

Fortification of Corn Masa Flour with Iron and/or Other Nutrients

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A Literature and Industry Experience Review



Prepared by
SUSTAIN

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**FORTIFICATION OF CORN MASA FLOUR WITH IRON
AND/OR OTHER NUTRIENTS -
A LITERATURE AND INDUSTRY EXPERIENCE REVIEW**

EXECUTIVE SUMMARY

Corn (*Zea mays L*) is one of the most important cereal grains in the Latin American diet, particularly in Mexico, Central America, Colombia, Venezuela and Brazil. In the rural areas of Central America, alkaline cooked corn based products contribute 39-65% and 27-53% of the daily caloric and protein intakes respectively. It also provides significant amounts of Ca, Fe, vitamin B₁ and niacin.

The amounts consumed, as well as the forms of consumption, are quite varied among these countries. In South America, arepas and other products are made from decorticated, degermed, precooked corn flour. These products are not cooked with alkali. Their pH is slightly below 7.0. In Mexico and Central America, corn is transformed into tortillas through a process called nixtamalization (from the Nahuatl words *nixtli* = ashes and *tamalli* = dough or masa).

Nixtamalization

Corns is cooked in boiling water and lime (1% based on corn weight) for 5-50 min, steeped overnight and washed to remove excess lime and pericarp. The nixtamal (washed, alkaline cooked corn) is stone ground into dough (masa) which is formed into discs that are baked on a hot griddle. The modern process ranges from relatively primitive hand processing to large capacity automated processes.

Nixtamalized corn flour (NCF)

NCF is produced by alkaline cooking of corn, washing, grinding the nixtamal and drying to produce flour. The flour is sieved into different particle sizes, which are blended to obtain NCF with optimum properties for different applications. Usually, 100 kg of corn yields 93 to 98 kg of NCF.

The use of NCF is increasing in popularity because it eliminates the tedious, labor-intensive cooking, washing, and grinding of corn to produce masa for tortillas and snacks. Grupo MASECA is the main NCF producer worldwide with 19 plants located in

the USA and Latin America. Other companies that produce NCF are MINSA (with 6 plants), AGROINSA (with 2 plants), Illinois Cereal Mills (Cargill) and Quaker Oats. In 1994, NCF accounted for 27% of the masa used for tortillas in the U.S. and Latin America (Torres et al 1996). The corn tortilla industry grew 36% in the United States between 1992 and 1996. Use of NCF is increasing significantly in Mexico, Central America, and the United States.

Major advantages of using NCF are reduced pollution, product flexibility, convenience, and reduced capital costs since corn processing equipment is not required (Rooney and Serna Saldivar, 1987; Serna Saldivar et al 1990). There are different NCFs available for the production of white and yellow table tortillas, restaurant style chips, tortilla chips, corn chips and tamales. By using NCF a consistent quality of the end product can be easily achieved. Another advantage is that it can be blended with other dry ingredients (i.e. preservatives, gums, enrichment mixes etc.).

Nutritional value of tortillas

Nixtamalization is a critical factor in the Latin American diet because it enhances the nutritional value of corn. The calcium content is increased, niacin becomes available and phytic acid levels are reduced; but lime-treated corn products are still deficient in other nutrients such as lysine, tryptophan, vitamin A, vitamin C, B vitamins and iron. Considerable information on the nutritional value of fresh masa exists.

Fortification of fresh masa and tortillas

No information on fortification of fresh masa with iron and other trace minerals was found. The nutritional value of tortillas can be increased by the addition of whole soybeans, defatted soybean flour, amaranth, fish meal, torula yeast and by addition of lysine and tryptophan (Bressani and Marengo 1963; Bressani et al. 1968, 1974, 1979; Del Valle and Perez Villaseñor, 1974; Sanchez Marroquin et al. 1987; Tonella et al. 1983; Serna Saldivar et al. 1988). Fortification with soybeans is the most feasible because of their low cost, high availability, high protein content and complementary effect on corn proteins. Defatted soybean flour has the advantage of better shelf stability although full-fat soybean flour produces a higher calorie tortilla.

Fortification of dry NCF and related products

The Nutrition Institute “Salvador Zubiran” of Mexico recently conducted a NCF enrichment study. NCFs (MASECA and MINSA) were enriched with a premix containing soy-oat-rice flour (10%) and seven micronutrients (iron, zinc, vitamin A, C, riboflavin, niacin and folic acid).

During a three-month pilot test, MINSA and MASECA produced 2,300 MT of enriched NCF. The micronutrients were added in a quantity necessary to meet 100% of the recommended daily intake by consuming six tortillas/d. An increase in digestibility (10%), protein quality (40%) and organoleptic quality (70%) was obtained. The flavor of the tortillas was different but close to the original corn flavor. No significant odor change was detected and most people said they would buy the product in the future (Torres et al. 1996).

Iron enrichment of precooked corn flours for arepas

Commercially prepared precooked arepa flours that require only hot water to produce a dough are made by cooking the corn grits to gelatinize the starch, putting the cooked grits through flaking rolls, and grinding the dry flakes into flour or meal of acceptable particle size distribution. Approximately 700, 000 tons of corn are processed annually into arepa flour in nine plants in Venezuela (Cuevas et al. 1985).

Precooked corn flours for arepas were enriched in Venezuela (1993) with 50 mg Fe (as ferrous fumarate)/kg flour. The consumption of precooked corn flour for arepas per capita was 80 g/d for the total population in 1994. A preliminary survey was carried out in 1994 in a population of 307 children aged 7, 11, and 15. The results showed that the prevalence of iron deficiency (determined by measuring the serum ferritin concentrations) and the prevalence of anemia were reduced from 37% and 19%, respectively in 1992, to 15% and 10%, respectively in 1994. The color, odor, texture, and flavor of the flours did not change for six months. The only adverse effect observed during the initial two years of the fortification program occurred in two regions of the country in which hard water is used for making arepas; the bread turned slightly dark due to the addition of the hard water (Layrisse et al.1996).

Fortification with vitamins

NCF can be fortified with vitamins by adding them to the flour in the form of a dry premix. When mixing dry vitamins with flours a careful selection of the physical characteristics of the compounds is important to ensure adequate mixing and to minimize segregation during storage. Vitamin A is sufficiently stable in flours at warehouse temperatures for up to six months and it has no apparent tendency to segregate during handling or shipping in bags. The form of vitamin A most commonly used in the fortification of flour is dry stabilized vitamin A palmitate (type 250-sd) powder form. Thiamin, riboflavin, niacin, pyridoxine, folate and calcium pantothenate were used in their pure crystalline form and were stable during storage.

Parrish et al. (1980) added to dry milled cornmeal a premix formulated with vitamins and iron (as reduced iron at 10 g cwt.). Losses of vitamin A were less than 20% when stored up to 6 months at room temperature. In Venezuela (1993) precooked arepa flours containing 50 mg of ferrous fumarate/kg plus vitamin A, thiamine, riboflavin, and niacin had good stability at room temperature storage at normal moisture levels (6.5%). Retention of the vitamins after cooking was also good. These studies show that vitamin fortification of precooked corn flours is feasible.

Recommendations

Our current knowledge of and technology for fortification of NCF with iron is very limited. However, the studies conducted on iron enrichment of corn dry-milling products, precooked corn flours for arepas, and wheat flours give preliminary knowledge and techniques for the development of an iron fortified NCF. An important difference between these products and NCF is pH. NCFs are neutral while corn dry-milling products, precooked corn flours for arepas, and wheat flours have a lower pH.

NCFs available in the market have a pH ranging from 6 to 7.2. At pH 7 ferrous ion will precipitate as FeOH_2 , having a solubility of about 10^{-1} M. Ferric ion is much less soluble than ferrous, precipitating as FeOH_3 , which has a solubility of about 10^{-16} M at pH 7. The formation of sparingly soluble hydroxides with increasing pH has nutritional significance, as solubility of iron is a prerequisite to its absorption in the gut. In the acid medium of the stomach, the iron is in a soluble, hydrated form. Upon passing into the alkaline medium of the small intestine, where all iron absorption takes place, most of the

iron would be expected to precipitate. However, it is evident that extensive precipitation does not occur because appreciable iron absorption takes place in the small intestine.

The most logical approach would be to enrich NCF with iron by mixing up to 50 mg of ferrous fumarate/kg. This compound is known to have certain organoleptic properties that can be appealing and could be added to the product at a reasonable cost (\$9.15/kg). It also has a bioavailability comparable to that of ferrous sulfate. Ferrous fumarate added to NCF would not segregate significantly and the quality of the products made with this flour would most likely be acceptable since it is less reactive than ferrous sulfate, leading to fewer problems with off-coloration and catalysis of fat oxidation reactions.

The high pH of NCFs might not reduce the availability of ferrous fumarate significantly since the pH of the small intestine is also high. The use of iron-complexes like ferrous gluconate to increase iron absorption by preventing precipitation might not be justified since they are six times more expensive than ferrous fumarate.

Since there is relatively little information on the effect of iron and other nutrients on what happens to the flavor, color, odor and stability of the NCF and tortillas it is desirable to conduct storage studies and quality evaluation of tortillas made from iron enriched NCF. Such studies would establish baseline information on the major factors that might affect organoleptic properties of the products. No major problems are anticipated but a few controlled experiments would clarify the situation. There is a need to evaluate the availability of the iron in tortillas at least using *in vitro* methods. There does not appear to be a need for extensive human feeding trials which would be expensive.

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I

NUTRITIONAL QUALITY OF NIXTAMALIZED CORN MASA FLOUR. ACHIEVEMENT THROUGH FORTIFICATION WITH MICRONUTRIENTS

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SUMMARY

Results from a relatively large number of surveys show that corn (*Zea mays*) is one of the most important cereal grains for the nutrition of large groups of the Latin American population, particularly for Mexico, Central America, Colombia and Venezuela.

In South America, corn is consumed in a number of ways as a decorticated, degermed precooked flour and one of the most popular forms is the arepa. In Mexico and in Central America, corn is also consumed in many different forms made by processing the grain by nixtamalization.. This process consists in cooking sound hard endosperm white or yellow corn with lime for 40 to 60 minutes, allowing a soaking period of 8-12 hrs, washing to remove the seed coat and excess lime, before grinding into a dough which can be made into tortilla or dried into a flour. Lime-treated corn flour represents the whole corn grain without the seed coat. Therefore, it has more protein, more fat and is of a better protein quality than the degermed corn flour consumed in South America. Furthermore, it is a very rich source of calcium due to the lime-cooking process.

In rural areas of Mexico and of the Central American countries nixtamalization is carried out daily by the traditional process of using whole corn to produce a dough from which tortillas are made. In recent years industrialized lime-treated corn flour has become available in Central America, a product first introduced in Mexico some 50-60 years ago.

The nixtamalization process, as done at home level, is relatively simple to carry out; however, it is time consuming, requires hard work and uses relatively large amounts of water. The industrial process is basically similar but with changes in cooking operations to decrease processing times, followed by dehydration and grinding, producing a convenient dry flour for the market.

Intake of corn as tortillas is higher in rural areas than in urban areas. Tortillas contribute 39-65% of the daily intake of calories in the rural areas of the Central American countries and 27 to 53% of the daily protein intake. They also provide some amounts of calcium and vitamins B₁ and niacin.

During processing by the alkaline cooking procedure, no major changes occur in the macro-nutrients. However, the losses in thiamine, riboflavin, and niacin are significant. There is a significant increase in Ca content and small decreases in dietary fiber. Losses in phytic acid have also been reported. The calcium from lime-treated corn is highly bioavailable, as is niacin. On the other hand, Fe availability, which is low in raw corn, does not change much with the process. The results of studies have suggested a negative relationship between iron bioavailability and phytic acid content.

Because of the nutritional importance of corn to many Latin American populations, efforts have been made to improve its nutritional value. The approaches used have included supplementation with lysine and tryptophan as crystalline amino acids or as components of protein supplements such as vegetable or animal protein sources. Another approach was the development of corn varieties containing high levels of the two limiting amino acids, as in Quality Protein Maize. Fortification formulas containing protein as well as vitamins and minerals were also developed in Guatemala and Mexico. However, the technologies have not been implemented, although acceptability and market trials have been promising. All the nutritional improvement approaches, when tested with children, have shown a highly significant improvement in the nutritional value of the fortified corn masa flour.

The fortification of lime-treated flour is recommended for several reasons:

- 1) the importance of corn in the diets of Latin American countries,
- 2) the wide availability and use of dried masa flour,
- 3) the losses of B-vitamins during nixtamalization,
- 4) the usually low bioavailability of iron in nixtamalized corn products.

It is important, however, to have available some basic information on the factors which are known to decrease the bioavailability of iron, as applicable to lime-treated corn flour, alone and in typical rural diets.

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INTRODUCTION

Corn (*Zea mays*) represents one of the most important cereal grains for the nutrition of the Latin American population. The amounts consumed, as well as the forms of consumption, are quite variable among different countries in the region. In Mexico and in Central America, where consumption of corn is higher than anywhere else on the continent, the grain is transformed into edible products by a unique process known as **nixtamalization**, which was developed by ancient Maya and Aztec civilizations. The main edible product of such a process is the **tortilla**. In the northern part of South America, particularly in Colombia and Venezuela, corn processed by milling is converted into a precooked degerminated flour for consumption in the form of **arepas**. In other countries, maize grits, polenta and corn meal are products of direct consumption or used as ingredients for other forms of consumption.

The transformation of corn into edible forms, mainly tortillas, is based on processes which were and still are carried out at home level. These usually require large amounts of labor, long times of preparation, and large amounts of water and energy, and yield products of variable quality. Today, however, there is an increasing demand and use of industrially prepared flours. These provide benefits to the consumer, such as convenience, availability, and a more stable quality. The availability of a precooked corn flour for arepa preparation was initiated around 1962 in Venezuela and Colombia and there are at least 12 different brands in Venezuela (1). Industrially produced tortilla flour or lime-treated corn flour was first introduced in the Mexican market in the early 1950's with some 6-7 different brands (2). Throughout the years it has expanded to a very large industry in Mexico and in the United States (2, 3, 4) and some of the brands available in the Mexican market are today found in various markets in the Central American countries. In Guatemala, lime-treated corn flour was first introduced around 1962, and its production and availability has increased up to the present time (5). The Central American market has available both locally-produced and imported brands of lime-treated corn flours.

The industrial process to convert raw corn into a lime-treated corn flour is based on the operations conducted at home level consisting in grain cleaning and washing;

cooking in water for some 40-60 min at an alkaline pH using lime; cooling and soaking for around 12 hrs; washing to remove the seed coat and excess lime; and grinding to make a dough. This in turn can be shaped into tortillas with an additional cooking operation, or it can be dehydrated and finely ground into flour, the product now available. The chemistry, technology and nutritional value of corn subjected to this multi-step process have been reviewed (3, 4, 6).

The adaptation and improvement of the home technology to an industrial level has resulted in more efficient production of quality products. The industry has made important contributions in defining the quality attributes of the raw material and in maximizing production through agronomic technologies. Also, more efficient operations of the process have been introduced, including the mechanical production of the final product, the tortilla. The quality of the flour in terms of its conversion to tortillas and other products, its shelf-life with various packaging materials, its safety and organoleptic characteristics have also received attention by the industry and research institutions. One brand (Torti-Ya) produced in Guatemala, has been enriched with vitamins and iron since its introduction in the market in 1972¹.

As early as 1960, a number of efforts have been made in Central America by INCAP to find practical solutions to improve the nutritional quality of foods consumed by the majority of the population. Plans were made and implemented for salt iodization (7,8) for the addition of vitamin A to sugar (9) and for the development of high-quality protein foods (10). These efforts have been successful and have contributed in a significant way to reduce the problems of malnutrition in the region. Because corn is such an important food item in the diet, activities were also conducted as early as 1968 to improve the nutritional value of lime-treated corn, through protein, protein/calorie, vitamin and mineral fortification, both at the home and at the industrial levels (11). The information developed was and is sound, but it proved difficult to implement at the time.

With the increased production and availability of industrially manufactured lime-treated corn flour and with the convenience it provides, it appears timely to improve its

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nutritional quality through fortification.

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THE CENTRAL AMERICAN DIET

Food intake

The nutritional evaluation of the population in Central America has been one of the main objectives of INCAP since the initiation of its activities in 1949 (1). Between 1969 and 1996 various surveys have been conducted which show in general little change in the kinds and amounts of foods ingested (2); however significant nutritional improvements have been made with the almost total elimination of iodine deficiency (due to the addition of iodine to salt) and vitamin A deficiency (due to the addition of vitamin A to sugar). The Micronutrient National Survey of 1995 showed that sugar fortified with vitamin A contributed 49% of the total retinol ingested by children 1 to 5 years of age. The contribution was even higher in rural areas (56%) where vitamin A deficiency is higher. The analysis of the data indicated that the vitamin A problem, improved after 1967 and especially after 1988 when sugar fortification began (3).

The data shown in Table 1, taken from the surveys published in 1969 (1), summarize the kinds and amounts of foods ingested by the rural population, while those in Table 2 present the same data for the urban population. The data show a relatively wide variety of food items consumed, with cereal grains being of first importance as expected. Of the cereal grains, corn tortillas are consumed in largest amounts in El Salvador, Guatemala and Honduras in both rural and urban areas, consumption decreases to some extent in Nicaragua and more so in Costa Rica and Panama. In the latter, corn meal replaces the tortilla in the urban areas. The data show small intakes of foods from animal origin in most countries, with relatively high intakes of foods from vegetable origin, mainly in rural areas. Such diets provide relatively high intakes of dietary fiber, which is of importance with respect to the bioavailability of microelements.

Food consumption surveys conducted in Guatemala in 1991 (2) showed that the rural population had an intake of 454 g/person per day of food products prepared from corn while the urban population had an intake of 251 g. The surveys showed further a strong association between corn products consumption and income. Those with an

income of less than Q550/month consumed 439 g of corn derived food products; those with income between Q550-Q900 consumed 327 g, and those with income over Q900/month consumed only 256 g. There were also high regional differences with the urban population consuming less corn products than the rural population. As in 1965, the 1991 survey indicated that corn in various food products is the cereal grain of the greatest importance, representing around 79% of all cereal grain intakes consumed in 99% of the homes at national level. It is of interest to point out that the other food item of major importance is beans, consumed in 97% of the homes in Guatemala.

Table 1						
Food intakes by the rural population in the Central American countries - 1969						
(g/person per day)						
Foods	Guatemala	El Salvador	Honduras	Nicaragua	Costa Rica	Panamá
Milk products*	125	190	194	243	193	73
Eggs	17	10	13	12	15	11
Meats	40	37	41	58	40	90
Beans	50	59	56	72	57	20
Fresh vegetables	63	53	51	27	66	25
Fruits	14	17	40	41	7	50
Bananas & Plantain	26	16	43	72	47	99
Root crops	14	13	22	33	46	82
Cereals:						
Rice	16	27	29	54	100	186
Corn tortillas	491	528	338	196	62	6
Corn tamales	5	5	2	3	-	-
Wheat bread**	40	26	20	35	66	47
Other	11	6	5	21	-	29
Sugar	53	41	39	58	89	51
Fats	8	15	16	19	19	26
Miscellaneous:						
Coffee	9	7	9	8	10	6
Carbonated drinks	1	-	3	14	-	8
Beer	-	-	-	-	-	-
Other	-	2	-	-	-	21

*Expressed as liquid/fluid milk

**Wheat derived food products (bread, pasta, etc.) ; Reference (1).

Table 2
Food intakes by the urban population in the Central American countries - 1969
(g/person per day)

Foods	Guatemala	El Salvador	Honduras	Nicaragua	Costa Rica	Panamá
Milk products*	304	237	289	377	350	163
Eggs	28	31	21	21	23	19
Meats	65	77	87	90	24	134
Beans	45	52	47	50	48	19
Fresh vegetables	120	90	56	74	126	68
Fruits	63	71	54	52	60	99
Bananas & plantain	37	49	49	75	57	75
Root crops	22	12	24	25	55	70
Cereals:						
Rice	27	55	50	80	103	150
Corn tortillas	155	249	202	84	21	6
Corn tamales	2	-	1	-	-	-
Wheat bread**	134	66	83	51	96	74
Other	15	5	7	33	4	7
Sugar	71	38	45	63	77	42
Fats	20	37	21	29	41	35
Miscellaneous:						
Coffee	8	6	9	4	13	6
Carbonated drinks	12	7	18	22	18	33
Beer	3	-	-	-	-	-
Other drinks	-	4	2	-	-	-
Other	-	4	1	-		4

*Expressed as liquid milk

**Wheat derived food products (bread, pasta, etc.); Reference (1).

Nutritional Intake and Adequacy

Nutrient intakes associated with rural and urban diets are presented in Table 3 (rural diets) and in Table 4 (urban diets). These results show higher nutritional intakes in urban as compared to rural areas as it would be expected from food intake data (1). However, this is not the case for calcium in Guatemala, El Salvador and Honduras, where the intake is larger in rural areas; in Nicaragua, Costa Rica and Panama calcium intake is larger in urban areas.

