rooney, L.W., Kunze, O.R. & Waniska, R.D. 1988. Alkaline processing. Properties of stress-cracked and broken corn (*Zea mays*). *Cereal Chem.*, 65: 133-137.

Jellum, M.D. & Marion, J.E. 1966. Factors affecting oil content and oil composition of corn (*Zea mays*) grain. *Crop. Sci.*, 6: 41-42.

Johnson, B.A., Rooney, L.W. & Khan, M.N. 1980. Tortilla-making characteristics of micronized sorghum and corn flours. *J. Food Sci.*, 45: 671-674.

Khan, M.N., Desrosiers, M.C., Rooney, L.W., Morgan, R.G. & Sweat, V.E. 1982. Corn tortillas: evaluation of corn cooking procedures. *Cereal Chem.*, 59: 279-284.

Khan, N.H. & Bressani, R. 1987. Preparation and nutritional quality of high protein food extracts from immature corn, whole soybean and

dry whole milk. *Plant Food Human Nutr.*, 37: 141-149.

Kies, C., Williams, E. & Fox, H.M. 1965. Determination of first limiting nitrogenous factor in corn protein for nitrogen retention in human adults. *J. Nutr.*, 86: 350-356.

Krause, V.M. 1988. *Rural-urban variations in limed maize consumption and the mineral content of tortilla in Guatemala.* Center for Studies of Sensory Impairment, Aging, and Metabolism, Guatemala; School of Dietetics and Human Nutrition, McGill University, Montreal, Canada.

Landry, J. & Moureaux, T. 1970. Hétérogénéité des glutélines du grain de maïs: Extraction sélective et composition en acides aminés des trois fractions isolées. *Bull. Soc. Chim. Biol.*, 52: 1021-1037.

Landry, J. & Moureaux, T. 1980. Distribution and amino acid composition of protein groups located in different histological parts of maize grain. *J. Agric. Food Chem.*, 28: 1186-1191.

Landry, J. & Moureaux, T. 1982. Distribution and amino acid composition of protein fractions in opaque-2 maize grain. *Phytochemistry*, 21: 1865-1869.

Leibovits, Z. & Ruckenstein, C. 1983. Our experiences in processing maize (corn) germ oil. *J. Am. Chem. Soc.*, 60: 395-399.

Luna-Jaspe, G.H., Parra, J.O.M. & Serrano, S.P. 1971. Comparación de la retención de nitrógeno en niños alimentados con maíz común, maíz de gene opaco-2 y leche de vaca. I. Resultados con baja ingesta de proteína. *Arch. Latinoam. Nutr.*, 21: 437-447.

Lunven, P. 1968. *Le tryptophane dans l'alimentation intertropicale.* Ph.D. thesis, University of Paris.

Maciejewicz-Rys, J. & Hanczakowski, P. 1989. Improvement of the nutritive value of cereals by leaf protein supplementation. *J. Sci. Food Agric.*, 50: 99-104.

Mangelsdorf, P.C. & Reeves, R.G. 1939. The origin of Indian corn and its relatives. *Bulletin No. 574, Texas Agric. Exp. Stn.*, College Station, Texas, USA.

Martínez, A.B., Gómez-Brenes, R.A. & Bressani, R. 1980. Relación del contenido de lisina y triptofano con el de zeina, durante la germinación del grano de maíz y su posible vinculación con el ciclo vegetativo de la planta. *Arch. Latinoam. Nutr.*, 30: 607-633.

Martínez, M.L., Elías, L.G., Rodriguez, J.F., Jarquín, R. &

Bressani, R. 1970a. Valor nutritivo del maíz infectado con hongos en pollos y de tortilla de maíz fungoso en ratas. *Arch. Latinoam. Nutr.*, 20: 217-240.

Martínez, M.L., Schieber, E., Gómez-Brenes, R. & Bressani, R. 1970b. Prevalencia de hongos en granos de maíz (*Zea mays*, L.) de Guatemala. *Turrialba*, 20: 311-319.

Martínez, R.R. 1979. Las aflatoxinas en las tortillas. *Vet. Mex.*, 10: 37-44.

Martínez-Herrera, M.L. 1968. *Efecto de ciertos hongos sobre el valor nutritivo, calidad y conservación del maíz en Guatemala.* Thesis, Universidad de San Carlos de Guatemala.

Martínez-Herrera, M.L. & Lachance, P.A. 1979. Corn (*Zea mays*) kernel hardness as an index of the alkaline cooking time for tortilla preparation. *J. Food Sci.*, 44: 377-380.

Mertz, E.T., Bates, L.S. & Nelson, O.E. 1964. Mutant gene that changes protein composition and increases lysine content of maize endosperm. *Science*, 145: 279-280.

Mertz, E.T., Jambunathan, R., Villegas, E., Bauer, R. Kies, C., McGinnis, J. & Shenk, J.S. 1975. Use of small animals for evaluation of protein quality in cereals. In *High-quality protein maize*, CIMMYT-Purdue International Symposium on Protein Quality in Maize, 1972, El Batan, Mexico, p. 306-329. Stroudsburg, Pa., USA, Dowden Hutchinson & Ross.