The adequacy of the above nutrient intake is shown in Tables 5 and 6 for rural and urban diets, respectively (1). This information together with the biochemical data derived from the 1969 surveys showed that in 1969 protein/calorie deficiencies existed in all countries with the exception of Panama; deficiencies in vitamin A were noted in all countries, with the exception of Costa Rica; deficiencies in iron intakes were found in all countries with the exception of Costa Rica and Panama; a moderate deficiency in riboflavin existed in all countries; all countries had niacin deficiencies except Panama, with this country showing a deficiency in calcium intakes, as summarized in Table 7.

Since the data is derived from surveys conducted before 1969, the possible impact of fortification programs implemented since then have not been taken into consideration. As indicated previously, iodine fortification of salt and vitamin A fortification of sugar have significantly reduced the deficiency of these nutrients.

Wheat flour, a food item of relatively low but growing intake, has been fortified for some time with B-complex vitamins and iron. Recently it has been proposed that wheat flour in Central America be fortified with 60 mg/kg of flour with reduced electrolytic iron of a particle size less than 40 μ m (4).

Table 3
Nutrient intakes in rural diets in the Central American countries - 1969

Nutrient	Guatemala	El Salvador	Honduras	Nicaragua	Costa Rica	Panamá
Energy, kcal	1994	2146	1832	1986	1894	2089
Proteins, g	60.4	67.9	58.0	64.4	53.6	60.1
Fat, g	32.0	39.3	40.1	47.5	43.9	49.8
Carbohydrates, g	382	396	315	338	332	357
Ca, mg	994	1092	883	763	580	301
P, mg	1244	1341	1140	1184	981	932
Fe, mg	14.6	11.6	15.5	18.2	15.4	14.3
Vit A, mg	0.69	0.268	0.384	0.508	0.539	0.548
Thiamine, mg	1.05	1.06	0.89	0.86	0.76	0.92
Riboflavin, g	0.72	0.78	0.79	0.93	0.84	0.69
Niacin, mg	11.24	12.0	10.3	10.7	10.7	14.3
Vit C, mg	34	36	59	66	52	87

Nutrient	Guatemala	El Salvador	Honduras	Nicaragua	Costa Rica	Panamá
Energy, kcal	2065	2204	2061	2108	2330	2101
Protein, g	66	69.9	70.9	72.2	67.3	70.9
Fat, g	53	64.4	59.9	59.6	66.8	58.7
Carbohydrates, g	344	338	322	331	344	328
Ca, mg	866	865	864	901	855	419
P, mg	1203	1232	1203	1224	1157	1034
Fe, mg	14	13.8	13.3	15.4	16.3	14.9
Vit A, mg	0.8	0.915	0.686	0.941	1.106	1.110
Thiamin, mg	1.0	0.99	0.96	0.88	0.97	0.91
Riboflavin, mg	1.1	1.08	1.12	1.33	1.28	0.98
Niacin, mg	10.6	12.1	12.8	16.9	13.3	14.3
Vit C, mg	64	91	74	82	102	107

Table 5
Adequacy of nutrient intakes in rural diets of the Central American countries- 1969
%

Nutrient	Guatemala	El Salvador	Honduras	Nicaragua	Costa Rica	Panamá
Energy, kcal	103	109	89	96	91	104
Protein	123	128	108	115	98	112
Calcium	200	207	166	144	112	59
Iron	147	114	152	175	150	141
Vit A	37	24	74	44	49	49
Thiamin	136	136	109	105	93	116
Riboflavin	63	66	64	75	68	58
Niacin	88	93	75	78	78	118
Vit C	81	62	130	144	117	194

Table 6						
Adequacy of nutrient intakes in urban diets of the Central American countries - 1969						
Nutrient	Guatemala	El Salvador	Honduras	Nicaragua	Costa Rica	Panamá
Energy, kcal	98	106	99	99	106	98
Protein	118	122	124	119	111	120
Calcium	176	170	168	173	165	80
Iron	144	133	129	109	155	143
Vit A	50	79	60	83	97	97
Thiamin	121	121	116	104	111	107
Riboflavin	94	86	90	104	97	76
Niacin	77	88	93	84	92	105
Vit C	145	198	163	180	221	230

Table 7
Summary of nutrient deficiencies

Country	Calorie s	Protein	Vit A	Iron	Riboflavin	Ca	Niacin
Guatemala	D	D	D	D	D	A	D
El Salvador	D	D	D	D	D	A	D
Honduras	D	D	D	D	D	A	D
Nicaragua	D	D	D	D	D	A	D
Costa Rica	A	A	A	A	D	A	D
Panamá	A	A	D	A	D	D	A

A = adequate; D = deficient

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Consumption of lime-cooked corn in Central America

The data from Table 8 show the amounts of lime-treated corn and other cereal grains consumed as tortillas and in other food forms by the rural and urban population in the Central American countries (1). Other data are shown in Tables 9, 10, and 11 from various studies, mainly from Guatemala. Lime-treated corn intakes by children 1 to 5 years of age were published by Flores et al. (2). Intakes as dry corn were 64 g/day for 1-2 year olds and 89 g/day for 4-5 years olds. It provided 27-36% of the daily protein intake and 37-44% of the daily intake of calories for both age groups. Tortilla intakes by preschool children of various socioeconomic levels in Costa Rica were published by Menchú et al. (1973) (3). The intake varied from 12-15 g of tortillas/day, with rice and wheat bread being more important. Martorell et al. (1979) (4) and Valverde et al. (1979) (5) reported the data shown in Table 11. This study was conducted before and after an intervention in which there was a free distribution of corn and beans at the sites studied.

For the head of the family intakes were 425 g and 503 g/day before and after the intervention respectively. For women it was 272 and 342 g/day representing an increase of 69 g. In children 0-23 months of age intakes were 44 g. Intakes increased with age to 155 g (48-72 m) and also after the intervention. For the total of 40 children in the study, tortilla intake increased from 93 g/day to 125 g/day after the intervention. Although corn consumption was already high in all age groups studied, the corn and bean intervention resulted in a significant increase in consumption levels. Additional data were reported by García and Urrutia (1978) (6) and Urrutia and García (1978) (7) and are shown in Table 12. Three-year-old children from Santa María Cauqué, Guatemala, showed an intake of 226 g per day. Expectant mothers (3rd trimester of pregnancy) consumed 595 g, and lactating mothers (1st trimester) 660 g. These figures are quite high as compared with the values given by Martorell et al (4). In children 6 to 11 months of age in Santa María Cauqué, tortilla intakes varied from 4 g/day at 6 months to 23 g/day at 11 months. These data show that lime-treated corn is a very important food item for all family members in Guatemala. The same situation is likely to be found in all other countries in Central America where corn intakes are high. From the point of view of nutrition, much could be

achieved if the corn consumed was of a better nutritional quality. This may now be possible due to the greater availability of industrially-produced nixtamalized corn flour.

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Table 8
Lime-treated corn intakes and of other cereal grains in rural and urban populations in Central America - 1969
g/day

Country	Area	Corn	Rice	Wheat**	Other
Guatemala	R	496	16	40	11
	U	157	27	124	15
El Salvador	R	533	27	26	6
	U	249	55	66	5
Honduras	R	340	29	20	5
	U	203	50	83	7
Nicaragua	R	199	54	35	7
	U	84	80	51	7
Costa Rica	R	62	100	66	0
	U	21	103	96	4
Panamá	R	6	186	47	29
	U	6*	150	74	7

Reference (1)

*Corn meal

**Wheat products (bread, pasta, etc.)

Table 9
Lime-treated corn consumption and its contribution to daily energy and protein intakes
of children in a rural area of Guatemala - 1970

		Protein intake			Energy intakes		
Age (yrs)	Intake (g/day)	Corn (g/day)	Total (g/day)	% from corn	Corn (kcal/day)	Total (kcal/day)	% from corn
1-2	64 ¹ (97) ²	5.4	20.0	27	231	699	33
2-3	86(130)	7.3	21.7	34	310	787	39
3-4	120(185)	10.2	27.9	36	433	981	44
4-5	89(194)	7.5	23.3	33	321	819	39

¹ Dry grain.

² Tortilla (in parenthesis).

Flores, M. et al. (2).

Table 10
Tortilla intakes in preschool children in rural Costa Rica by socioeconomic level
g/children/day - 1973

	Socioeconomic groups		
Cereals	Low	Middle	High
Rice	39	45	22
Wheat bread	24	35	25
Pasta	4	4	3
Corn*	15	12	13
Other	10	4	4

*As tortilla
 Menchú et al. (3).

Table 11				
Corn consumption ($\bar{x}\pm 50$) before and during the intervention				
- 1979				
g/day				
Group	n	Before	After	Change
Adult				
Father	45	425±132	503±161	78 ^d
Mother	47	272± 68	342± 90	69 ^d
Children				
Age, months				
0 - 23	15	44±55	75±65	31 ^c
24 - 47	12	87±49	108±73	21 ^b
48 - 72	13	155±88	198±110	43 ^c
Total	40	93±66	125±84	32 ^d

Martorell et al. (4)

Valverde et al. (5)

b,c,d = Paired t-test significant at 5, 1 and 0.1 percent respectively.

Tale 12	
Lime-treated corn intakes in rural areas in Guatemala - 1984	
Guatemala	g/day
Three-year-old children (n=31) ⁶	226
Women's 3d.trimester pregnancy (n=50) ⁶	595
Mothers 1st trimester lactation (n=50) ⁶	666
Children ⁷ :	
6 m	4
7 m	6
8 m	9
9 m	16
10 m	19
11 m	23

Reference (6, 7).

Contribution of Nutrients from Tortilla to Total Intake

As shown in Table 8, the consumption of lime-treated corn is high in Guatemala, El Salvador and Honduras, lower in Nicaragua, and still lower in Costa Rica, particularly in urban areas. As can be seen from the data in Tables 13 and 14, lime-treated corn is an important source of nutrients to the diet of these countries. The figures in Table 13 represent the percent contribution from all cereals (corn, wheat, rice) to the total nutrient intake. The other nutrient sources of importance are animal derived food products and beans (for most nutrients) and sugar (for calories). Table 14 shows the percent of lime-treated corn foods relative to the total cereal grain intake, for both rural and urban areas. In the rural sector, lime-treated corn is the most highly consumed cereal grain in El Salvador, Guatemala, Honduras and Nicaragua, with consumption by country in that order. In Costa Rica, rice is the cereal of choice. In the urban sector, lime-treated corn is also of importance, although other cereal grains, particularly rice, make an important contribution to total intakes of nutrients.

In the past, emphasis was placed on the dietary calorie and protein contribution of lime-treated corn products, and little mention was made of other nutrients. Yet lime-treated corn products provide some amounts of Ca, and vitamins B₁, niacin, and even B₂. Animal food products contribute as much B₂ as lime-treated corn, followed by beans.

Table 13
Nutrient contribution of corn products (cereals) to the average intake of nutrients -1969

	Guatemala		El Salvador		Honduras		Nicaragua		Costa Rica	
	R	U	R	U	R	U	R	U	R	U
Energy	65.0	50.5	61.8	44.1	49.5	44.2	41.5	38.5	39.3	36.7
Protein	52.6	38.3	50.1	35.2	39.2	35.4	30.9	29.5	32.7	27.1
Ca	68.8	82.2	62.2	79.6	45.3	32.3	25.8	14.9	16.8	8.8
Fe	50.7	39.5	22.0	22.8	34.5	28.3	39.2	26.0	31.9	20.3
Vit A	15.4	1.5	1.2	0.5	10.0	0.2	0.9	0.4	1.2	0.4
B 1	61.9	43.9	54.9	39.3	44.2	40.0	32.3	33.7	34.3	31.6
B 2	36.1	15.9	30.7	14.7	22.9	16.5	19.3	15.4	11.6	7.9
Niacin	54.0	37.8	55.6	40.9	43.5	38.6	38.9	41.5	40.0	33.5
Vit C	0	0	0	0	0	0	0	0	0	0

R = Rural; U = Urban

Table 14					
Intakes of lime-treated corn versus rice intakes in the countries of Central America 1969					
		Cereal intakes, g/country/day			
Country		Rural		Urban	
		g	%	g	%
Guatemala	Total	563		333	
	Tortilla	496	88.1	157	47.1
	Rice	16	2.8	27	8.1
El Salvador	Total	542		375	
	Tortilla	533	98.3	249	66.4
	Rice	27	4.6	55	14.7
Honduras	Total	394		343	
	Tortilla	340	86.3	203	59.2
	Rice	29	7.4	50	14.6
Nicaragua	Total	309		248	
	Tortilla	199	64.4	84	33.9
	Rice	54	17.5	80	32.3
Costa Rica	Total	228		224	
	Tortilla	62	27.2	21	9.4
	Rice	100	43.9	103	46.0

THE NIXTAMALIZATION PROCESS

In Mexico and in some of the countries in Central America, the transformation of corn (*Zea mays*) into masa and edible products such as tortilla, tamales, tamalitos, tacos, and other forms is accomplished by cooking the whole grain in water to which lime is added. This operation is called nixtamalization and the product obtained is called nixtamal. The industrial process as practiced today is an adaptation of the traditional home process as carried out by the rural populations of these countries.

The Original Technology

The process has been described with some detail by a number of researchers (1-5). As shown in Figure 1, it involves cooking of the corn grain in water at a ratio of 3 parts water to 1 part corn. To this blend variable amounts of lime are added, up to 1.2% based on corn weight. Cooking at boiling temperatures continues for up to 60 minutes. The cooked corn is allowed to cool to ambient temperature and to soak in its cooking liquor for 8-14 hrs. After the soaking period, the cooked corn is washed 3 to 4 times in clean agitated water to remove the seed coat of the kernel, the excess lime and any solids which leached out of the kernel or parts of the grain. The creamy yellow liquor is discarded. The cooked corn or nixtamal, containing around 55% moisture, is ground either by hand or with kitchen disc mills, or motor driven stone mills into a dough (masa), sometimes with the aid of small amounts of water. The moisture content may go as high as 61% and the pH to around 7.6. The masa is then kneaded to a limited extent and variable portions are taken to be patted flat and placed over a hot surface made of clay or metal to be cooked for 3 minutes on each side usually about 3 times, so one surface is cooked twice. During this operation, the volume and surface area increase due to vapor expansion within the structure. The temperature of the hot plate varies between 180 °C on the edges, to 220 °C in the middle closer to the source of heat. To facilitate the removal of the already made tortilla, the hot plate is sprayed by hand with lime water or the hand is slightly wetted for the tortilla to attach to it. The tortillas are stacked one over the other in

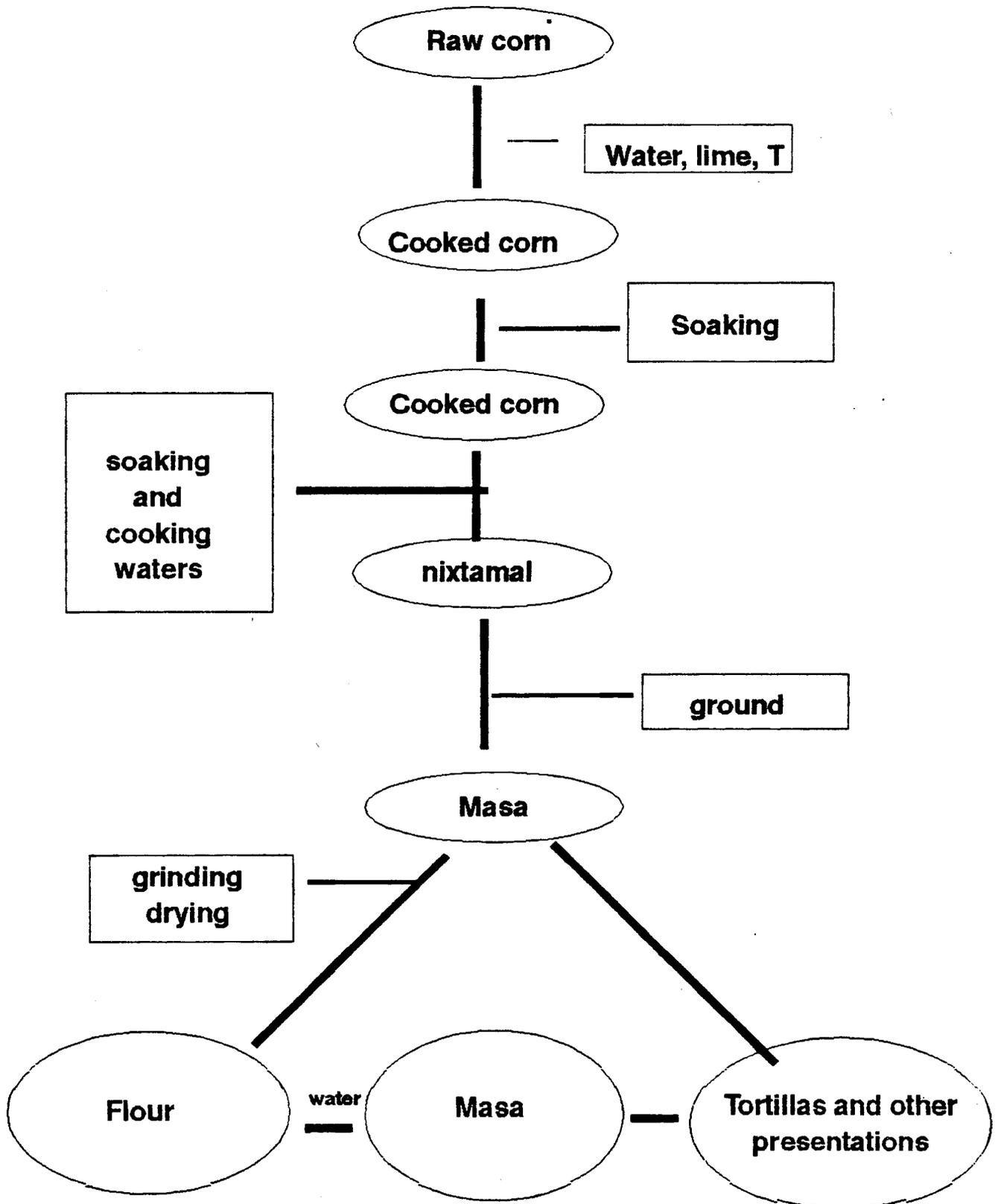
a small basket covered by a cloth where they remain hot, flexible and moist. The process described above is conducted on a daily basis at rural and urban homes and at commercial tortilla-making enterprises, using hand labor or machines.

With the availability of dry masa flours in the market, the cooking, soaking and grinding operations are eliminated; the flour is mixed with about 1.2-1.3 parts of water to 1 part of flour. These two ingredients are blended to make the dough, allowing some 15-20 minutes for the water to equilibrate before making the tortillas as indicated above. In general, 1 kg of whole clean and sound corn yields 1.3 - 1.4 kg of tortillas, with around 45% moisture content.

The Industrial Process

For all practical purposes, the industrial process to produce masa flour is based on the original nixtamalization process. The flow diagram is shown in Figure 1. Semi-hard or hard textured corn is stored in silos at an appropriate T (temperature) and RH (relative humidity) to keep a 10-12% moisture content. For processing, the grain goes through a cleaning process which usually gives a loss of 3-4%. Once clean water and lime are added (1:1.8; 5 - 6%), it is subjected to the nixtamalization process for about 30-50 minutes. It is then allowed to soak for some 60-120 minutes, after which it is washed. The cooking and washing waters are discarded and the cooked grain (nixtamal) is ground and simultaneously dried. It is allowed to cool and is ground again with classification of the flour with respect to particular particle size. It is then packaged, and stored for distribution. With sound hard or semi-hard endosperm corn, the yield of nixtamalized flour varies between 900 to 950 g per 1 kg of raw corn at equal moisture contents.