Mitchell, H.H., Hamilton, T.S. & Beadles, J.R. 1952. The relationship between the protein content of corn and the nutritional value of the protein. *J. Nutr.*, 48: 461-476.

Mitchell, H.H. & Smuts, D.B. 1932. The amino acid deficiencies of beef, wheat, corn, oats and soybeans for growth in the white rat. *J. Biol. Chem.*, 95: 263-281.

Molina, M.R., Baten, M.A. & Bressani, R. 1978. Características microbiológicas de la masa y tortillas con y sin agregado de harina de soya y el efecto de clorinación. *INCAP Informe Anual 1977*, p. 39-42. Guatemala, INCAP.

Molina, M.R., Letona, M. & Bressani, R. 1977. Drum-drying for the improved production of instant tortilla flour. *J. Food Sci.*, 42: 1432-1434.

Mosqueda Suarez, A. 1954. La arepa criolla. *Arch. Venez. Nutr.*, 5: 407-423.

Murillo, B., Cabezas, M.T. & Bressani, R. 1974. Influencia de la densidad calórica sobre la utilización de la proteína en dietas elaboradas a base de maíz y frijol. *Arch. Latinoam. Nutr.*, 24: 223-241.

Navarrete, D.A. & Bressani, R. 1981. Protein digestibility and protein quality of common beans (*Phaseolus vulgaris*) fed alone and with maize, in adult humans using a short-term nitrogen balance assay. *Am. J. Clin. Nutr.*, 34: 1893-1898.

Nelson, O.E., Mertz, E.T. & Bates, L.S. 1965. Second mutant gene affecting the amino acid pattern of maize endosperm proteins. *Science*, 150: 1469-1470.

Norad, M.N., Iskander, F.Y., Rooney, L.W. & Erp, C.F. 1986. Physicochemical properties of alkali-cooked corn using traditional and presoaking procedures. *Cereal Chem.*, 63: 255-259.

Oke, O.L. 1967. Chemical studies on the Nigerian foodstuff "ogi". *Food Technol.*, 21: 202-204.

Onigbinde, A.O. & Akinyele, I.O. 1989. Association between chemical composition and protein digestibility of heat-damaged maize (*Zea mays*) flour. *Food Chem.*, 34: 35-39.

Orr, M.L. & Watt, B.K. 1957. *Amino acid content of foods.* Home Economics Research Report No. 4. Washington, D.C., USA, US Dept. Agric. 88 pp.

Ortega, E.I., Villegas, E. & Vasal, S.K. 1986. A comparative study of protein changes in normal and quality protein maize during tortilla making. *Cereal Chem.*, 63: 446-451.

Paredes-Lopez, O. & Saharopulos-Paredes, M.E. 1983. A review of tortilla production technology. *Baker's Dig.*, 57(5): 16-25.

Patterson, J.I., Brown, R.R., Linkswiler, H. & Harper, A.E. 1980. Excretion of tryptophan-niacin metabolites by young men: effects of tryptophan, leucine and vitamin B_6 intakes. *Am. J. Clin. Nutr.*, 33: 2157-2167.

Paulsen, M.R. & Hill, L.D. 1985. Corn quality factors affecting dry milling performance. *J. Agric. Eng. Res.*, 31: 255-263.

Pearson, W.N., Stempfel, S.J., Valenzuela, T.S., Utley, M.H. & Darby, W.J. 1957. The influence of cooked vs. raw maize on the growth of rats receiving a 9% casein ration. *J. Nutr.*, 62: 445-463.

Pelaez, J. & Karel, M. 1980. Development and stability of intermediate moisture tortillas. *J. Food Proc. Preserv.*, 4: 51-65.

Pérez, R.A., Tuite, J. & Baker, K. 1982. Effect of moisture, temperature and storage time on subsequent stability of shelled corn. *Cereal Chem.*, 59: 205-209.

Pflugfelder, R.L., Rooney, L.W. & Waniska, R.D. 1988a. Dry matter losses in commercial corn masa production. *Cereal Chem.*, 65: 127-132.

Pflugfelder, R.L., Rooney, L.W. & Waniska, R.D. 1988b. Fractionation and composition of commercial corn masa. *Cereal Chem.*, 65: 262-266.

Poey, F.R., Bressani, R., García, A.A., García, M.A. & Elías, L.G. 1979. Germ-endosperm relationship in the nutritional improvement of maize grain. In *Seed protein improvement in cereals and grain legumes*, Vol. 1, p. 369-384. Vienna, IAEA.

Pomeranz, Y., Czuchajowska, Z. & Lai, F.S. 1986. Comparison of methods for determination of hardness and breakage susceptibility of commerically dried corn. *Cereal Chem.*, 63: 39-43.

Pomeranz, Y., Czuchajowska, Z., Martin, C.R. & Lai, F.S. 1985. Determination of corn hardness by the Stenvert hardness tester. *Cereal Chem.*, 62: 108-112.

Pomeranz, Y., Martin, C.R., Traylor, D.D. & Lai, F.S. 1984. Corn hardness determination. *Cereal Chem.*, 61: 147-150.

Poneleit, C.G. & Alexander, D.E. 1965. Inheritance of linoleic and oleic acids in maize. *Science*, 147: 1585-1586.

Poneros, A.G. & Erdman, J.W. Jr. 1988. Bioavailability of calcium from tofu, tortillas, non-fat milk and mozzarella cheese in rats: effect of supplemental ascorbic acid. *J. Food Sci.*, 53: 208-210.

Pradilla, A., Linares, F., Francis, C.A. & Fajardo, L. 1973. El maíz de alta lisina en nutrición humana. In *Simposio sobre desarrollo y utilización de maíces de alto valor nutritivo, junio 29-30, 1971. Colegio de Postgraduados, ENA*, Chapingo, México, SAG.

Price, R.L. & Jorgensen, K.V. 1985. Effects of processing on aflatoxin levels and on mutagenic potential of tortillas made from naturally contaminated corn. *J. Food Sci.*, 50: 347-349.

Quackenbush, F.W. 1963. Corn carotenoids: effects of temperature and moisture on losses during storage. *Cereal Chem.*, 40: 266-269.

Rajan, P., Daniel, V.A., Padmarani, R. & Swaminathan, M. 1975. Effect of insect infestation on the protein efficiency ratio of the proteins of maize and cowpea. *Indian J. Nutr. Diet.*, 12: 354-357.

Ranhotra, G.S. 1985. Nutritional profile of corn and flour tortillas. *Cereal Foods World*, 30: 703-704.

Ranhotra, G. & Gelroth, J. 1988. Soluble and total dietary fiber in white bread. *Cereal Chem.*, 65: 155-156.

Reinhold, J.G. & Garcia J.S. 1979. Fibre of the maize tortilla. *Am. J. Clin. Nutr.*, 32: 1326-1329.

Robles, R.R., Murray, E.D. & Paredes-Lopez, O. 1988. Physico-chemical changes of maize starch during the lime-heat treatment for tortilla making. *J. Food Sci. Technol.*, 23: 91-98.

Robutti, J.L., Hoseny, R.C. & Deyoe, C.W. 1974. Modified opaque-2 corn endosperm. I. Protein distribution and amino acid composition. *Cereal Chem.*, 51: 163-172.

Robutti, J.L., Hoseny, R.C. & Wasson, C.E. 1974. Modified opaque-2 corn endosperm. II. Structure viewed with a scanning electron microscope. *Cereal Chem.*, 51: 173-180.

Rodriguez, O.F. 1972. Industrial production, market and distribution of maize flour for the preparation of arepa in Venezuela. *In* R. Bressani, J.E. Braham & M. Béhar, eds. *Nutritional improvement of maize.* INCAP Pub. L-4, p. 107-112. Guatemala, INCAP.

Rooney, L.W. & Serna-Saldivar, S.O. 1987. Food uses of whole corn and dry-milled fractions. *In* S.A. Watson & P.E. Ramsted, eds. *Corn: chemistry and technology.* St Paul., Minn., USA, Am. Assoc. Cereal Chem.

Rosenberg, H.R., Rohdenburg, E.L. & Eckert, R.E. 1960. Multiple amino acid supplementation of white corn meal. *J. Nutr.*, 72: 415-422.

Rubio, M.J. 1972a. Tortilla and process using epichlorohydrin. *US Patent 3,690, 893.*

Rubio, M.J. 1972b. Tortilla and process using carboxylic acids and their anhydrides. *US Patent 3,694,224.*

Rubio, M.J. 1973. Tortilla and process using hydrophylic inorganic gels. *US Patent 3,709,696.*

Rubio, M.J. 1974a. Tortilla and process using sorbic acid and its salts. *US Patent 3,853,997.*

Rubio, M.J. 1974b. Tortilla and process using methyl, ethyl, butyl and propyl esters of parahydroxy benzoic acid. *US Patent 3,853,998.*

Rubio, M.J. 1975. Tortilla and process using acetic and propionic acid. *US Patent 3,859,449.*

Saldana, G. & Brown, H.E. 1984. Nutritional composition of corn and flour tortillas. *J. Food Sci.*, 49: 1202-1203.

Sanderson, J., Wall, J.S., Donaldson, G.L. & Cavins, J.F. 1978. Effect of alkaline processing of corn on its amino acids. *Cereal Chem.*, 55: 204-213.

Sandstead, H.H., Munoz, J.M., Jacob, R.A., Kelvay, L.M., Reck, S.J., Logan, G.M. Jr., Dintzis, F.R.,

Inglett, G.E. & Shvey, W.C. 1978. Influence of dietary fiber on trace element balance. *Am. J. Clin. Nutr.*, 31: 5180-5184.

Sandstead, R.M., Hites, B.H. & Schroeder, H. 1968. Genetic variations in maize: effects on the properties of the starches. *Cereal Sci. Today*, 13: 82-94, 156.

Sauer, D.B. & Burroughs, R. 1980. Fungal growth, aflatoxin production and moisture equilibrium in mixtures of wet and dry corn. *Phytopathology*, 70: 516-521.