THE NIXTAMALIZATION PROCESS OF CORN



Other Processes

There is much interest for economic and environmental reasons to use other processes for corn nixtamalization. Present industrial technology employs large amounts of water, which is later discarded with its load of organic solutes and calcium. Energy consumption is high, as the lime-cooked corn contains 50% moisture, which has to be reduced to 10-12% by heated air or by other processes. To this date none of the new technologies proposed has been completely adequate. A promising new technology is extrusion-cooking, which still needs some additional research and development (6).

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con harinas instantáneas obtenidas por extrusión continua. *Arch. Lat. Amer. Nutr.* **41**: 315-319, 1996.

Changes in Nutrient Content

As indicated previously, the conversion of corn into dough and then into tortillas is a process affected by the type of corn used, as well as other variables, such as water, lime, heat treatments, washing, steeping times, grinding and final cooking. All these factors induce changes in the structure of the grain and influence functional characteristics, chemical composition and nutritive value (1,2).

The chemical composition of the tortilla taken from Food Composition Tables is shown in Table 15 (3,4,5). The figures are very similar to values published from various laboratories in spite of the fact that the tortillas were produced with different varieties of corn, varying processes and equipment, and both at home level and industrially. Figures for the macro-nutrients are relatively abundant with less information on dietary fiber and mineral content and little with respect to vitamins, for which more values should be obtained. Fatty acid contents as well as amino acid content are also available (1,2,6). Chemical composition values for U.S. products have also been published (7, 8).

Only a few studies on how the process affects the chemical composition of corn tortillas are available. Information on this topic has been published by Bressani et al. (9), Cravioto et al. (10), Serna-Saldívar et al. (11) and recently, by Weber et al. (12), and Gómez-Aldapa et al. (13) on extrusion-cooked flours.

Table 16 shows the changes in macro-nutrients which take place in white and yellow corn as processed by rural people in Guatemala (9). As indicated, no major change takes place in the macro-nutrients, with the exception of low values in fat soluble substances in the dough and tortilla, with respect to the raw grain. Others have reported similar changes (10,11).

The changes in thiamine, riboflavine, niacin and carotenes during the lime-cooking process are shown in Table 17 for white and yellow corn (9). Thiamine losses varied from 48.9 to 68.8% from corn to tortillas for white corn, and from 54.2 to 64.9%

for yellow corn. Most of the losses took place during nixtamalization. Riboflavin losses varied from 38.5 to 49.6% in white corn and from 16.7 to 18.4% in yellow corn, from corn to tortilla. These losses also took place during the wet alkaline cooking operation. Niacin losses varied from 16.1 to 20.4% for white corn and from 8.5-12.3% in yellow corn, all of the loss occurring during the wet alkaline cooking. Carotenes in yellow corn also decreased, but most of the decrease took place in the baking step. Cravioto et al. (10) found similar results. Recently Gómez-Aldapa et al. (13) have reported relatively high vitamin losses due to alkaline processing of corn by extrusion-cooking as shown in Table 18. Thus, it would be of interest to restore the B-vitamin content as is done for wheat flour due to the losses which take place during milling of the grain.

With respect to mineral content, most studies show a small increase in total ash, which is due to the calcium retained during cooking. There are other changes in mineral content as shown in Table 19, in samples of tortillas and corn collected at the rural level in Guatemala (14). Besides an increase in Ca, there are small increases in Fe, Cu and Zn. The reasons for such increases are not clear. Bressani et al. (14) suggest that the increase may be the result of the use of lime (which is not pure $\text{Ca}(\text{OH})_2$), and of the utensils used to grind the cooked corn. Krause et al. (15), who reported values of 1.9 to 6.6 mg/100 g of tortilla (dry weight), suggests the process of making tortillas, rather than the type of corn used, is probably the more important determinant of tortilla mineral content. Recently Weber et al. (12) showed that the mineral content of tortillas varied with the source of the samples. Values for Fe varied from 1.0 to 3.2 mg/100 g, on a wet weight basis, with an average of 1.7 mg/100 g. Although the mineral amounts are variable and increases may be observed, no information is reported on their bioavailability.

With respect to dietary fiber (DF) content, values from various workers (6,11,12,14) are shown in Table 20. Insoluble DF decreases from corn to the dough, followed by a small increase from the dough stage to tortilla. On the other hand, soluble dietary fiber increases to a small extent from raw corn to masa and tortillas; therefore total dietary fiber (TDF) is slightly lower in tortillas as compared to raw corn, with lower values in the dough stage suggesting the formation of dietary fiber during baking.

The lime-cooking process induces other changes which are of nutritional interest.

One change which is of significance is the decrease in phytic acid content. Recently Urizar and Bressani (16) showed a 35% decrease associated with the amount of lime used for nixtamalization. In raw corn, phytic content averaged 1.03% while the nixtamal cooked with 1.2% lime showed a value of 0.75%. Values reported by various workers (13,17,18,19,20) are shown in Table 21. The variability is relatively large, which could be due to the variety of corn, or to the alkaline processing conditions used, and possibly to the analytical method used. This is an area which deserves additional attention.

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Table 15					
Chemical composition of tortillas*- 1961					
	Central America		Latin America	Mexico	
	Yellow	White	white	yellow	White
Corn color	Yellow	White	white	yellow	White
Moisture, %	47.8	47.8	53.9	47.5	42.4
Energy, kcal	206	204	192	214	224
Protein, %	5.6	5.4	4.2	4.6	5.9
Fat, %	1.3	1.0	4.4	1.8	1.5
Carbohydrate, %	44.5	44.9	36.9	45.3	47.2
Ash, %	0.8	0.8	0.6	ND	ND
Fiber, %	ND	ND	0.6	2.09	4.47
Calcium, mg/100 g	158	124	198	196	108
Phosphorus, mg/100 g	122	123	111	ND	ND
Iron, mg/100 g	2.5	0.2	4.0	2.6	2.5
Thiamin, mg/100 g	0.12	0.10	0.04	0.15	0.17
Riboflavin, mg/100 g	0.05	0.04	0.04	0.05	0.08
Niacin, mg/100 g	1.02	1.02	0.5	1.0	0.9
Vitamin C, mg/100 g	0	0	0	0	0
Retinol equiv., mcg/100 g	15	2	5	20	2

* From Food Composition Tables

ND - Not given in Tables

Table 16				
Changes in macro nutrient content during corn nixtamalization, g/100 g				
Nutrient	Corn	Nixtamal	Masa	Tortilla
Moisture	15.9	48.6	60.5	47.8
Protein*	9.6	10.3	10.4	10.3
Fat*	5.7	3.9	4.0	2.0
Crude fiber*	1.9	1.4	1.2	1.4
Ash*	1.5	1.5	1.6	1.6
Carbohydrate*	81.3	84.2	82.8	84.8

*Dry weight basis
Bressani et al. (9)

Table 17				
Losses of vitamin content of corn processed by nixtamalization, %				
Loss of vitamins from raw corn to:				
Nutrient	Raw corn, mcg/g	Nixtamal	Masa	Tortilla
Thiamin ¹	3.84	40.6-66.2	40.1-61.4	48.9-68.2
Riboflavin ¹	1.14	34.8-1.5	37.8-45.9	38.5-49.6
Niacin ¹	20.00	11.0-16.0	10.1-25.9	16.1-20.4
Thiamin ²	4.78	52.8-57.7	49.3-67.1	54.2-64.9
Riboflavin ²	1.00	19.2-28.1	15.8-21.9	16.7-18.4
Niacin ²	18.98	8.6-14.8	16.6-21.3	8.5-12.3
Carotene ²	3.03	0 - 11.0	0 - 14.2	26.1-34.5

¹ White corn

² Yellow corn

Bressani et al. (9)

Table 18 Losses of vitamins during nixtamalization of corn by extrusion-cooking - 1996	
Vitamin	% loss
Thiamin	53
Riboflavin	30-39
Niacin	66.7
Pyridoxine	45
Folic acid	28-43

Gómez-Aldapa et al. (13)

Table 19 Changes in mineral content mg/100 g (d.wt.b.)			
Mineral	Corn	Tortilla	Change %
Phosphorus	299.6±57.8	309.0±34.2	+3.1
Potassium	324.8±33.9	272.9±16.0	-16.0
Calcium	48.3 ± 12.3	216.6±41.5	+348.4
Magnesium	147.9± 9.9	123.1±15.4	-16.8
Sodium	59.2± 4.1	71.2±7.9	+20.3
Iron	4.8 ± 1.9	7.0±7.4	+45.8
Copper	1.3 ± 0.2	2.0± 0.5	+53.8
Manganese	1.0 ± 0	1.0 ± 0	0.0
Zinc	4.6 ± 1.2	5.4 ± 0.4	+17.4

Bressani et al. (14)

Table 20
Changes in dietary fiber taking place in the lime-cooking of corn to tortilla
(g/100%) (d.wt.b.)

Product	Dietary fiber		Total
	Insoluble	Soluble	
Raw corn	13.05	0.99	13.80
Masa	6.32	1.75	7.64
Tortilla	6.87	1.74	8.63

Bressani et al. (6).

Table 21 Phytic acid content, corn masa or tortilla % (d.wt.b.)	
0.5 - 0.6	Reinhold & García, 1978
0.95	Poneros & Erdman, Jr., 1988
0.565	Martínez-Torres et al., 1987
0.488±40	Wyatt & Triana-Trejos, 1994
0.76 - 0.24	Gómez-Aldapa et al.1996
0.739	Urizar & Bressani (to be published)

THE NIXTAMALIZATION PROCESS OF CORN AND SPECIFIC NUTRIENT

BIOAVAILABILITY

The nixtamalization process to convert corn into an edible cooked product causes losses of carbohydrates, nitrogen-containing compounds, and vitamins. However, it also induces some changes of nutritional benefit.

Calcium. Due to the use of lime during the cooking and soaking process, the cooked corn absorbs and retains significant amounts of calcium. Most of it remains in the germ, with lower amounts in the endosperm (1). Bioavailability studies with experimental animals conducted in 1966 (2) showed masa calcium to be 83.4 - 90.1% available as compared to 91.8% for milk calcium Table 22. Higher levels of bioavailable Ca were obtained with lime-treated corn supplemented with lysine and tryptophan to improve its protein quality. Other workers have confirmed the above results. Serna-Saldivar et al. (3,4) found only 59.0% of the Ca in raw corn to be bioavailable, but this climbed to 86.7% in tortillas. For Quality Protein Maize (QPM), bioavailability of Ca was 75.0% in raw corn and 89.4% in the tortilla. QPM is a corn variety with a high lysine and tryptophan content, and with an overall better essential amino acid balance than common corn. Serna-Saldivar et al (4) also reported better bone formation (tibia and femur) in animals fed on tortillas as compared to raw corn. Calcium intakes from tortillas

improved phosphorus and magnesium retention. Poneros & Erdman Jr. (5) also confirmed the high bioavailability of Ca in tortillas in the presence or absence of ascorbic acid. Therefore, one of the nutritional advantages of the nixtamalization process is the increased bioavailability of calcium in the lime-treated corn flour.

Niacin. During the nixtamalization of corn, niacin losses of around 16 - 20% take place. However, populations consuming high intakes of lime-treated corn do not develop niacin deficiency disease (pellagra). The accepted explanation (6,7) is that the lime-cooking process increases the bioavailability of nicotinic acid (a component of niacin), as documented from studies in experimental animals and human subjects. In addition to the increased bioavailability, during cooking the essential amino acid leucine becomes less available, resulting in a better balance with isoleucine and tryptophan, a precursor of niacin (8).

Iron. According to studies of various workers, the bioavailability of iron from whole corn in several culinary preparations is quite low, less than 2% for healthy persons, and less than 5% for iron-deficient persons. Iron absorption from corn is in general lower than that from other cereals (9). In a study involving 276 subjects, Martínez-Torres et al. (10) found iron absorption from corn to vary from 1.2 to 2.3% for groups of individuals whose absorption from a reference dose was less than 35%. There was an abrupt increase from 2.3% to 4.5% in corn iron absorption between subjects whose absorption from the reference dose was about 30.0% - 34.9% and those with absorption of 35.0 - 39.9% ($P < 0.02$). These same authors reported iron absorption from “precooked maize flour arepas” to be $5.8 \pm 1.2\%$; and $6.7 \pm 1.2\%$; from “polished corn arepas” to be $5.5 \pm 1.2\%$; from “whole maize arepas” to be $3.6 \pm 1.2\%$ and $4.0 \pm 1.3\%$; and from “whole corn tortillas” to be $2.8 \pm 1.2\%$, for a total of 22 and 13 subjects in two studies, respectively (Table 23). They further found iron absorption to be highly associated with the level of phytic acid in the different corn preparations, which ranged from 120 mg/100 g in the “precooked maize flour arepas,” 213 mg/100 g in the “polished corn arepas,” 318 mg/100 g in the “whole corn arepas”, and 565 mg/100 g in the “whole corn tortillas.” The phytate content of the whole corn grain, of the polished corn and of the precooked corn flour was 772, 557 and 251 mg/100 g respectively.

Wyatt and Triana-Trejos (11) found corn tortillas from various locations in Mexico to average 488 ± 4.0 mg/100 g dry weight of total phytate of which 68.04% was insoluble. These workers reported average values for iron of 4.06 ± 1.04 mg/100 g dry weight and for zinc of 2.55 ± 0.42 mg/100 g (d.wt), of which 73.69% was insoluble iron and 69.93% was insoluble Zn. Recently Urizar and Bressani (12) reported phytic acid to decrease upon cooking corn with various levels of lime. Using in vitro assays, they found iron absorption from lime-cooked corn to increase to levels similar to those reported by Martínez-Torres et al (10) for human subjects.

During processing of corn by nixtamalization, a change in the amount of Total Dietary Fiber (TDF) has been reported (13,14). Dietary Fiber (DF) has been shown to bind iron. Reinhold et al (15,16) reported that the Neutral Detergent Fiber (NDF) present in tortillas could bind up to 0.3 mg of Fe per gram of NDF at a pH of 6.45. Likewise, García- López and Wyatt (17) indicated that the iron in a corn/bean diet is around 50% less available, as compared to ferrous sulphate used as reference. The association between DF and iron availability in lime-treated corn was also reported by Reinhold et al (1981) (16), and García-López & Wyatt (1982) (17). There do not appear to be any published studies of the effect on iron availability of cooking the dough into tortillas, although some effect may be inferred from the changes in DF which take place. An additional concern is the possible inhibitory effect of calcium intake from tortillas on nonheme- and heme-iron absorption as reported by Hallberg et al. (18, 19). This is an area which should be studied in the case of lime-treated corn tortillas.

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Table 22			
Biological utilization of calcium from tortilla			
	Absorption % of intake	Retention % of intake	Reference
Corn	78.6±4.6	77.0±4.6	Braham & Bressani '66 (2)
Tortilla	85.4±4.6	83.6±4.6	Braham & Bressani '66 (2)
Milk	97.0±4.4	91.8±1.2	Braham & Bressani '66 (2)
Maize + Lysine	80.7±3.5	79.3±3.4	Braham & Bressani '66 (2)
Tortillas + Lysine	89.8±3.0	87.8±3.7	Braham & Bressani '66 (2)
Milk	96.5±0.6	94.5±1.2	Braham & Bressani '66 (2)
Corn grain	63.2	59.0	Serna-Saldívar et al. '91 (2)
Tortilla	86.7	85.2	Serna-Saldívar et al. '91 (2)
QPM grain	75.0	75.0	Serna-Saldívar et al. '91 (3)
QPM tortilla	91.9	89.4	Serna-Saldívar et al. '91 (3)
Relative Biological value			
Tortilla	93%		Poneros & Erdman, Jr. '88 (5)
Tortilla + Ascorbic acid	98%		Poneros & Erdman, Jr. '88 (5)
NFDA	95%		Poneros & Erdman, Jr. '88 (5)

Table 23			
Absorption of iron from various kinds of maize food products (%)			
	Study 1	Study 2	Phytate content, mg %
Whole maize tortilla	2.8±1.2	-	565
Whole maize arepa	4.0±1.3	3.6±1.2	318
Precooked maize flour arepa	5.8±1.2	6.7±1.2	120
Polished maize arepa	-	5.5±1.2	213

Martínez-Torres et al.(10)

PHYSICAL CHARACTERISTICS OF TORTILLAS

The chemical composition of whole corn tortillas is shown in Table 24 for Latin America and Mexico (1,2). Nutrient contents have also been published by Ranhotra (3), Saldana & Brown (4), Serna-Saldívar et al. (5), Grijalva-Haro et al. (6), Weber et al (7) and Bressani et al. (8). There is a high similarity among all the analyses; the differences may be due to the corn variety and to the processing methodology, which ranges from rural practices to mechanical preparation of the tortilla.

As indicated previously, the tortilla is a flat cake made from lime-treated corn by cooking it over a clay or metal surface at temperatures between 180-220 C for about 6 minutes. There are several methods to prepare the flat cakes, which include kitchen-ware, machines, and by hand. The latter is at present the most common way. As shown in Table 25, there are significant variations in tortilla physical characteristics. Most workers (9,10,11) have reported significant differences in tortilla size between communities. Usually tortillas made at the rural level are heavier and larger than tortillas made at urban homes or urban areas, which tend to be smaller. Likewise, machine-made tortillas are not as thick as those made by hand. Tortilla size is also different among the lime-treated corn consumers in other countries in Central America and Mexico (12). Weber et al. (7) reported values from 19±3.4 to 22± 2.3 g in Arizona. Tortilla size should be taken into consideration to improve accuracy in quantitative studies of dietary intakes.

In specific regions of Guatemala, the tortilla is replaced by the tamalito, which is a small cake weighing around 86 grams. This food item is made by placing the dough into the husks of the ear of corn and placing the whole unit into hot water to gelatinize all the starch. It is a lime-treated corn preparation very popular in urban centers and is sold by street food vendors. The chemical composition and nutritive value of this dough preparation is the same as that of the tortilla. The number of tamalitos consumed per day varies from 15.8 ± 1.7 in urban areas to 12.7 ± 1.0 in rural areas (10,11).

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Table 24		
Chemical composition of corn tortillas in 100 g		
Nutrient	White corn*	White corn**
Energy, kcal	204	224
Moisture, g	47.3	42.4
Protein, g	5.4	5.9
Fat, g	1.0	1.5
Fiber, g	-	4.5
Ash, g	0.8	-
Carbohydrates, g	44.9	47.2
Calcium, mg	124	108
Phosphorus, mg	123	111
Iron, mg	0.2	2.50
Thiamine, mg	0.10	0.17
Riboflavine, mg	0.04	0.08
Niacin, mg	1.02	0.90
Vitamin C, mg	0	0
Retinol equiv., mcg	2	2

* Food Composition Tables for Use in Latin America.

** Muñoz de Chávez, M., A. Chávez Villasana, J. A. Roldán Amaro, JA. Ledesma Solano, E. Mendoza Martínez, F. Pérez-Gil Romo, S. L. Hernández Cordero, A. G. Chaparro Flores. *Tablas de Valor Nutritivo de los Alimentos de Mayor Consumo en México*. 1a Edición, enero 20, 1996. México D. F., México.