Schneider, K. 1987. *Experiencia del proyecto para la reducción de pérdidas postcosecha en Honduras.* Honduras, Ministerio de Recursos Naturales, Honduras y Cooperación Suiza al Desarrollo

Scrimshaw, N.S., Béhar, M., Wilson, D., Viteri, F., Arroyave, G. & Bressani, R. 1961. All-vegetable protein mixtures for human feeding. V. Clinical trials with INCAP mixtures 8 and 9 and with corn and beans. *Am. J. Clin. Nutr.*, 9: 196-205.

Scrimshaw, N.S., Bressani, R., Béhar, M. & Viteri, F. 1958. Supplementation of cereal proteins with amino acids. 1. Effect of amino acid supplementation of corn-masa at high levels of protein intake on the nitrogen retention of young children. *J. Nutr.*, 66: 485-499.

Serna-Saldivar, S.O., Canett, R., Vargas, J., Gonzalez, M., Bedolla, S. & Medina, C. 1988a. Effect of soybean and sesame addition on the nutritional value of maize and decorticated sorghum tortillas produced by extrusion cooking. *Cereal Chem.*, 65: 44-48.

Serna-Saldivar, S.O., Knabe, D.A., Rooney, L.W. & Tanksley, T.D. Jr. 1987. Effect of lime cooking on energy and protein digestibilities of maize and sorghum. *Cereal Chem.*, 64: 247-252.

Serna-Saldivar, S.O, Knabe, D.A., Rooney, L.W., Tanksley, T.D. Jr. & Sproule, A.M. 1988b. Nutritional value of sorghum and maize tortillas. *J. Cereal Sci.*, 7: 83-94.

Shannon, J.C. & Garwood, D.L. 1984. Genetics and physiology of starch development. *In* R.L. Whistler, J.N. Bemiller & E.F. Paschall, eds. *Starch, chemistry and technology*, p. 25-86. Orlando, Fla., Academic Press.

Siriacha, P., Kawashima, K., Kawasugi, S., Saito, M. & Tomboon-Ik, P. 1989. Post-harvest contamination of Thai corn with *Aspergillus flavus. Cereal Chem.*, 66: 445-448.

Solorzano-Mendizabal, M. de C. 1985. *Destrucción de aflatoxinas durante el proceso de nixtmalización.* Thesis, Universidad de San Carlos de Guatemala.

Sproule, A.M., Serna-Saldivar, S.O., Buckholt, A.J., Rooney, L.W. & Knabe, D.A. 1988. Nutritional evaluation of tortillas and tortilla chips from quality protein maize. *Cereal Foods World*, 33: 233-236.

Squibb, R.L., Bressani, R. & Scrimshaw, N.S. 1957. Nutritive value of Central American corns. V. Carotene content and vitamin A activity of three Guatamalan yellow corns. *Food Res.*, 22: 303-307.

Sternberg, M., Kim, C.Y. & Schwende, F.J. 1975. Lysino-alanine: presence in foods and food ingredients. *Science*, 190: 992-994.

Tanaka, A. & Yamaguchi, J. 1972. Dry matter production, yield components and grain yield of the maize plant. *J. Fac. Agric. Hokkaido Univ.*, 57: 71-132.

Torreblanca, A., Bourges, H. & Morales, J. 1987. Aflatoxin in maize and tortillas in Mexico. *In* M.S. Zuber, E.B. Lillehoj & B.L. Renpro, eds. *Aflatoxin in maize, proceedings of the workshop*, p. 310-317. El Batan, Mexico, CIMMYT.

Torún, B., Caballero, B., Flores-Huerta, S. & Viteri, F. 1984. Habitual Guatemalan diets and catch-up growth of children with mild to moderate malnutrition. *Food Nutr. Bull. Suppl.*, 10: 216-231.

Torún, B. & Viteri, F.E. 1981. Capacity of habitual Guatemalan diets to satisfy protein requirements of preschool children with adequate dietary energy intakes. *Food Nutr. Bull. Suppl.*, 5: 210-228.

Trejo-González, A., Feria-Morales, A. & Wild-Altamirano, C. 1982. The role of lime in the alkaline treatment of corn for tortilla preparation. *Adv. Chem. Ser.*, 198: 245-263.

Truswell, A.S. & Brock, J.F. 1961. Effect of amino acid supplements on the nutritive value of maize protein for human adults. *Am. J. Clin. Nutr.*, 9: 715-728.

Truswell, A.S. & Brock, J.F. 1962. The nutritive value of maize protein for man. *Am. J. Clin. Nutr.*, 10: 142.

Tsai, C.Y., Dalby, A. & Jones, R.A. 1975. Lysine and tryptophan increases during germination of maize seed. *Cereal Chem.*, 52: 356-360.

Tsai, C.Y., Huber, D.M. & Warren, H.L. 1978. Relationship of the kernel sink for N to maize productivity. *Crop. Sci.*, 18: 399-405.

Tsai, C.Y., Huber, D.M. & Warren, H.L. 1980. A proposed role of zein and glutelin as N sinks in maize. *Plant Physiol.*, 66: 330-333.

Tsai, C.Y., Warren, H.L., Huber, D.M. & Bressani, R.A. 1983. Interaction between the kernel N sink grain yield and protein nutritional quality of maize. *J. Sci. Food Agric.*, 34: 255-263.