Table 25					
Some special characteristics of tortillas in Central America					
Country and area	Preparation	Weight, g	Diameter, cm	Moisture, %	Reference
Rural (white)	hand	45.7±1.2	11 - 17		Valverde et al. (9)
Urban (white)	hand	27.4±4.2	10.7±5.2	48.4±5.2	Krause et al. (10)
Semi-urban (white)	hand	40.9±2.2	12.2±0.2	-	Krause et al. (10)
Rural (white)	hand	53.1±1.8	13.7±0.2	-	Krause et al. (10)
Rural (yellow)	hand	26.1±2.8	10.0	42.5	Bressani (11)
Rural (white)	hand	23.3±2.5	10.8	43.5	Bressani (8)
Rural (white)	hand	41.9±2.0	11.9	50.3	Bressani (11)
Rural (white)	hand	41.3±3.4	11.7	47.4	Bressani (11)
Rural (white)	hand	23.5±3.9	9.9	41.4	Bressani (11)
Urban (white)	machine	28.8±1.2	14.2±0.2	47.5	Bressani (11)
		Small	Medium	Large	
Guatemala	hand	37 g	44 g	55 g	Menchú (12)
Nicaragua	hand	78 g	96 g	128 g	Menchú (12)
Costa Rica	hand	18 g	25 g	40 g	Menchú (12)

¹ Guatemala

CHEMICAL, PHYSICAL CHARACTERISTICS AND SHELF-LIFE OF LIME-COOKED CORN FLOUR

The chemical composition of lime-treated corn flours from Guatemala and Mexico are shown in Table 26 (1,2). Chemical composition data for corn tortillas has been presented earlier. The data in this table show little variation in the macro-nutrients between the two flours, and some variation in the micronutrients, particularly the mineral content. The pH of the nixtamalized corn flour varies between 7.5 and 8.5 which is of importance during storage, since this retards the development of acidity. Some work has been done to establish physical standards of quality for nixtamalized corn flour for tortilla preparation. Molina et al. (3) evaluated an industrially-produced lime-treated corn flour and a flour produced in the laboratory using drum drying. The criteria included soluble sugars, protein and calcium content as well as total and damaged starch, available lysine, sedimentation number (Zeleny), water absorption (by farinography) and viscosity at 85 °C by amylography. Differences found included a higher sedimentation value in the industrial flour, higher water absorption in the laboratory produced flour and higher viscosity at 85 °C by amylography in the industrial flour. Bedolla & Rooney (4) evaluated six samples of lime-treated corn flour from the US and three from Mexico, from various manufacturers. The criteria used were moisture, protein, total and enzyme-susceptible starch, pH, density, water absorption index and viscoamylography. The study indicated that lime-cooked flour for tortilla production should have a uniform particle size (32% in 60, 70, 80 mesh), a maximum viscosity of 220-330 BU, a pH of 7.2, water absorption of 1.3 cc of water/g of flour at 70 °C, and a white color. Gómez et al. (5) reached the same conclusions.

Very few studies have been published on the keeping quality of lime-cooked corn flour. Paredes-López & Mora-Escobedo (6) studied, with accelerated storage techniques, the stability of corn flours stored at 55, 62, 75 and 83% RH for 10 to 90 days at a storage temperature of 35 °C. The results showed an increase in fat acidity with respect to time, which was greater as the RH increased. Protein solubility tested at 55 and 75% RH

decreased up to 60 days, and remained constant between 60 and 90 days. This observation may be a consequence of the hydrolytic changes in lipids during storage. Available lysine as well as in-vitro protein digestibility and PER decreased with storage time, effects which were more drastic at the higher storage RH values. Sensory qualities of tortillas prepared from the corn flour, such as flavor, color, aroma, texture and consistency tended to decrease with respect to RH, with the flour stored at 75% RH being no longer acceptable at 60 days of storage. These results indicate the importance of the kind of material that is used for packaging.

The effects of storage conditions and packaging materials on the physico-chemical, microbiological, and sensory properties of nixtamalized corn masa flour were reported by Carrillo-Pérez et al. (7). These workers stored the flour for 6 months in paper and polyethylene bags at ambient conditions (36 °C and 29% RH). They found that polyethylene bags had more resistance to tearing and a lower rate of water vapor transfer than paper bags. The masa flour stored in paper bags deteriorated faster than the masa flour stored in polyethylene bags. In this study, the masa flour deteriorated at the lowest rate when stored at ambient conditions. On the other hand, masa flour stored in polyethylene bags or paper bags at 25 °C and 60% RH retained its properties longer than when stored at 40 °C and 60% RH. Carrillo-Pérez et al. (7) indicated that the main factor that affected product shelf-life was the moisture content. Serna-Saldívar et al. (8) have indicated that dry masa flour has a long shelf life, probably up to two years in dry storage. Various additives, such as gums, acidulants and preservatives are used in many masa flours for table tortillas to enhance tortilla texture and shelf-life (5,8,9).

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Table 26 Chemical composition of industrial lime-treated corn flour (in 100 g)		
Nutrient	Central America*	Mexico**
Energy, kcal	365	377
Moisture, g	9.0	7.1
Protein, g	9.3	7.1
Fat, g	3.8	4.5
Fiber, g	-	9.5
Carbohydrate, g	76.3	77.4
Ash, g	1.6	-
Calcium, mg	141	140
Phosphorus, mg	223	238
Iron, mg	3.4	3.9
Thiamine, mg	0.31	0.22
Riboflavine, mg	0.05	0.05
Niacin, mg	2.40	1.30
Vitamin C, mg	0	0
Retinol equiv., mcg	1	1
Sodium, mg	-	1
Zinc, mg	-	2.50

*Tabla de Composición de Alimentos de Centro América.

M.T. Menchú, H. Méndez, M. A. Barrera, L.Ortega.
INCAP. Sept. 1996. (1)

**Chávez et al. (2)

IMPROVEMENT OF THE NUTRITIONAL QUALITY OF LIME-TREATED CORN FLOUR

Experimental studies with protein and micro nutrients

Chemical analysis of lime-treated corn and of biological studies conducted with experimental animals and human subjects, suggest that the quality of the protein of nixtamalized whole corn flour is not different from that of raw corn (1).

Due to its high importance in rural diets in Mexico and Central America, many studies have been conducted to address the low protein quality of this flour by adding the limiting amino acid in corn (1,2,3), or with various lysine-rich protein sources, which have been reviewed by Bressani (4). The first approach will only improve protein quality, while the second also increases protein content. Besides the two methods indicated, a third possibility was through the use of QPM (5).

Amino acids. The addition of lysine and tryptophan to lime-treated corn resulted in a significant increase in the protein quality both in experimental animals (3,6) and in human subjects, both children and adults (2). The increase was also evident when the two amino acid supplements were added to mixtures of 87/13 and 70/30 corn and beans (7). The latter findings are of importance since one of the arguments against improving the quality of corn protein with amino acids or with supplementary protein is that the consumption of beans would make up the amino acid deficiencies in lime-treated corn protein, which is not completely true.

Protein. Many workers have published studies on the effects of protein supplementation of lime-treated corn, and have been reviewed by Bressani (4). Table 27 shows a summary of data from Bressani and Marengo (3). All proteins tested, both animal and vegetable, when added in levels between 2.5 and 8.0% increased the protein quality of lime-treated corn. The same type of result was obtained whether the effect was measured through changes in PER or in nitrogen balance (6). Other workers confirmed the above findings for other foods: McPherson & Suh Yon (7) and Green et al. (10) with cottonseed flour; Franz (11), del Valle et al. (12,13), Collins and Sánchez (14), Serna-Saldívar et al (15), with whole soybeans or soybean derived products (16,17). All these

studies indicate that both the protein content and the protein quality of lime-treated corn are significantly improved through protein fortification.

Other supplements. Results have also been published which show that the protein quality of lime-treated corn products can be improved by the addition of high protein content vegetables. This approach is not recommended for industrially-produced lime-treated corn flour due to changes in color; however, it is a very good practice in programs addressing nutrition and food preparation (4).

Addition of cereal grains or pseudo cereal grains such as amaranth and quinoa which are higher in lysine content than lime-treated corn have also been proposed, such is the case of the use of rice and of amaranth (4).

Quality protein maize. The protein quality problem of lime-treated common corn would easily be solved through the use of QPM which has higher levels of lysine and tryptophan in its protein (5) and thus a higher protein quality than common corn. The results of nixtamalization of either high lysine corn or QPM show the process not to affect the protein quality of the masa or of the tortilla as compared to the raw corn (4,5).

Vitamin and mineral addition

Studies on the nutritional effects of adding vitamins and minerals to lime-treated corn are very limited. Table 28 describes experimental results in young growing rats in which the effect was evaluated by the weight gained and by changes in PER. In the first groups of results, the effect of adding riboflavin alone, or with B₁ and niacin, or with other B-vitamins is evident in terms of weight gained. In the second set of results, both riboflavin and thiamine added with lysine and tryptophan to lime-treated corn resulted in a significant increase in both weight gain and PER (3). These effects are also observed when vitamins, minerals and amino acids are added to a basal diet of corn and beans (18). The vitamin and mineral supplemental effects are also observed when they are added to lime-treated corn/cooked bean diets in a proportion 80/20, as shown in Table 29. The results shown indicate a significant effect on growth of rats from the addition of minerals, followed by a smaller but significant effect due to the addition of vitamins; both are important when the quality of the protein is improved through the addition of lysine and

tryptophan (18). In other single-nutrient-omission studies using corn/bean diets, the vitamins which appeared important were vitamin A, riboflavin, niacin, and thiamine, and the minerals, magnesium, zinc and iron (19). Du Plessis et al. (20) through a comprehensive chemical, biochemical and technological investigation found that the addition of 1 mg of riboflavin and 10 mg of niacin/400 g of corn meal was adequate to effectively reduce the incidence of subclinical deficiency of these two vitamins among Bantu school children.

Past and ongoing experiences in the improvement of the nutritional quality of lime-treated corn

Guatemala

Studies of lime-treated corn intakes by human population in Central America, its contribution of nutrients, method of processing, forms of consumption and nutritional limitations of the tortilla, have led to the development of a fortifying mixture, the effects of which when added to lime-treated corn were tested in Santa María Cauqué in 1972. Although the fortifying mixture contained thiamine, riboflavin, niacin, iron and vitamin A (Table 30), the study focused on the impact of the protein supplement. The logistics of fortification of the nixtamal (home-treated corn) at the time of milling were complex; however the problems were solved as described by Urrutia et al. (21). The study ended in February 1976 due to an earthquake which destroyed the village where the study was being conducted. The findings of this study were as follows:

1. There was no difference in the intake of calories and protein of preschool children in the high and low fortification groups up to the age of 3 years.
2. There was no apparent impact of the fortification on birth weight (fetal growth)
3. Maize fortification had no significant effect upon physical growth and bone development
4. There was a significant difference in the morbidity and mortality between children with high and low fortification during the second and third year of life, as well as a significant reduction in preschool mortality in the group of children consuming

the highly fortified masa.

5. The intervention also had a positive effect during the weaning period (12-30 months of age) when infectious illnesses are more frequent and of longer duration.

Since March 1997, INCAP has been conducting a feeding study in schools of rural areas of Guatemala, in which the mothers have been instructed to add, to each pound of corn, seven tablespoons of a product made from soybean flour 30, corn flour 70, vitamins and ferrous fumarate. No nutritional evaluation has been done yet (22); however, the acceptability of the nutrient-fortified tortilla has been high.

Of the various brands of lime-treated corn flour available in the market in Guatemala only one (Torti-Ya) is fortified with vitamins and iron. The levels indicated in the package are as follows: B₁ 4.8 mg/kg, B₂, 2.9 mg/kg, niacin 35.3 mg/kg and Fe, 30.9 mg/kg. The iron added is in the electrolytic form (23).

Mexico

Interest in fortifying lime-treated corn flour also exists in Mexico, as indicated by Carrión Hernández (24) and Contreras Medellín (25). A fortifying mixture has been developed by the Mexican National Nutrition Institute (INN Salvador Zubirán). This mixture consists of micronized soybean flour, oat and rice flour, and seven micronutrients; vitamin A, vitamin C, riboflavin, niacin, folic acid, iron and Zn. This blend is added at a level of 10% w/w which results in a 10% increase in protein digestibility, a 40% increase in protein content and a 70% increase in protein quality. Furthermore, the sensory qualities improve with respect to texture and elasticity. Carrión Hernández (24), describes a three-month marketing trial selling 2300 MT, in rural stores of Distribuidora e Impulsora Comercial de CONASUPO, S.A. (DICONSA). The cost of the flour was subsidized to 1.10 pesos/kg and the cost of the supplements was absorbed by the manufacturer. The survey indicated the following results:

1. The flavor of the product was different but resembled that of corn
2. The color of the flour was slightly more yellow
3. There was no change in odor

4. The majority of the purchasers indicated that they would buy the enriched flour to be utilized by the family

Blind taste tests were also conducted with two brands, the results of which indicated that 55% preferred the control tortilla and 45% the fortified product.

Contreras-Medellín (25) has described the fortification process, which is a simple mixing operation. To obtain a good distribution of the vitamin and mineral premix, this is blended first as a 1:4 w/w mixture with a portion of the corn flour; and this blend is then added to the nixtamalized flour for a final concentration of 10%. The fortified flour produces a tortilla with a greater retention of water and thus it gives a larger yield of tortillas. The protein content increases (8.4% to 10.5%), with higher levels of lysine and tryptophan. The iron level increases from 11.5 ppm to 44.5 ppm, and there is also an increase in the vitamins B₁, B₂ and niacin as expected. Serna-Saldívar (1995, cited by Contreras-Medellín) (25) showed that an intake of 5 fortified tortillas (150 g) increased the nutrient intake of children quite significantly.

Past and ongoing experiences of lime-treated corn with added iron

Studies on the addition of iron to lime-treated corn have not been done, except as a component of studies of fortifying mixtures, as described in the previous section. The evaluation of its potential impact in reducing iron deficiency has not been measured in lime-treated corn-eating countries except in the reports from Venezuela (26) where precooked degermed non-nixtamalized corn flour is fortified with a mixture containing 1.5 mg/kg of thiamine, 2.0 mg/kg of riboflavin, 20.0 mg/kg of niacin and 20 mg/kg of iron. Based on surveys conducted in Caracas in 1992, when the corn flour was still not enriched, 37% of the population had iron deficiency, which decreased to 16% in 1994. Anemia decreased from 18% in 1992 to 10% in 1994. According to the publication, these are promising results of the impact of the fortification program of corn flour in Venezuela. Brooke (27) reviewed the experiences in the U.S. on the enrichment of corn meals and grits.

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Table 27 Supplementary effect of various protein sources on the quality of lime-treated corn		
Protein source	Level, %	Protein Efficiency Protein
None	-	1.00
Egg	3.0	2.24
Casein	4.0	2.21
Beef	4.0	2.24
Fish Protein Concentrate.	2.5	2.04
Soy protein isolate	5.0	2.30
Soy flour	8.0	2.25
Cottonseed flour	8.0	1.83
Torula yeast	2.5	1.47

Bressani & Marengo (3)

Table 28 Effect of the addition of vitamins, minerals and amino acids on the nutritional quality of lime-treated corn		
Nutrient	Average weight gain g	Protein efficiency ratios
None*	38	0.91
+ Riboflavin	53	1.09
+ Riboflavin + Thiamine	50	1.00
+ Riboflavin + Thiamine + Niacin	54	1.05
+ All B complex	65	1.13
None*	35	0.77
+ Lysine + Tryptophan (=A)	70	1.62
+ Riboflavin + Thiamine (=B)	51	1.30
A + B	136	2.28
All B complex (=C)	54	1.16
A + C	162	2.37
None**	26±2.3	1.09±0.07
+All vitamins (A)	49±4.0	1.52±0.06
+ Minerals (B)	65±4.3	1.41±0.06
+ Lysine + Tryptophan (C)	26±2.5	1.10±0.08
+A + B + C	107±4.9	2.55±0.06

*Only lime-treated corn flour

**Lime-treated corn flour (90%) + cooked black bean flour (10%)
Bressani & Marengo (3).

Table 29
Effect of supplementation of a lime-treated corn (80%) cooked black beans (20%) diet with vitamins, minerals and amino acids

Diet	weight gain, g	PER
Basal diet (B)	26±2.3	1.11±0.07
+ Vitamins	49±4.0	1.55±0.06
+ Minerals	65±4.3	1.94±0.06
+ Lysine + Tryptophan	26±2.5	1.13±0.08
+ Vit + Min	70±2.1	1.91±0.05
+ Vit + A.A.	54±3.4	1.73±0.08
+ Min + A.A.	89±3.3	2.38±0.06
+ Vit + Min + A.A.	107±4.9	2.56±0.06

Table 30 A protein, vitamin and mineral supplement for lime-treated corn flour		
Ingredient	g/100 g	Contribution to 100 g of corn flour, g*
Soy flour (50% protein)	97.5000	7.800000
L-Lysine HCl	1.5000	0.120000
Thiamine	0.0268	0.002196
Riboflavine	0.0162	0.001296
Niacin	0.1930	0.015444
Ferrous orthophosphate**	0.6000	0.048000
Vitamin A 250 SD***	0.0313	0.002504
Corn starch	0.1327	0.010616

Bressani et al. (18).

* When added at a level of 8 grams/100 g.

** As the salt containing 28.1% iron.

*** Containing 75,000 mcg retinol/g.

RECOMMENDATIONS REGARDING IRON FORTIFICATION OF LIME-TREATED CORN FLOUR

Factors in the lime-cooking process for tortilla preparation, which could affect micronutrient bioavailability.

The factors in corn as such, which could interfere with micronutrient bioavailability are no different from those in other cereal grains, (e.g. dietary fiber and phytates). However, the conversion of corn to the edible tortilla is an alkaline process giving a product with a pH of around 7 and retaining relatively high levels of Ca, depending on the amount of lime used for the cooking process.

With respect to dietary fiber, it has been shown by Reinhold & García (1) that neutral detergent fiber (NDF) as well as acid detergent fiber (ADF) increase from masa to tortilla, while hemicellulose decreases. They suggested that differences in fiber concentration of the corn used and not the techniques of preparing masa or of baking were the main source of variation. However, Reinhold & García (1,2) and García & Wyatt (3) indicated that the increase in NDF and ADF that occur during the baking of tortillas is the result of the formation of fiber-like substances that are measured as lignin. These are produced by a non-enzymatic browning reaction between peptides and free amino acids and carbohydrates, particularly sugars. These authors claimed that the NDF fraction from corn tortillas could bind as much as 0.3 mg of iron per gram of NDF at pH 6.45. Bressani et al (4) showed that insoluble dietary fiber (IDF) increases from 8.69% in masa to 9.43% in tortillas, while soluble dietary fiber (SDF) increases from 1.46% to 1.54%, with total DF increasing from 10.15% to 10.97%, results which confirm those of Reinhold and García (1, 2). There is a decrease in SDF and IDF from corn to masa, mainly due to the loss of the seed coat, which has a high content of DF. It is well accepted that fiber binds bivalent metals making these unavailable for absorption. Therefore, it would be of interest to establish the effect of the conversion of dough (masa) to tortilla on iron bioavailability.

Phytates are also known as substances that bind bivalent metals making them unavailable for absorption. Corn, as with other vegetable foods, contains relatively high amounts of phytates. In corn, the phytates are localized in the germ, which contains about 90% of the phytic acid in corn. The results of various workers, shown in Table 21, indicate a relatively high variability in phytic acid content in tortillas. Urizar & Bressani (5) showed that lime cooking of corn reduced the phytic acid content up to 35% from raw corn to masa; the amount of reduction was associated with the level of lime used for cooking which varied from 0 to 1.2%. They further showed using in-vitro techniques, an increase in iron bioavailability. Phytic acid binds calcium to give calcium phytate. One phytate molecule has the capacity to bind 6 calcium molecules so 0.364 mg of calcium would saturate 1 mg of phytic acid. Martínez Torres et al. (6) showed iron absorption in various edible corn foods to be associated to the level of phytic acid, with better absorption in degermed corn flour than in lime-treated corn.

Further studies are thus needed to establish the behavior of phytic acid during the various stages of converting corn into tortilla and on how this affects iron bioavailability.

The lime cooking process produces a flour which has a pH of around 7. This high pH may influence iron absorption since the pH affects the solubility of iron food complexes and added iron salts. The pH of the food is reduced to a pH of 2-3 at the stomach, and it enters the intestine at a higher pH. The bioavailability of iron is then determined by the solubility of the iron complexes which form and the strength of the affinity of the ligands for iron. Thus, the effect of pH on iron bioavailability deserves some attention (7).

The final problem which may be of importance in iron bioavailability is the high content of calcium in masa flours and in tortilla. As was indicated in a previous section, the calcium in masa flours and tortilla depends on the amount of lime used for cooking. Some values taken from the literature are shown in Table 31. Other factors influence the level of calcium in tortillas, such as corn variety, (with soft endosperm grain absorbing more than the hard types), cooking time, soaking time and extent of washing. There is some evidence which indicates that increasing the amount of Ca will reduce Fe bioavailability (7,8). However, using in vitro techniques, Urizar and Bressani (5)

showed that Fe absorption from masa flour increased up to calcium levels of 250 mg per 100 g. Nevertheless, it is possible that iron absorption may be reduced at higher levels of calcium in the lime-treated corn flour, a point which deserves to be studied.