Ulloa-Sosa, M. & Schroeder, H.W. 1969. Note on aflatoxin decomposition in the process of making tortillas from corn. *Cereal Chem.*, 46: 397-400.

Urrutia, J.J., Garcia, B., Bressani, R. & Mata, L.J. 1976. Report of the maize fortification. *In* H.L. Wilcke, ed. *Improving the nutrient quality of cereals. Report of 2nd Workshop on Breeding and Fortification*, Boulder, Colorado, USA, p. 28-68. Washington, D.C., USA, USAID.

Valverde V., Delgado, H., Belizan, J.M., Martorell, R., Mejia-Pivaral, V., Bressani, R., Elías, L.G., Molina, M.R. & Klein, R. 1983. The Patulul project: production, storage, acceptance and nutritional impact of opaque-2 corns in Guatemala. *Pub. INCAP*, p. 179. Guatemala, INCAP.

Valverde, V., Martorell, R., Delgado, H., Pivaral, V.M., Elías, L.G., Bressani, R. & Klein, R.E. 1981. The potential nutritional contribution of opaque-2 corn. *Nutr. Rep. Int.*, 23: 585-595.

Van Soest, P.J., Fadel, J. & Sniffen, C.J. 1979. Discount factors for energy and protein in ruminant feeds. In *Proceedings, Cornell nutrition conference for feed manufacturers*, p. 63-75. Ithaca, N.Y., USA, Cornell Univ.

Vargas, E., Muñoz, R. & Gómez, J. 1986. Composición química y valor biológico de tortillas y pan producidos a nivel industrial en Costa Rica. *Arch. Latinoam. Nutr.*, 36: 456-465.

Vineyard, M.L., Bear, R.P., MacMasters, M.M. & Deatherage, W.L. 1958. Development of "Amylo-maize" corn hybrid with high amylose starch: 1. Genetic considerations. *Agron. J.*, 50: 595-598.

Viteri, F., Martínez, C. & Bressani, R. 1972. Evaluación de la calidad proteínica del maíz común, del maíz opaco-2 y del maíz común suplementado con aminoácidos y otras fuentes de proteína. *In* R. Bressani, J.E. Braham & M. Béhar, eds. *Mejoramiento nutricional del maíz.* INCAP Pub. L-3, p. 195-208. Guatemala, INCAP.

Vivas, N.E., Waniska, R.D. & Rooney, L.W. 1987. Effect of tortilla production on proteins in sorghum and maize. *Cereal Chem.*, 64: 384-389.

Wang, Y.Y.D. & Fields, M.L. 1978. Germination of corn and sorghum in the home to improve nutritive value. *J. Food Sci.*, 43: 1113-1115.

Watson, S.A. 1962. The yellow carotenoid pigments of corn. *In* W. Hechendorn & J.J. Sutherland, eds. *Proc. Corn. Res. Conf., 17th.* Washington, D.C., USA, Am. Seed Trade Assoc.

Watson, S.A. 1987. Structure and composition. *In* S.A. Watson & P.E. Ramstad, eds. *Corn: chemistry and technology*, p. 53-82. St Paul, Minn., USA, Am. Assoc. Cereal Chem.

Weber, E.J. 1987. Lipids of the kernel. *In* S.A. Watson & P.E. Ramstad, eds. *Corn: chemistry and technology*, p. 311-349. St Paul, Minn., USA, Am. Assoc. Cereal Chem.

Wichser, W.R. 1966. Comparison of the dry milling properties of opaque-2 and normal dent corn. *In* E.T. Mertz & O.E. Nelson, eds. *Proc. High Lysine Corn Conference*, p. 104-116. Lafayette, Ind., USA, Purdue Univ.

Wolf, M.J., Buzan, C.L., MacMasters, M.M. & Rist, C.E. 1952. Structure of the mature corn kernel. *Cereal Chem.*, 29: 321-382.

Wolf, M.J., Khoo, V. & Seckinger, H.L. 1969. Distribution and subcellular structure of endosperm protein in varieties of ordinary and high-lysine maize. *Cereal Chem.*, 46: 253-263.

Wolzak, A., Bressani, R. & Gómez-Brenes, R. 1981. A comparison of in vivo and in vitro estimates of protein digestibility of native and thermally processed vegetable proteins. *Plant Foods Human Nutr.*, 31: 31-43.

Woodworth, C.M. & Jugenheimer, R.W. 1948. Breeding and genetics of high protein corn. *Ind. Res. Rep.*, 3: 75-83.

Yen, J.T., Jensen, A.H. & Baker, D.H. 1976. Assessment of the concentration of biologically available vitamin B_6 in corn and soybean meal. *J. Anim. Sci.*, 42: 866-870.

Young, V.R., Ozalp, I., Chokos, B.V. & Scrimshaw, N.S. 1971. Protein value of Colombian opaque-2 corn for young adult men. *J. Nutr.*, 101: 1475-1481.

Further reading

Adeyemi, I.A. 1983. Dry-milling of sorghum for ogi manufacture. *J. Cereal Sci.*, 1: L221-227.

Akingbala, J.O., Rooney, L.W. & Faubion, J.M. 1981. Physical, chemical and sensory evaluation of ogi from sorghum of differing kernel characteristics. *J. Food Sci.*, 46: 1532-1536.

Akinrele, I.A. 1964. Fermentation of cassava. *J. Sci. Food Agric.*, 15: 589-594.

Akinrele, I.A. & Edwards, C.C.A. 1971. An assessment of the nutritive value of maize-soya mixture, Soy-Ogi, as a weaning food in Nigeria. *Br. J. Nutr.*, 26: 177-185.

Andah, A. & Muller, H.G. 1973. Studies on koko, a Ghanaian fermented maize porridge. *Ghana J. Agric. Sci.*, 6: 93-102.

Bagley, E.B. 1979. Decontamination of corn containing aflatoxin by treatment with ammonia. *J. Am. Oil Chem. Soc.*, 56: 808-811.

Banigo, E.O.I. & Adeyemi, A.I. 1975. A comparative study of the commercial practice of traditional ogi manufacture using high-lysine (opaque-2) corn and normal corn. *Proc. 10th Int. Congr. Nutr.*, Kyoto, Japan, p. 402.

Bothast, R.J., Anderson, R.A., Warner, K. & Kwolek, W.F. 1981. Effects of moisture and temperature on microbiological and sensory properties of wheat flours and corn meal during storage. *Cereal Chem.*, 58: 309-311.

Braham, J.E., Flores, M., Elías, L.G., de Zaghi, S. & Bressani, R. 1969. Mejoramiento del valor nutritivo de dietas de consumo humano. II. Suplementación con mezcla vegetal INCAP 9 y leche. *Arch. Latinoam. Nutr.*, 19: 253-264.

Brekke, O.L. 1968. Corn dry-milling: stress crack formation in tempering of low-moisture corn, and effect on degerminator performance. *Cereal Chem.*, 45: 291-303.

Bressani, R. 1974. Complementary amino acid patterns. *In* P.L. White & D.C. Fletcher, eds. *Nutrients in processed food proteins*. Acton, Mass., USA, Publishing Sciences

Group.

Bressani, R. 1981. The role of soybeans in food systems. *J. Am. Oil Chem. Soc.*, 58: 392-400.

Bressani, R., Campos, A.M., Squibb, R.L. & Scrimshaw, N.S. 1953. Nutritive value of Central American corns. IV. The carotene content of thirty-two selections of Guatemalan corn. *Food Res.*, 18: 618-624.

Bressani, R. & Conde, R. 1961. Changes in the chemical composition and in the distribution of nitrogen of maize at different stages of development. *Cereal Chem.*, 38: 76-84.

Bressani, R., Elías, L.G., Scrimshaw, N.S. & Guzmán, M.A. 1962. Nutritive value of Central American corns. VI. Varietal and environmental influence on the nitrogen, essential amino acids and fat content of 10 varieties. *Cereal Chem.*, 39: 59-67.

Bressani, R., Elías, L.G. & Gómez-Brenes, R.A. 1969. Protein quality of opaque-2 corn. Evaluation in rats. *J. Nutr.*, 97: 173-180.

Brown, R.B., Fulford, G.N., Daynard, T.B., Meiering, A.G. & Otten, L. 1979. Effect of drying methods on grain corn quality. *Cereal Chem.*, 56: 529-533.

Brunson, A.M. & Quackenbush, F.W. 1962. Breeding corn with high provitamin A in the grain. *Crop Sci.*, 2: 344-347.

Cantor, S.M. & Roberts, H.J. 1967. Improvement in protein quality in corn-based foods. *Cereal Sci. Today*, 12: 443-445, 460-462.

Choto, C.E., Morad, M.M. & Rooney, L.W. 1985. The quality of tortillas containing whole sorghum and pearled sorghum alone and blended with yellow maize. *Cereal Chem.*, 62: 51-55.

Cravioto, B., Cravioto, R.O. & Cervantes, M. 1965. Eficiencia proteica de la harina de masa enriquecida con harina de soja y de la adicionada con proteína de ajonjolí. *Ciencia (Mes.)*, 24: 159-162.

De Padua, M.R. & Maroun, H.P. 1984. Rheological behavior of Venezuela arepa dough from precooked corn flour. *Cereal Chem.*, 61: 37-41.

Dutra de Oliveira, J.E. & de Souza, N. 1972. El valor nutritivo de productos de la molienda del maíz, de la suplementación con aminoácidos y de mezclas de maíz comun y de opaco-2. *In* R. Bressani, E. Braham & M. Béhar, eds. *Mejoramiento nutricional del maíz. INCAP Pub. L-3*, p. 209-215. Guatemala, INCAP.

Earle, F.R., Curtis, J.J. & Hubbard, J.E. 1946. Composition of the component parts of the corn kernel. *Cereal Chem.*, 23: 504-511.

Erdmenger, J.J., Elías, L.G., de Souza, N., Salomon, J.B., Bressani, R., Arroyave, G. & Habicht, J.P. 1972. Estudio, en ratas, del efecto de la suplementación proteínica de una dieta típica de una comunidad rural de Guatemala. *Arch. Latinoam. Nutr.*, 22: 179-190.