An additional aspect of concern with respect to the bioavailability of natural and added iron to lime-treated corn flour are the foods often consumed with tortillas. Some of these foods, such as animal products and sources of vitamin C, will help iron bioutilization, while some other foods such as beans, vegetables and coffee will reduce iron absorption. Since animal food consumption is low in the Central American rural diets, it is probable that vegetable foods will play an important role in iron bioutilization. Beans are the second most important food item in the rural diets, and beans are known to contain high levels of DF, polyphenolic compounds and phytic acid.

Recommendations regarding the possibility of iron fortification of lime-treated corn

Factors considered: The following are some of the factors considered regarding the possibility of iron fortification of lime-treated corn. We begin with the premise that iron deficiency and anemias, which affect more than 25% of pregnant women and children in the Central American countries (9), are due to Fe deficiency.

1. Consumption of lime-treated corn flour. As documented in the present report, corn intakes in at least four countries in Central America, are quite high, particularly in Guatemala, El Salvador and Honduras, mainly in the rural areas. Most of the corn consumed is processed at the home level; only recently has industrial lime-treated corn flour been used. The consumption of this flour is increasing in the above countries, as judged by the number of brands available in the market. For example, in Guatemala there are four brands available: **Torti-Ya**, a national brand, **Maseca** and **Minsa**, Mexican brands, and **Nixtamasa** a Salvadoran brand. El Salvador has three brands, **Maseca**, **Del Comal** and **Nixtamasa**, and Honduras has seven: **Maseca**, **Tortimasa**, **Sularina**, **Rapimasa**, **Masabrosa**, **Del Maizal**, **Minsa**, all produced at a national level. Thus, it is apparent that the production and use of these flours is likely to increase in the near future. **Maseca** production in Guatemala and Honduras is between 90-105 MT of flour/day in each country (10). Table 32 shows an estimate of the potential need and present

production of industrial NCF. The values for present consumption of industrial NCF are low; however, they are expected to increase for various reasons. First, NCF has only recently been introduced to the market (1994); second, it is a convenient food that is easy to prepare. The cost is still higher than that of raw corn; however, when the cost of labor and of energy (wood) are added, the cost between the two is about the same. Finally, it is expected that fluctuations in price will be minimal for NCF, with respect to the fluctuations for raw corn. Table 33 shows the same kind of data, provided by industry.

2. Tortilla consumption is highly frequent in rural areas and less frequent in urban areas, but in all cases it is consumed on a daily basis.
3. There is no information available on the organoleptic characteristics of iron fortified lime-treated corn flour, however, based on experience with other flours, no changes are expected to take place.
4. No information is available on stability during storage
5. No information is available on iron bioavailability
6. The cost of fortification would not be different from that in other cereal flours, such as wheat flour.
7. The bulkiness of tortillas should minimize the possibility of an excessive intake.

Based on the above, the recommendation would be to fortify lime-treated corn flour with iron and other nutrients.

Possible iron sources to be used

The iron sources commonly used for fortification have been grouped according to their water solubility. Those highly soluble in water include ferrous sulphate, ferrous lactate and gluconate. Their iron bioavailability is high; some are more expensive than ferrous sulphate; however, the most important aspect is the negative effects on food color, taste and stability.

A second group of iron compounds with a limited solubility in water, but soluble in acid media. Ferrous fumarate belongs to this group of compounds. It is being used as an iron supplement in cereal grain flours and is only slightly more expensive than ferrous sulphate. It would be of interest to learn about its behavior when added to an alkaline

food such as NCF.

A third group includes compounds soluble in water, with a low solubility in acid media. This group includes compounds used in many foods, that however, have a relatively low iron availability. They do not alter the organoleptic properties of the food.

An additional group includes compounds such as EDTA (ethylene-diamine-tetra-acetic acid) chelates among which NaFe EDTA is often used. This compound can be used as an iron fortificant with a relatively good iron bioavailability. One disadvantage is its higher cost relative to ferrous sulphate and ferrous fumarate. Furthermore, its iron content is low (14%) which means that more of it has to be used to provide the same amount of iron. This may be convenient in a product such as lime-treated corn flour, which may have high levels of phytates, high levels of calcium and alkaline pH. Its addition would probably not affect the organoleptic properties of the NCF. This compound together with ferrous fumarate can be screened as iron sources as indicated in the following section.

Supplemental iron and iron available from the local diet

From food consumption intakes, estimations were made of the intake of nutrients for the rural and urban diets of Central America. According to these calculations, the iron intake from rural diets varied between 11.6 to 18.2 mg and from 13.3 to 16.3 mg from urban diets. However, only between 13 and 18% is heme iron and the rest is from vegetable sources, to which grain legumes and cereal grain contribute around to 65 to 68%. Ascorbic acid seems to be consumed in adequate amounts. However, the average rural diet is high in phytic acid, polyphenolic compounds and dietary fiber which could reduce the availability of iron.

Recommendations for research needed on iron fortification of lime-treated corn

The following represents a research approach to the problem of iron fortification of lime-treated corn.

1. Sensory trials of fortified lime-treated corn flour.
2. Measurements of bioavailability in vivo in experimental animals.

3. Field trials with fortified corn masa flour.

Sensory trials

Products:

fortified lime-treated corn flour fresh and stored for three months at 25 °C and 35 °C (lowland and highland temperature)

Preparation:

atole (a hot drink, 12 to 15 % solids), cooked in water.

tamalitos (gelatinized flour), heated with steam for 30 to 40 min.

tortillas (moist flour), cooked using dry heat

The first step would be to learn of the effects of the iron source added on the acceptability of the food products listed. In the case of tortillas, the test will be conducted following the pattern of consumption in most homes in Guatemala. Usually tortillas are made once a day to be used for the three meals: lunch, dinner and breakfast. Sometimes they are served 24 hrs later. They are usually warmed for dinner and breakfast.

Based on the above, sensory trials would be conducted with the products listed, prepared with lime-treated corn flours supplemented with one or two levels of ferrous fumarate, for example. If ferrous fumarate were to be added at the level of 0.025 g per 100 g of corn flour, this would contain 7.5 mg of iron per 100 g of flour. From 100 g of flour, 120 g of dough is to be obtained or 4 tortillas of 30 g each. Each tortilla would contain 1.875 mg Fe. On the basis of a consumption of 6 tortillas/child/day, the iron intake would be 11.25 mg/day which is close to the recommended value of 12 mg/day. Tortillas would be made and tested with a panel: fresh, heated at dinner time and heated again at breakfast the following day. In the case of the atole and of the tamalitos, the acceptability trials will be made only once per day.

Measurements of bioavailability

If results from the above tests are poor, lower levels of the iron source will be added, or a new iron source would be tested. If tests are positive, the

bioavailability of the added iron is to be measured, as well as to stability upon storage. If bioavailability studies show poor results by in-vivo tests with experimental animals, the factors responsible would need to be studied. These include the effects of processing to reduce phytic acid levels (lime level, cooking time, soaking time), and the effects of baking the dough into tortillas, as well as the effects of calcium on iron bioavailability.

Field trials

If up to this point all tests were positive, the iron-fortified flour would be tested in field trials for improvement of iron stores in children or adults, before implementation at the industrial level.

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Table 31 Calcium content in corn masa flour and on tortilla	
mg/100 g - dry weight	Author
178 - 214	Cravioto et al., 1945 (11)
228 - 346	Bressani et al., 1958 (12)
150	Serna-Saldívar et al., 1987 (13)
216.7±41.5	Bressani et al., 1989 (14)
209.2±57.3 (Highland corn)	Bressani et al., 1990 (4)
196.1±50.4 (Lowland corn)	Bressani et al., 1990 (4)
210 (QPM)	Bressani et al., 1990 (4)
138	Serna-Saldívar et al., 1991 (15)
195 (QPM)	Serna-Saldívar et al., 1991 (15)
99 - 477 (202±74)	Krause et al., 1992 (16)
362	Weber et al., 1993 (17)

Table 32					
Estimated consumption of industrial lime-treated corn flour					
Country	Total population 6	Population distribution %		Tortilla-consuming population %	
		Rural	Urban	Rural	Urban
Guatemala	9.2	53	47	88	47
El Salvador	5.3	56	44	98	66
Honduras	5.1	56	44	86	59
Nicaragua	3.9	40	60	64	34
Costa Rica	3.0	53	47	27	9

Country	Corn intake g/person		Corn flour MT potential demand			Industrial production MT*
	Rural	Urban	Rural	Urban	Total	
Guatemala	496	157	2128	319	2447	210 (8.6%)
El Salvador	523	249	1550	383	1983	140 (7.2%)
Honduras	340	203	835	269	1104	140 (12.7%)
Nicaragua	199	84	199	67	266	-
Costa Rica	62	21	27	3	30	-

* Produced at national level and imported

Table 33			
Intake of NCF flour with respect to corn intake *			
Country	Corn intake TM/year	Corn intake/person/year kg	Consumption of NCF, %
Guatemala	896,400	83	3.2
El Salvador	407,100	69	5.9
Honduras	429,200	74	7.2
Nicaragua	229,200	68	5.6
Costa Rica	210,800	62	12.8

* Personal communication, Mr. E. Eushner, President HARISA, El Salvador, 1997

II

**FORTIFICATION OF NIXTAMALIZED CORN FLOUR
AND RELATED PRODUCTS WITH IRON AND/OR OTHER
NUTRIENTS**

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INTRODUCTION

Latin American Indians developed the traditional method to process corn into tortillas (nixtamalization). They cooked corn in a boiling water and lime (ash) solution for 5-50 min and steeped it overnight. The steep liquor (nejayote) was discarded. Then, they washed the cooked-steeped corn (nixtamal) to remove excess lime and pericarp. They ground the nixtamal with a pestle and stone into dough (masa). Finally, they flattened the masa into discs to form tortillas that were baked on a hot griddle for 30-60 sec on each side (Bedolla and Rooney 1982).

Modern methods of corn nixtamalization (alkaline cooking)

Modern nixtamal producers in the United States and Latin America cook corn through a commercial process similar to the traditional method (Figure 1). The purpose of nixtamalization is to improve flavor, starch gelatinization and water uptake, and partially remove the germ and pericarp of the corn kernels . Important process variables are cooking time, temperature, kind and concentration of lime, type and frequency of agitation to keep the lime suspended, and the nixtamal washing procedures (Rooney and Serna-Saldivar 1987).

A common procedure for cooking corn uses Hamilton steam kettles, in which the dry corn is added to the water along with dry, powdered lime. Steam is injected until a temperature of 94°C is reached. The cooking water is circulated with a pump, and the corn is stirred to suspend the lime. Alternatively, compressed air and live steam injection are used to maintain temperature and suspend the lime. The corn is held at near boiling for 30-40 min or more. The time to reach boiling (rise time) is critical. Afterwards the cooked corn is steeped for 8-12 hr. Steeping is necessary to distribute moisture and lime uniformly throughout the cooked grain. After steeping, corn is pumped to the washers using a hydraulic transport system or it is dropped into the washers by gravity. Most washers are horizontal rotating barrels or drums that spray the nixtamal with pressurized water that removes the pericarp, lime, and solubles from the cooked corn (Rooney and Serna-Saldivar 1987).

The washed nixtamal is then ground, using lava or aluminum oxide stones that cut, knead, and mash the nixtamal to form masa. Attrition milling is essential. A typical mill contains a stationary stone (4 in. thick and 16 in. in diameter) and another of the same dimensions that is turned by a 30-hp motor that can process up to 1,360 kg/hr. Additional water is often added to the nixtamal during grinding to increase the moisture level in the masa and to cool the stones (Rooney and Serna-Saldivar 1987).

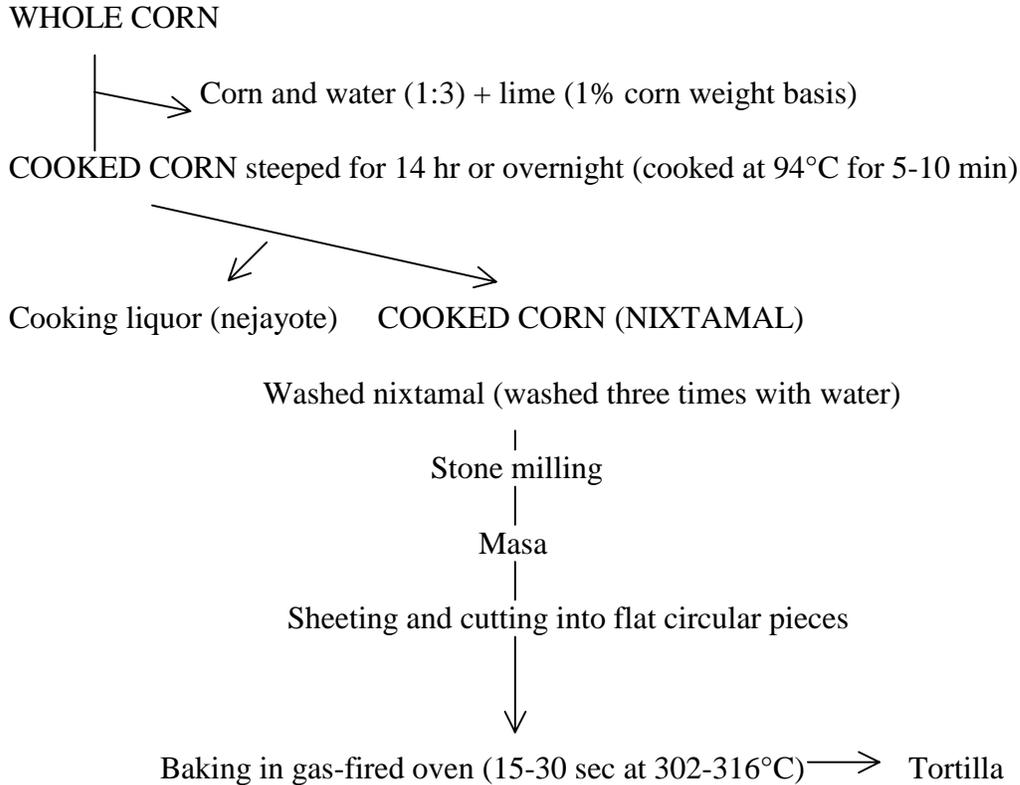


Figure 1. Commercial processing of corn into tortillas.

Tortilla producers are the main masa consumers. They form the masa into tortillas with an automatic sheeter that presses it into a thin sheet of dough that is cut into circular pieces. Finally, a gas-fired oven bakes the tortillas for 15-30 sec at 302-316°C.(Gomez et al.1987).

Corn properties for nixtamalization

Both environment and genetics affect the properties of corn. Industrial nixtamal producers prefer uniformly sized kernels. A low variability in size among corn kernels helps to achieve a more uniform cooking process. They also require a high proportion of hard or flinty endosperm and intact kernels free of fissures or stress cracks. The harder kernels tend to cook more uniformly and retain their integrity during subsequent handling

and processing. Broken kernels or kernels with fissures take up water and alkali more quickly and thus tend to overcook. They also prefer kernels without prominent dents in the crown with easily removed pericarp, and clean yellow or white color. Varieties with deeper dents are generally softer and are more easily overcooked. Overcooking causes excessive dry matter losses and forms masa with a sticky consistency. Kernels with a high pericarp content also produce very sticky masa. The preferred color of corn tortillas is based on regional preferences. However, the cleanest, brightest-colored products are obtained when the corn kernels have a white or yellow uniform color without red or pink shades (Rooney and Serna-Saldivar 1987).

Nixtamalized corn flour (NCF) production

NCFs or instant masa flours need only to be rehydrated to produce masa (1:1 NCF to water ratio). Several companies produce NCF using various procedures based on alkaline cooking and washing of the corn, followed by grinding and drying (figure 2). The dried masa is sieved and reformulated into flours with carefully controlled particle size distributions to meet the various product requirements (Rooney and Serna-Saldivar 1987). The flours used for tortillas need to be finer than the ones used for tortilla chips and taco shells (Gomez et al.1987).

Montemayor and Rubio (1983) described continuous and batch processes for cooking corn to produce NCF. In the continuous processes, the lime (0.6-1.0% based on corn) is mixed with equal parts of corn and water in a large screw conveyor fitted with steam jets. The corn is cooked by steam injection as it slowly passes along a large conveyor. Then, the cooked corn is washed to remove the pericarp and lime, equilibrated (to reach an equal moisture content throughout the kernels) and ground with a hammer

mill. The particles are flash-dried and fed into a sifter, and the appropriate fractions are recombined to provide flours with desired particle size distribution and other properties (Figure 2). Sometimes additional grinding to reduce coarse particles is required. In batch processes, the corn is mixed with lime, cooked, steeped, washed, ground with a hammer mill or stone mill, dried, and formulated into dry masa (Rooney and Serna-Saldivar 1987). Other methods use stone grinding and flash-drying.

Good process control is critical to produce a good NCF (Rooney and Serna-Saldivar 1987). Incorrect particle size and improper drying impart a sandy texture to tortillas. Also, starch gelatinization and retrogradation occurring during processing of corn must be controlled to provide the desired functionality of NCF. This is required for the manufacture of tortilla and snacks with acceptable, consistent quality (Almeida-Dominguez et al.1996).

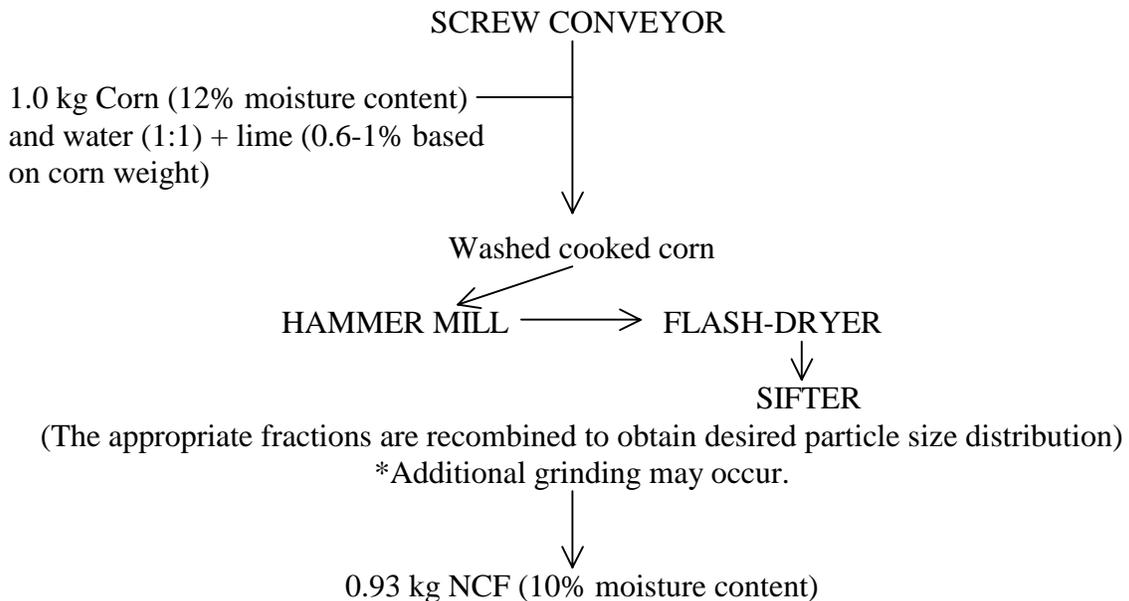


Figure 2. Production of NCF using a continuous process.

Alternate methods to produce NCFs have been proposed. Molina et al.(1977) cooked raw, whole ground corn flour with water (1:3) and lime (0.3% of raw corn weight). The mixture was cooked and dried in a double drum drier with a gap of 0.007 mm and an internal pressure of 110-183 kg/m² and 2-4 rpm. Supposedly, the tortillas produced by drum drying have similar physicochemical and organoleptic characteristics to those of tortillas prepared traditionally. Extrusion puffing has not proven feasible for preparation of NCF, however, the use of continuous cooking without puffing may have potential (Bazua et al.1979). In general the experimental processes do not produce NCFs that are comparable to the best commercial NCF. Subtle differences are important and must be overcome if extrusion or other processes are to be used in NCF production.

The major advantages of using instant masa flours are reduced production times, low equipment costs, and no sewage treatment (there is little or no wastewater). The preparation of masa for tortillas and snacks using NCF requires 30 min, compared with 12-24 hr for cooking, steeping, and grinding corn in the traditional process (Gomez et al.1987). NCFs are available for the production of white and yellow table tortillas, restaurant-style tortilla chips, and tamales. NCF also increases quality consistency of the end product and it can be easily blended with other ingredients (i.e., preservatives, gums, etc.) (Serna-Saldivar et al.1990).

NCF Market

The use of NCF is increasing in popularity because it eliminates the tedious, labor-intensive cooking, washing, and grinding of corn to produce masa for tortillas and snacks. Grupo MASECA is the main NCF producer worldwide (Table 1). MASECA

has 19 plants located in the USA and Latin America. Other companies that produce NCF are MINSA (with 6 plants) and AGROINSA (with 2 plants). In 1994 NCF companies covered 27% of the tortilla market in the U.S. and Latin America (Table 2). Traditional masa production companies cover the rest of the tortilla and related product market (Torres et al.1996). Sales in the corn tortilla industry grew 36% in the United States between 1992 and 1996 according to a survey completed recently by the Tortilla Industry Association, Encino, CA.

Table 1. Main NCF producers worldwide

Companies	
Grupo MASECA	Edinburg, TX (800) 262 7322 Plainview, TX (806) 293 0110
MINSA Corporation	Muleshoe, TX (800) 852 8291 Red Oak, IA (800) 701 5892
AGROINSA	Guadalupe, N.L. Mexico (52 8) 379 94 99 (52 3) 377 8383
Illinois Cereal Mills	Paris, IL (217) 465 5331
Quaker Oats Co.	Barrington, IL (847) 381 1980
Cereal Food Processors Inc.	Kansas City, MO (913) 262 1121

*Other dry milling companies like ConAgra (Atchinson KS 66002) have started to produce NCF to enter the market in the future.

Table 2. Potential market for NCF in the US and Latin America

Country	Potential market (based on a 122 kg/person/year consumption of NCF)
US (30 million Hispanics)	4 million tons/year
Mexico (90 million people)	11 million tons/year
Honduras (5.5 million people)	0.6 million tons/year
Guatemala (9 million people)	1.1 million tons/year
El Salvador (6 million people)	0.7 million tons/year
Costa Rica (3 million people)	0.4 million tons/year

(Torres et al. 1996)

FORTIFICATION OF FLOURS AND OTHER CEREAL-BASED PRODUCTS

Instant corn flours and other corn-based products can be enriched with iron and/or other nutrients such as vitamins, amino acids, and protein sources like soy flour and fish protein concentrate. In the fortification of flour, the required nutrient mixture is mixed with an appropriate diluent to produce a premix, which is accurately metered into the flour (Clarke 1995). The addition of vitamins B₁ and B₂, niacin, iron and calcium to corn flour is similar to wheat flour enrichment, which is a common practice in many developed countries. It is technologically feasible to add other vitamins and minerals as well.

Iron enrichment

Iron compounds

The success of iron fortification depends as much on the fortification compound as on the food vehicle. Fortification with iron is technically more difficult than with other

nutrients because bioavailable forms of iron are chemically reactive and often produce undesirable effects when added to the diet. Since the population will seldom accept the fortified vehicle if the added iron can be detected, inert iron compounds are commonly used, but these are poorly absorbed and have little effect on iron status. A critical step in the design of an iron fortification program is the selection of an iron compound that is both unobtrusive and well absorbed (Cook et al.1983). Table 3 presents the common iron sources and their relative costs.

Table 3. Iron compounds used for fortification (Cook 1983)

Iron compounds	Relative cost*/**
I. Soluble	
1. Ferrous sulfate	1
2. Ferrous fumarate	5
3. Ferrous lactate	7
4. Ferrous gluconate	11
5. Ferric ammonium citrate	7
II. Phosphate	
1. Ferric orthophosphate	6
2. Sodium ferric pyrophosphate	17
3. Ferric pyrophosphate	
III. Elemental	
1. Reduced iron (hydrogen,CO)	1.5
2. Electrolytic	1.5
3. Carbonyl	1.5
IV. Complex	
1. NaFeEDTA	11
2. Hb	

*Incorporates purchase price, percentage iron, and bioavailability. Values expressed relative to ferrous sulfate.

** The relative prices reported in Cook 1983 are still current (i.e. ferrous sulfate = \$ 1.83/lb (Mallinckrodt Baker, Inc. St. Louis, MO), ferrous fumarate = \$ 9.15/lb (Bio-Dar, NY) so ferrous sulfate is reported with a relative cost of 1 and ferrous fumarate with a relative cost of 5).

Soluble compounds

The soluble compounds (ferrous sulfate, ferrous fumarate, ferrous lactate, ferrous gluconate, and ferrous ammonium citrate) are both the best absorbed and the most chemically reactive. The use of soluble iron compounds often leads to the development of off-colors and flavors due to reactions with other components of the food material. Infant cereals have been found to turn gray or green on addition of ferrous sulfate. Off-flavors can be the result of lipid oxidation catalyzed by iron. The iron compounds themselves may contribute to a metallic flavor. Some of these undesirable interactions with the food matrix can be avoided by coating the fortificant with hydrogenated oil or ethyl cellulose (Clarke 1995).

Because of the high bioavailability of ferrous sulfate, it is used extensively to fortify foods such as bread and bakery products, which are stored for short periods of time. Ferrous fumarate is used to fortify corn-soy-milk. The absorption of ferrous fumarate is comparable to that of ferrous sulfate when it is given at therapeutic levels without food. Ferrous lactate and ferrous gluconate are well absorbed but their cost generally limits their use to milk and soy-based formulas (Parrish et al. 1980).

Phosphate compounds

Phosphate compounds (ferric orthophosphate, sodium ferric pyrophosphate, and ferric pyrophosphate) are at the opposite end of the spectrum in regard to bioavailability and product quality problems. Ferric orthophosphate and sodium pyrophosphate were used extensively for flour and cereal fortification in the USA because of their low cost and low vehicle reactivity, but their use declined when isotopic studies in humans showed

absorption to be far less than that of ferrous sulfate. The relative cost of these iron forms is high because of poor availability (Cook et al.1976).

Elemental iron powders

Elemental iron powders (reduced iron, electrolytic, and carbonyl) have been used extensively for flour and bread fortification. Surface area, bioavailability, and cost are lowest for hydrogen and CO-reduced iron, intermediate for electrolytic iron, and highest for carbonyl iron (Cook et al.1976).

Complexes

A complex like NaFe-EDTA (ethylenediaminetetraacetate) or Hb (hemoglobin) in which iron is bound might minimize adverse reactions with a food vehicle. NaFeEDTA minimizes iron precipitation and Hb has high bioavailability but imparts an intense color to food. Some of the technical problems of food iron fortification might be avoided by using a relative inert form of iron in combination with a substance that enhances absorption such as ascorbic acid (Cook et al.1976).

Bioavailability of iron compounds

The bioavailability of iron compounds is normally stated relative to a ferrous sulfate standard. The highly water-soluble iron compounds like ferrous sulfate have superior bioavailability. Bioavailability of the insoluble or very poorly soluble iron compounds can be improved by reducing particle size. Unfortunately this is accompanied by increased reactivity in deteriorative processes.

The problem of low bioavailability of some of the less reactive forms of iron is often avoided by the use of absorption enhancers added along with the fortificant.

Examples of such enhancers are ascorbic acid, sodium acid sulfate, vitamin A, and orthophosphoric acid. Ascorbic acid, sodium acid sulfate and orthophosphoric acid act as acidifying agents that enhance iron availability through the reduction of the less available ferric to the more available ferrous form. Investigations carried out in Venezuela in 1995 showed that vitamin A added to precooked corn flours for arepas prevents the inhibitory effect of phytates contained in the flour; therefore the absorption from iron-fortified food and iron-containing food is doubled (Layrisse et al., in press).

In some cases, iron bioavailability is affected by the methods used to process the food. It is known that ferrous sulfate absorption is considerably less when it is baked into bread than when consumed alone. Steinkamp and co-workers (1955) measured the radioactive hemoglobin in the blood of human subjects who consumed radioiron-labeled breads. The bread contained 2 to 4 mg of iron in the form of radioactive FS, EI, FOP and FSP, the four most common iron enrichment sources. Ferrous sulfate absorption was found to be considerably less when baked into bread and when consumed with unlabeled bread than when given alone. Absorption increased two to three times when bread containing any of the forms of iron is ingested with 1 g of ascorbic acid. There was no difference in absorption among the four forms of iron after they were baked in to the bread, i.e., most of the subjects absorbed between 1 and 12% of the iron, regardless of the form of iron added (Lee and Clydesdale 1979).

Iron enrichment of dry-milled corn products

Corn meal and corn grits have been fortified with iron in the USA for many years with no major technical problems. When these dry-milled products were fortified to a

level of 88 mg iron/kg with powdered iron (reduced either electrolytically or with hydrogen) , little or no segregation occurred and no adverse odors or flavors were encountered after 56 days of accelerated storage. On the other hand, stabilized ferrous sulfate (stabilized with sodium hexametasulfate to reduce its reactivity) at this level caused some deterioration in flavor and odor (Anderson et al.1976).

Iron enrichment of NCF and precooked corn flours for arepas

The only enrichment study found in the literature concerning NCF and iron was the one conducted in Mexico by the Instituto Nacional de Nutricion “Salvador Zubiran”. NCFs (MASECA and MINSA) were enriched with a premix containing soy-oat-rice flour (10%) and seven micronutrients (iron, zinc, vitamin A, C, riboflavin, niacin and folic acid).

During a three-month pilot test, MINSA and MASECA produced enriched NCF with a distribution volume of 2 300 MT. The micronutrients were added in a quantity necessary to meet 100% of the recommended daily intake by consuming six tortillas/d. An increase in digestibility (10%), protein quality (40%) and organoleptic quality (70%) was obtained. The flavor of the tortillas was different but close to the original corn flavor. No significant odor change was detected and most people said they would buy the product in the future (Torres et al. 1996).

Arepas are widely consumed in Colombia and Venezuela, where they are considered the national bread (Cuevas et al. 1985). Arepas are traditionally produced from partially degermed corn grits or meal that is soaked, cooked in water, ground to a

moist dough, and shaped into flat discs (approximately 7.5 cm in diameter and 1 cm thick). The discs are browned on each side and baked.

Commercially prepared precooked arepa flours that require only hot water to produce a dough are made by cooking the corn grits to gelatinize the starch, putting the cooked grits through flaking rolls, and grinding the dry flakes into flour or meal of acceptable particle size distribution. Approximately 700, 000 tons of corn are processed annually into arepa flour in nine plants in Venezuela (Cuevas et al. 1985). Smith et al. (1979) proposed an alternative method for production of precooked arepa flour by extrusion cooking moist corn grits followed by drying, grinding, sieving, and packaging.

Precooked corn flours for arepas were enriched in Venezuela (1993) with 50 mg Fe (as ferrous fumarate)/kg flour as part of a fortification program where wheat flour was also fortified with 20 mg Fe (as ferrous fumarate)/kg. The consumption of precooked corn flour for arepas per capita was 80 g/d for the total population in 1994. The consumption of wheat flour was 40 g/d as bread and 30 g/d as pasta. A preliminary survey was carried out in 1994 in a population of 307 children aged 7, 11, and 15. The results showed that the prevalence of iron deficiency (determined by measuring the serum ferritin concentrations) and the prevalence of anemia were reduced from 37% and 19%, respectively in 1992, to 15% and 10%, respectively in 1994. The organoleptic properties of the flours (color, odor, texture, and flavor) did not change for six months. The only adverse effect observed during the initial two years of the fortification program occurred in two regions of the country in which hard water is used for making arepas; the bread turned slightly dark due to the addition of the hard water (Layrisse et al.1996).

Enrichment of NCF and related products with other nutrients

Enrichment with amaranth

NCF can be mixed with milling fractions of raw amaranth seeds (\$4.98/kg) to increase protein and fat content of tortillas and arepas. Adding up to 50% of amaranth milling fractions produces no change in organoleptic characteristics (Sanchez-Marroquin and Maya 1985). Adding amaranth (*Amaranthus cruentus*) yields products of excellent nutritional quality due to its good amino acid profile and protein efficiency ratio (PER). Pacheco and Portillo (1990) analyzed arepas made with up to 30% of amaranth flour (\$3.50/kg). Results showed that arepas prepared with the mixture containing just 10% of the amaranth flour had a higher lysine content (1.6 g lysine/100g protein) than the commercial arepas (0.7 g lysine/100g protein).

Soy-enriched NCF and related products

Soy-enriched NCF can be produced through the typical NCF process by cooking whole raw corn-soybean mixtures containing 10% soybeans instead of pure corn (Del Valle et al.1976). Protein quantity and quality increase as follows through the addition of soybeans: protein content, from 9.2 to 12.6%, an increase of 36%; protein efficiency ratio (PER) from 1.45 to 2.62 (casein PER = 3.40), an increase of 81%. Also, no organoleptic changes are found between normal tortillas and tortillas prepared with the soy-enriched flour.

Bressani et al.(1966) formulated a vegetable mixture (58% corn flour, 38% soybean flour, 3% torula yeast, 1% calcium phosphate, and 4,500 I.U. vitamin A per

100g) with approximately 27% protein. This formula was evaluated in young dogs. Its main limiting amino acid was methionine, but only when fed at a low level of protein. The addition of lysine and threonine in the presence of methionine caused a highly significant increase in the protein efficiency ratio.

After developing the INCAP Vegetable Mixture 14, Bressani continued to work in increasing protein and calories of corn-based diets. His early studies indicated that the addition of 8% soybean flour, containing 50% protein, gave optimum protein quality when added to corn (Bressani and Marengo 1963). Since 8% soybean flour provided 4 g of protein, quantities supplying this additional amount were added when using whole soybeans for preparing soy-enriched tortillas (Bressani et al.1974). Eighty-five percent of whole corn was cooked with 15% of whole soybeans. To each batch of corn and whole soybeans were added 1.6 times water, by weight, and 1.7% calcium. The soy-enriched tortillas had similar physical characteristics to those of traditional tortillas, and water retention for both types of preparation was essentially the same. Furthermore, acceptability was very good among testers.

Fortification with coarse defatted corn germ

The dietary value of precooked corn flour can be improved through fortification with 11% coarse defatted corn germ (Rivero et al.1994). The results of tests in humans show that the total iron absorption from the fortified preparation is similar to that from the precooked corn alone, but the fortified flour is richer in several nutrients; protein (25%), zinc (61%), potassium (47%), magnesium (112%), and fiber (34%). Fortification with defatted corn germ is not practical in Mexico or Central America because there is

little if any corn dry milling. The use of imported germ would be economically impractical.

Fortification with vitamins

NCF can be fortified with vitamins by adding them to the flour in the form of a dry premix. When mixing dry vitamins with flours a careful selection of the physical characteristics of the compounds is important to ensure adequate mixing and to minimize segregation during storage. Vitamin A is sufficiently stable in flours at warehouse temperatures for up to six months and it has no apparent tendency to segregate during handling or shipping in bags. The form of vitamin A most commonly used in the fortification of flour is dry stabilized vitamin A palmitate (type 250-sd) powder form. Other vitamins like thiamin, riboflavin, niacin, pyridoxine, folate and calcium pantothenate are used in their pure crystalline form and are also stable during storage.

Parrish et al.(1980) added to cornmeal a premix formulated with vitamins and iron (as reduced iron at 10 g cwt.). Losses of vitamin A during storage at room temperature for up to six months were greater than with wheat flour but still remained below 20%. A similar study was carried out in Venezuela in 1993 with precooked corn flours for arepas. The flours contained 50 mg of ferrous fumarate/kg plus vitamin A, thiamine, riboflavin, and niacin. The vitamins had good stability at room temperature storage at normal moisture levels (6.5%). Retention of the vitamins after cooking was also good. These studies show that vitamin fortification of precooked corn flours is technically feasible.

CONCLUSIONS

The current knowledge of and technology for fortification of NCF with iron is very limited. Only one study on iron fortification of NCF was reported. However, the studies conducted on iron enrichment of corn dry-milling products, precooked corn flours for arepas, and wheat flours give preliminary knowledge and techniques for the development of an iron fortified NCF. An important difference between these products and NCF is pH. NCFs are neutral while corn dry-milling products, precooked corn flours for arepas, and wheat flours have a lower pH.

NCF available in the market have a pH ranging from 6 to 7.2. At pH 7 ferrous ion will precipitate as FeOH_2 , having a solubility of about 10^{-1} M. Ferric ion is much less soluble than ferrous, precipitating as FeOH_3 , which has a solubility of about 10^{-16} M at pH 7. The formation of sparingly soluble hydroxides with increasing pH has nutritional significance, as solubility of iron is a prerequisite to its absorption in the gut. In the acid medium of the stomach, which has a pH of about 1, the iron is in a soluble and hydrated form. Upon passing into the alkaline medium of the small intestine, where all iron absorption takes place, most of the iron would be expected to precipitate. However, it is evident that extensive precipitation does not occur because appreciable iron absorption does take place in the small intestine. The control of iron uptake in the small intestine must be determined by the ability of the iron to be chelated by low molecular weight ligands (Lee and Clydesdale 1979).

Increased iron absorption can be achieved by using iron complexes (with soluble low molecular weight chelates like ascorbate, fructose or glucose) as iron enrichment sources, which would prevent the formation of sparingly soluble (and thus less available)

hydroxides in the gut. The use of microencapsulated iron as a fortificant is not justified, as it would also precipitate in the gut (since it is not chelated).

The most logical approach would be to enrich NCF with iron by mixing up to 50 mg of ferrous fumarate/kg. This compound is known to have certain organoleptic properties that can be appealing and could be added to the product at a reasonable cost (\$9.15/kg). It also has a bioavailability comparable to that of ferrous sulfate. Ferrous fumarate added to NCF would not segregate significantly and the quality of the products made with this flour would most likely be acceptable since it is less reactive than ferrous sulfate, leading to fewer problems with off-coloration and catalysis of fat oxidation reactions.

The high pH of NCFs might not reduce the availability of ferrous fumarate significantly since the pH of the small intestine is also high. The use of iron-complexes like ferrous gluconate to increase iron absorption by preventing precipitation might not be justified since they are six times more expensive than ferrous fumarate.

The following questions must be answered in order to successfully develop an iron-fortified NCF: 1) What is the amount of iron consumed in other foods by the target population?, 2) Does NCF affect the bioavailability of iron?, 3) Is iron-enriched NCF stable during storage? and, 4) Will tortillas produced with iron-enriched NCF develop changes in color, odor, flavor, and/or texture?

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DATA BASES:

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OVID
PubMed (<http://www.ncbi.nlm.nih.gov/PubMed/>)
OMNI Web Site fortification update (<http://www.idrc.ca/mi/fort196.htm>)

*Relevant publications before 1970 were consulted.

KEY WORDS USED:

Corn
Flour
Fortification
Corn flour fortification
Tortilla
Tortilla fortification
Corn-based fortified foods
Enriched tortilla flour
Enriquecimiento de las tortillas
Enriquecimiento de arepas
Fortified corn meal
Fortified corn products
Enriched corn products

Enriched nixtamal
Enriched nixtamalized corn flour
Enriched instant tortilla flour
Fortified instant tortilla flour
Enriched dry masa flour
Fortified dry masa flour
Suplementacion de la tortilla
Nutricion de la tortilla
Suplementacion con hierro de la tortilla
Iron fortification
Maize flour
Harina de maiz
Fortification of maize
Enrichment of maize
Micronutrients

*NCF Market information obtained from MINSA Corporation.