Evans, J.W. 1941. Changes in biochemical composition of the corn kernel during development. *Cereal Chem.*, 18: 468-473.

FAO/WHO. 1973. *Energy and protein requirements*. FAO Nutrition Meetings Report Series No. 52; WHO Technical Reports Series No. 522. Rome, FAO.

Feria-Morales, A.M. & Pangborn, R.M. 1983. Sensory attributes of corn tortillas with substitutions of potato, rice and pinto beans. *J. Food Sci.*, 48: 1124-1134.

Fernandez, J.J., Guerra, M.J. & Racca, E. 1991. Precocción de harina de soya y maíz por microonda y su uso en la preparación de arepas. *Arch. Latinoam. Nutr.*, 41(3).

Fields, M.L., Hamad, A.M. & Smith, D.K. 1981. Natural lactic acid fermentation of corn meal. *J. Food Sci.*, 46: 900-902.

Flores, M. 1966. Food intake of Guatemalan Indian children aged 1 to 5. *J. Am. Diet Assoc.*, 48: 480.

Franz, K. 1975. Tortillas fortified with whole soybeans prepared by different methods. *J. Food Sci.*, 40: 1275-1277.

Frey, K.J. 1949. The inheritance of protein and certain of its components in maize. *Agron. J.*, 41: 113-117.

Garcia-Lopez, S. & Wyatt, C.J. 1982. Effect of fiber in corn tortillas and cooked beans on iron availability. *J. Agric. Food Chem.*, 30: 724-727.

Gopalan, C., Belavady, B. & Krishnamurthi, D. 1969. The role of leucine in the pathogenesis of canine black-tongue and pellagra. *Lancet*, 11: 956-957.

Graham, G.G., Lembcke, J. & Morales, E. 1990. Quality-protein maize as a sole source of dietary protein and fat for rapidly growing young children. *Pediatrics*, 85: 85-91.

Green, J.R., Lawhon, J.T., Cater, C.M. & Mattil, K.F. 1976. Protein fortification of corn tortillas with oilseed flour. *J. Food Sci.*, 41: 656-660.

Green, J.R., Lawhon, J.T., Cater, C.M. & Mattil, K.F. 1977. Utilization of whole undefatted glandless cottonseed kernels and soybeans to protein fortify corn tortillas. *J. Food Sci.*, 42: 790-794.

Grogan, C.O. & Blessin, C.W. 1968.

Characterization of major carotenoids in yellow maize lines of different pigment concentration. *Crop Sci.*, 8: 730-732.

Hamad, A.M. & Fields, M.L. 1979. Evaluation of the protein quality and available lysine of germinated and fermented cereals. *J. Food Sci.*, 44: 456-459.

Hansen, D.W., Brimhall, B. & Sprague, G.F. 1946. Relationship of zein to the total protein in corn. *Cereal Chem.*, 23: 329-335.

Holder, D.G., Glover, D.V. & Shannon, J.C. 1974. Interaction of shrunken-2 and five other carbohydrate genes of corn endosperm. *Crop Sci.*, 14: 643-646.

Jellum, M.D. 1967. Fatty acid composition of corn (*Zea mays* L.) as influenced by kernel position on ear. *Crop Sci.*, 7: 593-595.

Jellum, M.D. 1970. Plant introductions of maize as a source of oil with unusual fatty acid composition. *J. Agric. Food Chem.*, 18: 365-370.

João, W.S.J., Elías, L.G. & Bressani, R. 1980. Efecto del proceso de cocción-extrusión (Brady Crop Cooker) sobre el valor nutritivo de mezclas elaboradas a base de frijol caupí (*Vigna sinensis*)-maíz y de frijol caupí-yuca. *Arch. Latinoam. Nutr.*, 30: 539-550.

Kodicek, E., Braude, R., Kon, S.K. & Mitchell, K.G. 1956. The effect of alkaline hydrolysis of maize on the availability of its nicotinic acid to pigs. *Br. J. Nutr.*, 10: 51.

Kodicek, E., Braude, R., Kon, S.K. & Mitchell, K.G. 1959. The availability to pigs of nicotinic acid in tortilla baked from maize treated with lime water. *Br. J. Nutr.*, 13: 363-384.

Kodicek, E. & Silson, P.W. 1959. The availability of bound nicotinic acid to the rat. 1. The effect of lime water treatment of maize and subsequent baking into tortilla. *Br. J. Nutr.*, 13: 418.

Krehl, W.A., Henderson, L.M., de la Huerga, J. & Elvehjem, C.A. 1946. Relation of amino acid imbalance to niacin-tryptophan deficiency in growing rats. *J. Biol. Chem.*, 166: 531-540.

Laguna, J. & Carpenter, K. 1951. Raw versus processed corn in niacin deficient diets. *J. Nutr.*, 45: 21.

Lillehoj, E.B., Kwolek, W.F., Horner, E.S., Widstrom, N.W., Josephson, L.M., Franz, A.O. & Catalano, E.A. 1980. Aflatoxin contamination of preharvest corn: role of *Aspergillus flavus* inoculum and insect damage. *Cereal Chem.*, 57: 255-257.