APPENDICES

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APPENDIX B: Mineral Supplement Companies

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Balchem Corp.
Rte. 6 And 284, Box 175
Slate Hill, NY 10973
Phone: 914-355-2861
Fax: 914-355-6314

Barrington Chemical Corp.
16 School St.
Rye, NY 10580
Phone: 800-684-CHEM
Fax: 914-921-4412

Bio-Botanica, Inc.
75 Commerce Dr.
Hauppauge, NY 11788
Phone: 516-231-5522
Fax: 516-231-7332

Em Ind., Fine Chemicals Div.
5 Skyline Dr.
Hawthorne, NY 10532
Phone: 914-592-4660

Fcc Products Inc.
106 Naylor Ave.
Livingston, NJ 07039
Phone: 800-524-0708

Fmc Corporation, Phosphorus Chemicals Div.
1735 Market St.
Philadelphia, PA 19103
Phone: 215-299-6887

Fortitech Inc.
Rotterdam Industrial Park
Schenectady, NY 12306
Phone: 518-356-5155
Fax: 518-356-2729

Freeman Industries, Inc.
100 Marbledale Rd.
Tuckahoe, NY 10707
Phone: 914-961-2100
Fax: 914-961-5793

Gallard-Schlesinger Industries, Inc.
584 Mineola Ave.
Carle Place, NY 11514
Phone: 516-333-5600
Fax: 516-333-5628

Grow Company
195 Kenneth Ave.
Hackensack, NJ 07061
Phone: 201-342-2007
Fax: 201-342-9127

International Sourcing Inc.
121 Pleasant
Upper Saddle River, NJ 07458
Phone: 201-934-8900
Fax: 201-934-8291

Mallinckrodt, Inc.
Box 5439
St. Louis, MO 63147
Phone: 800-325-8888

McShares, Inc.
P.O. Box 1460
Salina, Kansas 67402-1460
(913) 825-2181
Fax (913)825-8908
Telex 417318 Repco Sal

Penta Manufacturing Co.
50 Okner Pkwy
Livingston, NJ 07039
Phone: 201-740-2300
Fax: 201-740-1839

Protein Research Associates
1999 Pike Ave.
San Leandro, CA 94577
Phone: 510-614-7716; Fax: 510-634-7720

Quest International
5115 Sedge Blvd.
Hoffman Estates, IL 60192
Phone: 708-645-7000; Fax: 708-645-7070

Reheis Inc.
500 N. Ninth St., P.O. Box 921
Midlothian, TX 76065
Phone: 214-775-2307
Fax: 214-775-3872

Ria International
23 Vreeland Rd. #240
Florham Park, NJ 07932
Phone: 201-660-0770
Fax: 201-660-0880

Scan American Seafood Co. Inc.
1410 80th St. S.W., Suite F
Everett, WA 98203
Phone: 206-514-0500
Fax: 206-514-0400

Seltzer Chemicals, Inc.
5931 Priestly Dr.
Carlsbad, CA 92008
Phone: 619-438-0089
Fax: 619-438-0336

Soluble Products Co., L.P.
480 Oberlin Ave., S.
Lakewood, NJ 08701-6997
Phone: 908-364-8855
Fax: 908-364-6689

Vitamins, Inc.
200 E. Randolph Dr.
Chicago, IL 60601
Phone: 312-861-0700
Fax: 312-861-0708

Watson Foods Co., Inc.
301 Heffernan Dr.
West Haven, CT 06516
Phone: 203-932-3000; Fax: 203-932-8266

Weinstein Nutritional Products
17751 Mitchell
Irvine, CA 92714
Phone: 714-833-9500; Fax: 714-833-9595

Wellington Foods, Inc.
21414 S. Alameda St.
Long Beach, CA 90810
Phone: 310-834-9311; Fax: 310-834-5008

APPENDIX C: Quality Assurance

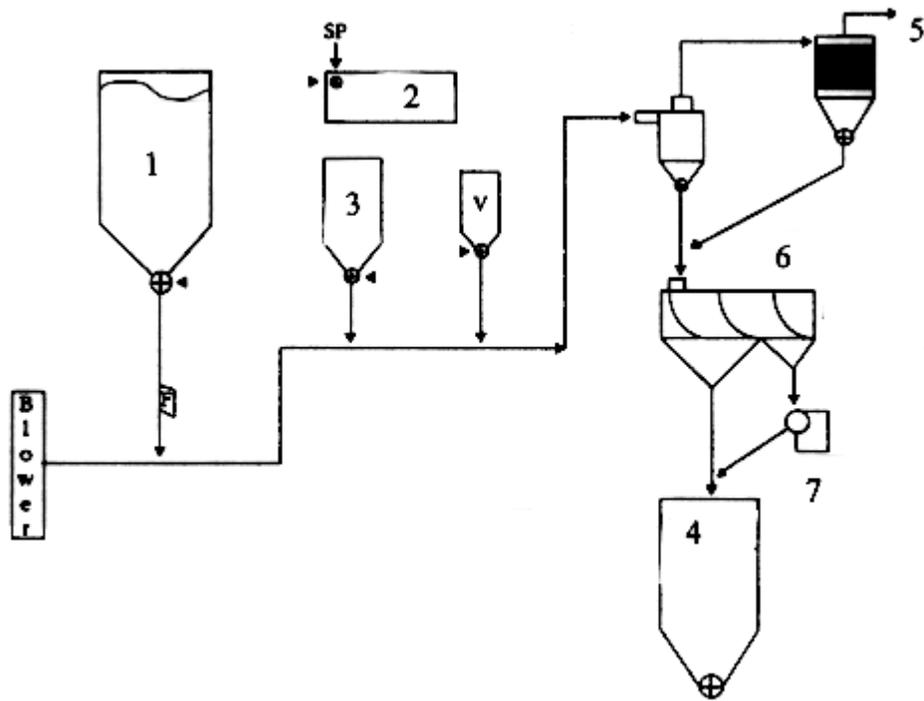
To assess the quality of the enriched NCF being produced, it would be necessary to test for iron content. This is necessary because of possible dosifier fluctuations during the production process, and because iron segregation might occur during shipping and handling. The NCF producers should advise tortilla plants to mix the flour prior to tortilla production. To monitor NCF quality, random samples of commercial tortillas should be tested for iron content and organoleptic characteristics such as flavor, color and aroma. An iron content determination test and a sensory panel evaluation could be run on 40 tortillas/day (assuming 4 batches/day and randomly sampling 10 tortillas/batch).

The iron content of the finished product can be determined by the Iron-Quantitative Method (AACC Method 40-41). The iron content of the NCF can be determined by the Iron (9) Method (AOAC Method 14.011) applicable to enriched flours. If the tortilla plant has a quality control lab the cost of the iron analysis would be the price of the necessary reagents and the pro-rated salary of a lab technician (\$10,000-20,000/year). The sensory panel can be conducted with 8 to 10 panelists trained to detect flavor, color and aroma differences in the tortillas. The panel can be formed by people working at the plant. Once they are trained, sensory evaluation can be supervised by the lab technician. Alternatively, NCF and tortilla samples can be sent to a commercial analytical laboratory to insure that proper levels of iron are present. A full service analytical laboratory would charge \$30 per sample (Coffey Laboratories, Portland, OR, Tel. 503- 254 1794).

APPENDIX D: Process and Equipment for Premix Addition

In a continuous mixing process there are natural occurring fluctuations that can be reduced with flow regulators. Another problem is the lumping of the dry ingredients due to moisture differences between the components and the environment. It is necessary to disperse the lumps, grind them and return them to the process. A premix of the iron and other micronutrients can be made using a small quantity of NCF, then this premix can be added to the rest of the NCF to reach the desired level (Figure A1). The initial micronutrient mix containing the vitamins and iron is difficult to add homogeneously to the flour, so the dilution premix with NCF is made (1:4 vitamin-iron:NCF). The vitamins, especially B1 and B2 are highly susceptible to light, so any exposure of the premix to light during the mixing process should be minimized.

The residual pneumatic transport system air from the cyclones usually still contains fine particles. This is why filters are needed to avoid the escape of these particles that would otherwise pollute the atmosphere (Torres et al.1996).



- V= vitamins + iron (diluted premix 1:4 with NCF)
- F= flour flux meter
- 1= NCF
- 2= digital controller
- 3= other flours (soy, amaranth, etc.)
- 4= fortified NCF
- 5= air
- 6= mixer
- 7= mill

Figure A1. Nixtamalized corn flour fortification process (Contreras 1996)

APPENDIX E: Comments on Other Industrial Corn Flours

The instant corn-soy flour produced by ConAgra is not a nixtamalized corn flour because they use corn flour (obtained from corn dry milling) as the main ingredient. A nixtamalized corn flour has to be produced by alkaline cooking of the corn followed by washing, grinding and drying.

APPENDIX F: Comments on Quaker Oats and ConAgra

Companies processing nixtamalized corn flour in the United States have in general not fortified with nutrients. In some cases, companies have added nutrients to meet specifications for local school lunch programs. We requested information on the specifics of these situations. However, we have not received the information although it was promised. We were told that there are no major problems with fortification of dry masa flour.

We were told that ConAgra had produced fortified masa flour for food aid programs. Upon further evaluation we found that the masa flour was actually precooked corn meal that had apparently been adjusted to basic pH during the processing. It could not really be used to produce high quality tortillas and related products. Today, ConAgra is attempting to market a NCF to the snack and tortilla industry. This new product is the result of extensive research to produce NCF that can compete with other established products, i.e., Maseca.

Quaker Oats produces dry masa. In the past, some NCF was apparently enriched for specific customers. Information on the details was not provided.

Our conclusions are that NCF can be easily enriched with iron and other nutrients similar to other corn products (dry milled flour, meal) but there is relatively little information on the effect of the nutrients on the flavor, color, odor and stability of the NCF and tortilla produced therefrom. Apparently, if there are problems they are not very significant. Therefore, it may be desirable to evaluate the effects of iron enrichment on attributes of NCF and its products along with longer-term storage studies.

III

THE FORTIFICATION AND ENRICHMENT OF CORN

TORTILLAS: AN INDUSTRIAL APPROACH

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INTRODUCTION

Maize (*Zea mays L*) is the basic staple for the people of Latin America. Traditionally the grain is cooked with alkali with the aim of producing nixtamal (from the Nahuatl word *nixtli* = ashes and *tamalli* = dough or masa). Nixtamal is generally stone ground into a dough or masa which is the basic material for the traditional or industrial elaboration of tortillas, nixtamalized flours and related products (Serna Saldivar et al. 1990; Serna Saldivar and Rooney, 1988).

Maize was the key factor for the development of pre-Columbian civilizations and nixtamalization was the critical process to enhance the nutritional value of the grain and the palatability of the products. Maize was so critically important for the development of mesoamerican civilizations that many scriptures, ceremonials and traditions are linked to this important grain. The Aztec civilization worshipped the corn god and goddess Centoetl and Chimecoatl, respectively, that represented the welfare of the population. Similarly, the Mayans worshipped the corn god named Yum Kaxx (Serna Saldivar et al. 1990, 1993).

The basic principles for the preparation of tortillas have been unchanged since the discovery of the Americas. Perhaps the most significant industrial advancement has been the process to manufacture dry nixtamalized flours, products that are increasing in popularity due to their extended shelf life and convenience. The market share for these flours has increased steadily during the past decade due to their convenience, as well as savings in labor, equipment, energy usage, plant space and processing time (Gomez et al. 1987). Currently, in Mexico more than 3 million metric tons of maize are channeled to the dry masa flour industry (Serna Saldivar 1996b). The big industrial consortiums utilize the same basic technology developed by the Mesoamerican civilizations. The current process consists of cooking the grain in excess water with lime in large cooking vessels or continuous reactors at temperatures near 100°C for times that range from 15 to 45 min. The extent of cooking is related to grain hardness and/or density, grain size and rate of heat transfer of the cooking vessels. After cooking the cooking liquor, called *nejayote*, is drained and the resulting nixtamal washed with pressurized water. During these steps the

grain absorbs part of the calcium hydroxide and loses the pericarp or bran tissue and other solids. The clean and washed nixtamal is then ground into a coarse dough in preparation for the critical step of dehydration. The dough is dried to reduce its moisture content to around 10% and the resulting particles classified by sifting and reground to formulate different flours for various end uses (Rooney and Serna Saldivar 1987; Serna Saldivar et al. 1990). In Mexico, most flours are tailor-made for the table tortilla industry. More than half of the tortillas produced in Mexico are manufactured from dry masa flours. The yearly *per capita* consumption of tortillas produced from dry masa flours is approximately 60 kg (Serna Saldivar 1996b).

Tortillas and related products have been the food most widely consumed by Mexicans. The annual per capita consumption in some groups reaches 120 kg (Serna Saldivar et al. 1990). The consumption is higher in rural areas and among low-income groups. In some areas of Mexico, tortillas provide more than 75% of the total caloric and protein intake.

OBJECTIVES

1) To review and compare the nutritional value of regular corn tortillas with that of tortillas fortified with soybean protein and enriched with various essential vitamins and iron.

2) To propose the best way to industrially produce fortified flours at the lowest possible cost, and the best alternative to distribute them so they can reach the poorest socioeconomic groups.

3) To present preliminary nutritional data (first year of the study) provided by the Instituto Nacional de Nutrición Salvador Zubirán on two Otomi (indigenous Mexican) groups consuming regular and fortified tortillas produced from nixtamalized flours (MASECA®).

MALNUTRITION IN LATIN AMERICA

Malnutrition is endemic to all Latin American countries, and is especially prevalent among children of poor and unemployed families without resources to purchase adequate amounts of quality food. The tortilla consuming countries (Mexico and most countries in Central America) have similar malnutrition problems because their diets are similar. In Mexico, 50% of the children are malnourished and most of these belong to poor families. The main malnutrition problems are related to the lack of good protein, iron, and vitamins A, C and the B-group. According to the IMSS (Instituto Mexicano del Seguro Social; the Mexican Social Security Institute), approximately 160,000 infants of less than 5 years of age die of problems associated with malnutrition. The malnutrition problem has worsened during recent years because the socioeconomic gap between poor and rich has been increasing. Malnutrition is prevalent in rural areas where the diet is monotonous and is based on tortillas. A tortilla-rich diet lacks adequate protein for infant growth, does not have enough vitamin A, iron and zinc, and in practical terms completely lacks vitamin C. The lack of adequate hygiene, potable water and medical services make these people more susceptible to diseases and death (Serna Saldivar 1996b).

NUTRITIONAL VALUE OF REGULAR CORN TORTILLAS

Maize undergoes several significant changes when it is processed into tortillas. At equivalent moisture contents, tortillas contain less dietary fiber and fat than the original grain. This is due to the losses of pericarp and germ tissues that occur during nixtamalization (Serna Saldivar et al. 1988a, 1988b; Serna Saldivar and Rooney 1988, Bressani, 1990; Bressani et al. 1958). However, both products contain similar quantities of carbohydrates (starch), protein and ash or minerals. There are, however, significant changes in the mineral content, due to the loss of minerals present in the pericarp and germ and the absorption of lime during the nixtamalization cooking and steeping (Serna Saldivar et al. 1988a, 1988b, 1990).

Energy. Tortillas are considered an excellent source of calories due to their high content of starch. Their energy digestibility is very high and approximates 93% (Table 1; Serna Saldivar et al. 1988a, 1988b; Sproule et al. 1988). The starch is in practical terms 100% absorbed and, during digestion, its glucose is transported to blood more slowly; a benefit for diabetic people. The consumption of 5 tortillas (150 g) provides about 230 kcal (Serna Saldivar 1996a).

Protein. Corn transformed into tortillas undergoes significant changes in its protein fraction. Lime cooking/baking decreases slightly the rate of protein digestibility and the bioavailability of lysine, the limiting essential amino acid in corn (Chu et al. 1976; De Groot and Slump 1969; De Groot et al. 1976; Serna Saldivar et al. 1988b). However, in practical terms tortillas and grain have similar protein nutritional values. The protein digestibilities of the grain and tortillas are around 88 and 85%, respectively (Table 1; Serna Saldivar et al. 1988a, 1988b; Sproule et al. 1988). The protein quality of both corn and tortillas is considered low because these products do not contain enough quantities of lysine and tryptophan to sustain regular growth in infants. These amino acids are present in about half of the amount required for optimum growth. Therefore, the biological and net protein utilization values and protein efficiency ratios are approximately one half of those for animal products (i.e., meat and milk). Thus, an infant can not live exclusively on tortillas.

Fiber. Dietary fiber in corn consists primarily of hemicellulose and B glucans (Serna Saldivar 1996b). Arabino-glucuronoxylans are the major hemicellulose components in the pericarp or bran, and B glucans and xylans dominate in endosperm cell walls. When corn is processed into nixtamal, it loses most of the pericarp and therefore most of the insoluble fiber. The extent of cooking and degree of washing dictate the amount of pericarp loss in the nejayote (Serna Saldivar et al. 1991). The xylans are susceptible to alkaline hydrolysis during processing (Ghali et al. 1984; Nyman et al. 1984). According to Reinhold and Garcia (1979) the neutral and acid detergent fiber contents of masa from 20 Mexican tortilla factories were 6% and 3%, respectively. Baking masa into tortillas increased values to 6.6% and 3.8%. The increase in dietary fiber was attributed to the formation of insoluble Maillard browning products. Serna

Saldivar et al. (1988a, 1988b, 1990) reported that corn tortillas contain 10.9% insoluble and 1.2% soluble dietary fiber. Ranhotra (1985) reported 4.1% dietary fiber in corn tortillas.

Minerals. Corn tortillas contain adequate amounts of the macrominerals phosphorus and calcium, although some researchers question their availability. Serna Saldivar et al. (1991, 1992) recently demonstrated that the calcium from tortillas is well absorbed, metabolized and deposited in the bones of rats fed diets of tortillas and beans. The same authors calculated that around 50% of the calcium ingested by the Mexican population comes from tortillas and related products. Krause (1992) estimated that most of the dietary calcium in the diets of Guatemalan Indians was provided by tortillas.

Table 1. Effect of Tortilla Processing on Nutrient Digestibility of Corn, Tortillas and Tortillas Chips

	----- Digestibility Rate -----		
	Dry Matter (%)	Energy (%)	Protein (%)
Ileum-cannulated swine^a			
Digestibility at terminal ileum			
Corn cooked without lime	76.3	77.8	76.5
Corn cooked with lime	76.3	78.4	72.8
Digestibility over total digestive tract			
Corn cooked without lime	91.5	91.6	85.4
Corn cooked with lime	92.0	92.6	84.6
Weanling laboratory rats^b			
Raw corn	90.3	91.1	86.2
Nixtamal	92.9	93.8	79.3
Tortillas	93.2	93.8	81.6
Laboratory rats^c			
Raw corn	91.4	92.9	86.1
Tortillas	94.3	94.3	83.8
Tortilla chips ^d	94.4	94.7	82.1

^aData from Serna-Saldivar et al (1987).

^bData from Serna-Saldivar et al (1988b).

^cData from Sproule et al (1988).

^dDefatted prior to feeding studies.

In an early study, Braham and Bressani (1966) found that rats absorbed 86.6% of tortilla calcium. The supplementation of tortillas with lysine and other essential amino acids enhanced calcium absorption. Tortillas are considered a poor source of microminerals iron, zinc and copper, and their bioavailability is limited due to the presence of phytic acid. Tortillas are a very poor source of iron because the raw grain has low quantities of this important mineral and part is lost during nixtamalization. A 150 g portion of tortillas provides about 1 mg iron, less than 10% of the recommended daily allowance (Serna Saldivar 1996a). Reinhold et al. (1984) reported that the fiber and zein (corn endosperm protein) bind a substantial proportion of added ferrous and ferric iron at pH 6 or higher but released these iron forms completely at pH values lower than 5. The buffering effect of lime diminishes the release of iron bound by fiber or protein in tortillas. Additionally, the concentration of calcium in tortillas is within the range reported to produce interference with the uptake of iron by the intestinal epithelium.

Vitamins. Corn tortillas are a poor source of liposoluble vitamins (A, D, E, and K), and of vitamins C and B₁₂. However, they contain significant amounts of the other B vitamins, in spite of the losses that occur during nixtamalization (Bressani et al. 1958; Bressani 1990). Gomez et al. (1996) documented losses that ranged from 30% to 70% during lime cooking in a continuous extruder. Vitamins are partially lost due to losses of pericarp, germ and aleurone tissues. Two of the documented advantages of lime-cooking are that during this process the bound niacin is released, and that the isoleucine-leucine amino acid ratio is improved (Bressani and Scrimshaw 1958; Katz et al. 1974, Koetz and Neukom 1977, Wall and Carpenter 1988). Therefore, the bioavailability of niacin is greatly improved (Cravioto et al. 1952). That is the reason why pellagra² is virtually nonexistent in tortilla-eating countries.

² Pellagra is a disease due to niacin deficiency and is characterized by eruption on the skin (dermatitis), severe functional disturbances and mental derangement (dementia).

TORTILLA FORTIFICATION

Many efforts have been made to improve the nutritional value of tortillas by protein fortification, utilization of high-lysine corn (Bressani and Marengo 1963; Bressani et al. 1988; Bressani et al. 1974, 1979; Del Valle and Perez Villaseñor, 1974; Sproule et al. 1988; Serna Saldivar et al. 1988) and vitamin/mineral addition (Bressani and Marengo 1963, Reinhold et al. 1984; Serna Saldivar 1996; Muñoz de Chavez and Chavez 1997).