Makinde, M.A. & Lachange, P.A. 1976. Tryptophan: first limiting amino acid in ogi. *Nutr. Rep. Int.*, 14: 671-679.

Makinde, M.A. & Lachange, P.A. 1989. Optimization of protein nutritive value of ogi. *Niger. J. Nutr. Sci.*, 10: 85-93.

Mitchell, H.H., Hamilton, T.S. & Beadles, J.R. 1952. The relationship between the protein content of corn and the nutritional value of the protein. *J. Nutr.*, 48: 461-476.

Mottern, H.H., De Buckle, T.S. & Pardo, C. 1970. Protein enrichment of Colombian corn cakes. *Cereal Sci. Today*, 15: 108-112.

Nofsinger, G.W. & Anderson, R.A. 1979. Note on inactivation of aflatoxin in ammonia-treated shelled corn at low temperatures. *Cereal Chem.*, 56: 120-122.

Pardo T., F., Mora, J.O., Páez F., J., de Onshuss, Y. & de la Cruz de Villota, M. 1972. Aceptabilidad del maíz opaco-2 en Colombia. *Arch. Latinoam. Nutr.*, 22: 561-575.

Plahar, W.A. & Leung, H.K. 1983. Composition of Ghanaian fermented maize meal and the effect of soya fortification on sensory properties. *J. Sci. Food Agric.*, 34: 407.

Plahar, W.A., Leung, H.K. & Coon, C.N. 1983. Effect of dehydration and soy fortification on physico-chemical nutritional and sensory properties of Ghanaian fermented maize meal. *J. Food Sci.*, 48: 1255.

Reddy, V. & Gupta, C.P. 1974. Treatment of kwashiorkor with opaque-2 maize. *Am. J. Clin. Nutr.*, 27: 122-124.

Reinhold, J.G., Garcia, J.S. & Garzon, D. 1981. Binding of iron by fiber of wheat and maize. *Am. J. Clin. Nutr.*, 34: 1384-1391.

Sanchez-Marroquin, A., Feria-Morales, A., Maya, S. & Ramos-Moreno, V. 1978. Processing, nutritional quality and sensory evaluation of amaranth enriched corn tortillas. *J. Food Sci.*, 52: 1611-1614.

Sauberlich, H.E., Chan, W.U. & Salmon, W.D. 1953. The amino acid and protein content of corn as related to variety and nitrogen fertilization. *J. Nutr.*, 51: 241-250.

Singh, J. & Koshy, S. 1974. Role of opaque-2 maize in child nutrition. *Indian J. Genet.*, 34: 1182-1190.

Sirinit, K., Soliman, A.G.M., Van Loo, A.T. & King, K.W. 1965. Nutritional value of Haitian cereal-legume blends. *J. Nutr.*, 86: 415.

Smith, O., De Buckle, T.S., de Sandoval, A.M. & Gonzalez, A.E. 1979. Production of precooked corn flours for arepa making using an extrusion cooker. *J. Food Sci.*, 44:

816-819.

Squibb, R.L., Braham, J.E., Arroyave, G. & Scrimshaw, N.S. 1959. A comparison of the effect of raw corn and tortillas (lime-treated corn) with niacin, tryptophan or beans on the growth and muscle niacin of rats. *J. Nutr.*, 67: 351-361.

Tellez-Girón, A., Acuff, G.R., Vanderzant, C., Rooney, L.W. & Waniska, R.D. 1988. Microbiological characteristics and shelf-life of corn tortillas with and without antimicrobial agents. *J. Food Prot.*, 51: 945-948.

Tuite, J., Foster, G.H., Eckhoff, S.R. & Shotwell, O.L. 1986. Sulfur dioxide treatment to extend corn drying time. *Cereal Chem.*, 63: 462-464.

Ulloa, M., Herrera, T. & Taboada, J. 1977. Pozol, a fermented maize dough consumed in Southeastern Mexico. In *Symposium on indigenous fermented foods*. Bangkok, Thailand. Government of Thailand, UNEP, Unesco, IFS, CFT, ICRO.

Viteri, F.E., Torún, B., Arroyave, G. & Pineda, O. 1981. Use of corn-bean mixtures to satisfy protein and energy requirements of preschool children. *In* B. Torún, V.R. Young & W.M. Rand, eds. *Protein-energy requirements of developing countries: evaluation of new data*, p. 202-209. Tokyo, United Nations Univ.

Watson, S.A. & Yahl, K.R. 1967. Comparison of wet milling properties of opaque-2 high-lysine corn and normal corn. *Cereal Chem.*, 44: 488-498.

Wolzak, A., Elías, L.G. & Bressani, R. 1981. Protein quality of vegetable proteins as determined by traditional biological methods and rapid chemical assays. *J. Agric. Food Chem.*, 29: 1063-1068.

Yamaguchi, J. 1974. Varietal traits limiting the grain yield of tropical maize. IV. Plant traits and productivity of tropical varieties. *Soil Sci. Plant Nutr.*, 20: 287-304.

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