Protein. Protein supplementation offers various advantages such as increased protein content, improved essential amino acid balance and improved protein utilization and growth. The nutritional value of tortillas can be upgraded by the addition of whole soybeans, defatted soybean flour, amaranth, fish meal, torula yeast and by addition of lysine and tryptophan (Bressani and Marengo 1963; Bressani et al. 1968, 1974, 1979; Del Valle and Perez Villaseñor, 1974; Sanchez Marroquin et al. 1987; Tonella et al. 1983; Serna Saldivar et al. 1988). Fortification with soybeans is the most feasible because of its low cost, high availability, high protein content and especially due to its proven complementary effect on corn proteins. Defatted soybean flour has the advantage of better shelf stability although full-fat soybean flour produces a higher caloric-density tortilla. The addition of 8% soybean improves the amount of protein, lysine and tryptophan, significantly improves nitrogen retention (Biological and Net Protein Utilization Values) and almost doubles the PER (Protein Efficiency Ratio) in weaning rats (Bressani et al. 1974, 1979; Serna Saldivar et al. 1988). At this level of fortification the overall quality and sensory characteristics of tortillas are not adversely changed (Almeida Dominguez 1984; Serna Saldivar et al. 1988).

Feeding trials have demonstrated the nutritional superiority of the maize varieties opaque 2 and quality protein maize (QPM) with respect to regular corn (Sullivan et al. 1989, Sproule et al. 1988, Bressani et al. 1968; Serna Saldivar et al. 1992). This is because QPM has higher levels of lysine and tryptophan that result in better nitrogen retention and growth. The preparation of tortillas from QPM significantly improves the protein quality, as measured with rats and other animals. The growth rates of

experimental animals fed these varieties are almost double those of animals fed regular corn diets (Bressani 1992).

Iron. Iron is the most important micromineral in human nutrition, especially in developing and poor countries (Serna Saldivar 1996b). The main cause of anemia is the lack of this important mineral. It is estimated that at least 1.2 billion of the 5.4 billion people that inhabit the world suffer from iron deficiency. Infants, and pregnant and lactating women are the most affected. In México, iron deficiency affects from 10% to 70% of the population, and anemia is prevalent among economically disadvantaged families and people living in urban marginal areas. Planning for the control of iron deficiency and the betterment of nutritional status of children requires special initiatives by government leaders and policymakers. The tortilla can be an excellent vehicle for the incorporation of iron into the diets, much better than wheat flour or breakfast cereals. A precise understanding of the benefits of prevention programs, a knowledge of the magnitude of the problem and an understanding of its causes, are prerequisites for effective policy and program development.

Enrichment has been recommended as the best long-term approach for combating iron deficiency and anemia in large segments of the population. Its major advantage is that it does not require the conscious cooperation of the target group. When widely practiced, it improves nutrient consumption, work performance, and public health (by reducing susceptibility to infection and diseases). In general, iron from vegetable and grain sources is poorly absorbed by humans, whereas iron from animal sources is highly available. The consumption of vitamin C enhances iron absorption. The addition of iron to flours and other common cereal-based foods has been practiced since the Second World War. However, iron enrichment still does not reach segments of the population that need it the most. Table 2 summarizes the main iron forms used to enrich or fortify foods, or as oral supplements.

The selection of the iron form is based on its relative cost, solubility, production of off-flavors and its bioavailability to the human system (Clydesdale and Wiemer, 1985 and Arce, 1997). The least expensive sources that have good bioavailability include ferrous sulfate and ferrous fumarate. The fumarate form has low water solubility.

Ferrous lactate and ferric citrate have excellent water solubility and bioavailability, but are more expensive than the other forms. Iron is a promoter of rancidity and produces off-colors and flavors in certain food systems. Since nixtamalized flour contains low quantities of fat (around 1.5%), this suggests that the more reactive forms of iron can be used or tested (i.e. ferrous forms and highly bioavailable forms).

Table 2. Iron Sources Commonly Used to Fortify Foods and for Oral Treatment to Prevent and Treat Anemia

Iron Source	Fe Mean Content (%)	Relative Average Bioavailability -----		Relative Cost ^a
		Rats	Humans	
High Solubility				
Ferrous Sulfate 7H ₂ O	20	100	100	1.0
Ferrous Gluconate	12	97	89	5.1
Lactate	19	---	106	4.1
Ammonium Ferric Citrate	18	107	---	5.2
Ammonium Ferric Sulfate	14	99	---	2.1
Ferric Pyrophosphate +	---	---	---	---
Sodium Citrate	10	103	---	5.3
Choline Ferric Citrate	14	102	---	11.0
Low-Medium Solubility				
Dry Ferrous Sulfate	33	100	100	0.7
Ferric Glycerophosphate	15	93	---	10.5
Ferric Sacarate	3 - 35	92	74	4.1
Ferric Citrate	17	73	31	4.6
Ferric Sulfate	22	83	34	1.1
Ferric Pyrophosphate	---	---	---	---
Ammonium Citrate	16	83	---	2.6
Low Solubility				
Ferrous Fumarate	33	95	100	1.3
Ferrous Succinate	35	119	92	4.1
Ferrous Tartrate	22	77	62	3.9
Ferrous Citrate	24	78	74	3.9
Insoluble				
Ferric Pyrophosphate	25	45	39	2.3
Ferric Orthophosphate	28	7 - 32	31	4.1
Iron Sodium Pyrophosphate	15	14	15	3.5
Iron Reduced Element	97	8 - 76	13 - 90	0.5 - 1.0

Sources: Clydesdale and Wiemer (1985) and Arce (1997).

^aRelative to cost of Ferrous Sulfate 7H₂O (362 German Marks/100kg, Feb. 1984)

INDUSTRIAL FORTIFICATION OF NIXTAMALIZED FLOUR

Industrial Production of Dry Masa Flour

Dry masa is produced by alkaline cooking of corn, followed by washing and grinding to produce masa, which is dried and ground into flour (Fig. 1). The flour is sieved into different particle sizes, which are blended to obtain dry masa flours with optimum properties for different applications. One hundred kg of corn usually yield around 93 kg of dry masa flour (Serna Saldivar et al. 1990). During blending, masa flours for table tortilla production are usually supplemented with gums and preservatives and acidulants before packaging. In-home-preparation of table tortillas, using dry masa flours, is rapidly growing in Mexico. Nixtamalized dry flours are convenient because they have a shelf life up to one year in dry storage and require only water to reform the masa. One major advantage of using dry masa flour is production flexibility. This is because in the final step of the production process different streams of dry masa flour with different particle size or granulometry can be mixed to produce an array of final products with a variety of characteristics. This production flexibility is achieved by mixing flours with different particle size distribution, color (yellow and white corn varieties can be used, as well as different concentrations of lime, which affect the color) and additives (gums, emulsifiers, mold inhibitors, acidulants etc). In the non-industrial nixtamalization process this range of varieties can be obtained, but with difficulty. For Central American tortillas (which are thicker than Mexican tortillas) the industry can process dry masa flours with the appropriate granulometry, color, and shelf-life (Rooney and Serna Saldivar, 1987; Serna Saldivar et al. 1990).

Masa flours for the production of white and yellow table tortillas, restaurant style chips, tortilla chips, corn chips and tamales are available. Dry masa flour also improves the consistency of the end product. It can be easily blended with other dry ingredients (i.e. preservatives, gums, enrichment mixes etc.) during processing. In the dry masa process (Fig. 1) corn is cooked with a lime or CaO solution, steeped for short periods of time and the resulting nixtamal washed with water. The clean nixtamal is ground into a coarse masa which is then dried.

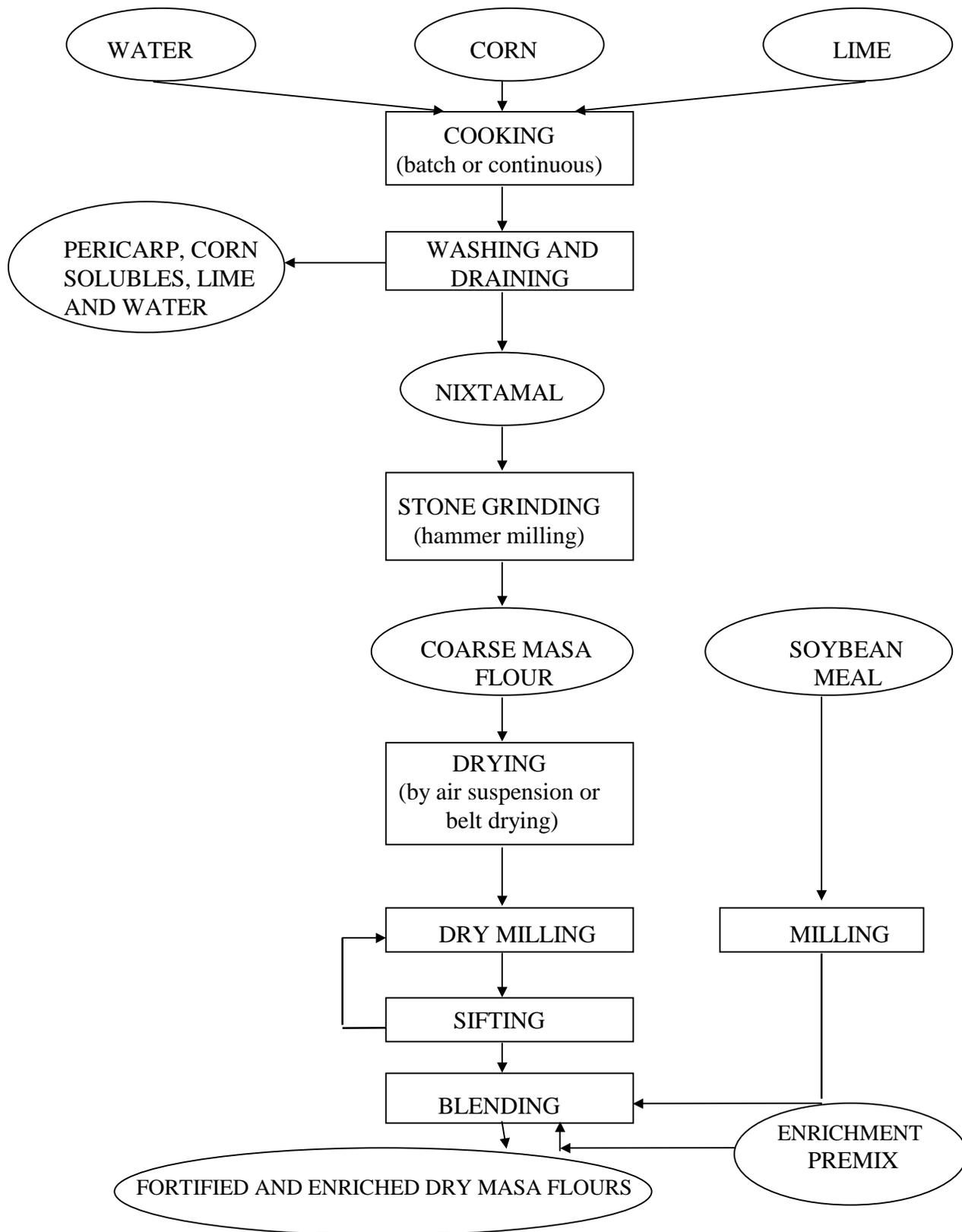


Fig. 1. Flowchart of the industrial production of regular and fortified dry masa flours.

Drying is the most critical operation and is generally done in large tunnels or drying towers in which warm air flows countercurrent to the pieces of masa. The masa is dried to a moisture content of about 8-10% and the resulting particles are sieved after hammer or roller milling to achieve the desired particle size. Various streams with different particle sizes are selected and blended to produce dry masa flours with optimum particle size distribution for different applications (i.e. table tortilla, snacks, flours for tamales and atoles). The flour is packaged in 1 to 22 kg bags (Serna Saldivar et al. 1990).

Dry masa flour is reconstituted with tap water to produce masa. The correct addition of water and adequate mixing are the most critical steps for obtaining the best masa properties. The flour is blended with the water in a slow and gentle manner for times between 5-10 min. The amount of water commonly required to hydrate masa flour for tortilla production is 1.1 to 1.2 L per kg of dry masa flour (Serna Saldivar et al. 1990; Gomez et al. 1987). In large processing plants, the production of fortified and enriched dry masa flour, such as the one proposed and described in this study, will not cause any logistic problems because the required equipment to perform the fortification or enrichment is available. For instance, soybean meal can be ground into the desired particle size using either hammer or roller mills existing in the plant. The ground soybean can be easily incorporated into the formulated dry masa flour at the blending step indicated in Fig. 1. Finally, the addition of an enrichment premix can be incorporated, in the predetermined dosage, along with other desired additives at the end of the dry masa flour formulation or diluted with nixtamalized flour to achieve a better distribution. Perhaps the only additional problems that the industry would encounter are the logistics of purchasing the soybean meal and vitamin/mineral premix and providing the facilities to store these products.

One of the big advantages of producing masa flours is that they can very efficiently reach the rural areas and small villages or towns. For instance, in Mexico DICONSA (Distribuidora e Impulsora Comercial de CONASUPO, S.A., where CONASUPO means Comercializadora Nacional de Subsistencia Popular; the Mexican subsidized food distribution agency) has distribution centers strategically located throughout the country. Due to the excellent distribution channels, the fortified/enriched

masa flours can reach practically all the population in Mexico. Ten or twenty years ago fortification would not have been possible because most of the tortillas were home-made or produced from fresh masa in small, local tortillerias.

Fortification of Dry Masa Flour

The Consortium MASECA designed a fortified/enriched dry masa flour by mixing 93.5% dry masa flour for table tortilla production with 6% defatted soybean flour and 0.5% of a vitamin/mineral premix which contains vitamins A, B₁, B₂, niacin, folic acid and iron (Serna Saldivar 1996a; Muñoz de Chavez and Chavez 1997). With the exception of folic acid and iron, these micronutrients were added in quantities that provided between 50 and 75% of the recommended daily requirements, according to the Instituto Nacional de Nutrición Salvador Zubirán (Muñoz de Chavez and Chavez 1997). The premix provided 130% of the folic acid and 100% of the iron recommended. The source of iron used was ferrous fumarate and provided approximately 4 mg/100 g nixtamalized flour. The fortification and enrichment have the objective of decreasing protein malnutrition (kwashiorkor), pellagra, anemia and xerophthalmia/xerosis.

Defatted soybean flour was added because numerous previous studies have clearly demonstrated the complementary effect of soybean protein in cereal based foods (Bressani et al. 1974, 1979; Serna Saldivar et al. 1988). Other advantages are its high availability, long shelf-life (it is practically free of lipids) and relative low cost. The concentration of 6% was chosen to improve the protein quality of the tortilla (almost doubles the PER) and to maintain the organoleptic properties of the tortillas without significantly affecting the cost. Six grams of defatted soybean flour with around 50% protein provide 3 g of protein rich in lysine and tryptophan, the two limiting amino acids in corn. With this level of fortification, the PER of the new tortilla mix is approximately 80% that of casein (Muñoz de Chavez and Chavez 1997).

For the mixing step of the premix with the flour, no additional equipment is required because the dry masa flour processors normally blend different streams of flours at the end of the process. The premix can be directly added during this last step. The equipment used is a blender. To add a premix containing vitamins and minerals to the

flour, this will be done with the equipment commonly used by the mills for blending flours at the final step of processing before storage.

Effect of Fortification/Enrichment on the Chemical Composition of Tortillas

Chemical Composition. Table 3 shows the chemical analyses of regular and fortified/enriched dry masa flours and the corresponding values for the processed tortillas (42% moisture). The moisture levels of the processed tortillas were evaluated in a different test. It was observed that the soybean-fortified tortillas retained more moisture after tortilla baking and cooling, and that therefore the fortified mix yielded more tortillas per kilogram of dry masa flour than the regular tortilla mix. This is due to the high water-retention capacity of the soybean proteins. This high water retention should also improve the textural properties of the tortillas. The improvement in water retention and texture of soybean flour have been previously documented by Serna Saldivar et al. (1988) for bread, by Gonzalez Agramon and Serna Saldivar (1988) for wheat flour tortillas and Serna Saldivar et al. (1988) and Bressani et al. (1974, 1979) for corn tortillas.

Table 4 shows a comparison between the nutrient composition of regular and fortified/enriched corn tortillas with wheat flour tortillas, wheat bread, Corn Flakes[®], meat, milk and egg (Serna Saldivar 1996a). As expected, the fortified/enriched tortilla system contained higher values of protein, minerals and crude fiber than regular tortillas (Tables 3 and 4). This is because soybean flour contains higher amounts of these components in comparison with corn. On the other hand, both regular and fortified tortillas have similar values of ether extract or fat. The fortified tortillas contain lower values of NFE or carbohydrates because they contain higher concentrations of protein, minerals and fiber. The protein content was the proximate composition value that changed the most. The fortified flour and tortilla contained approximately 2% and 1% more protein respectively, than their corresponding regular counterparts. This is very

relevant, especially for the nutrition of low-income socioeconomic groups that cannot afford animal foods.

Table 3 also shows the gross energy values determined with an adiabatic bomb calorimeter, the estimated digestible calories for both types of tortillas, and their starch contents. In previous studies with laboratory rats and swine, Serna Saldivar et al. (1988a, 1988b) found that around 93% of the gross energy of the tortillas was digestible (Table 1). This is due to the high amount of starch, that in practical terms is 100% digestible.

Table 3a. Nutrient Composition of Nixtamalized Regular and Fortified/Enriched Corn Flours and Their Corresponding Tortillas (100 g)^a.

Nutrients	Regular Maseca		Fortified/Enriched Maseca	
	Flour	Tortilla	Flour	Tortilla
Water (g)	10.4	42.0	10.4	42.0
Food energy (kcal)	368	236	366	234
Starch (g)	73.8	42.8	66.7	38.7
Total lipid (fat) (g)	1.7	1.1	1.9	1.2
Crude fiber (g)	0.9	0.6	1.1	0.7
Ash (g)	1.2	0.8	1.4	0.9
NFE (g)^b	77.4	50.1	74.7	48.3
Protein (g)	8.4	5.4	10.5	6.8
Amino acids (%):				
Phenylalanine	0.41	0.27	0.50	0.32
Histidine	0.28	0.18	0.31	0.20
Isoleucine	0.29	0.19	0.38	0.25
Leucine	1.12	0.73	1.17	0.76
Lysine	0.25	0.16	0.39	0.25
Methionine	0.19	0.12	0.23	0.15
Threonine	0.32	0.21	0.38	0.25
Tryptophan	0.034	0.020	0.041	0.024
Valine	0.40	0.26	0.49	0.32

Table 3b. Nutrient Composition of Nixtamalized Regular and Fortified/Enriched Corn Flours and Their Corresponding Tortillas (100 g)^a.

Minerals :				
Calcium (mg)	106	68	98	63
Phosphorus (mg)	285	182	290	186
Magnesium (mg)	102	65	111	71
Sodium (ppm)	325	208	295	189
Potassium (ppm)	315	202	440	282
Iron (ppm)	11.5	7.4	44.5	28.5
Zinc (ppm)	20.5	13.1	22.5	14.4
Copper (ppm)	1	0.6	1	0.6
Manganese (ppm)	5.5	3.5	7	4.5
Vitamins:				
Vitamin B ₁ (mg)	0.36	0.23	0.89	0.56
Vitamin B ₂ (mg)	0.11	0.07	0.38	0.24
Niacin (mg)	1.33	0.86	4.17	2.63

^a Data from Serna Saldivar (1996). The average moisture content of the nixtamalized flours was 10.4 % and for the tortillas 42 %.

^b Nitrogen Free Extract is an indication of total soluble carbohydrate content (starch, sugars, etc.)

This data shows that tortillas are a high-energy food and that the consumption of 5 tortillas (150 g) provides about 27% of the recommended daily intake for a 1-3 year old child (See Table 9 in Appendix). Serna Saldivar et al. (1988) found that the digestibility of the protein in the tortilla was around 85%, and that the addition of soybean protein did not affect protein digestibility values.

Table 5 shows the effect of soybean meal addition on the amino acid composition of the regular and fortified tortillas. For comparison purposes, the typical amino acid composition of other common foods is also presented. The fortified tortilla contains more protein (Table 3) but more importantly contains a better balance of essential amino acids (Table 5). The “chemical score” gives an indication of the quantity of the limiting amino acid (which for both types of tortillas is lysine), in contrast to the amount required by a growing infant (see Tables 10 and 11 in the Appendix). It is noteworthy that an infant requires about three times more lysine per kg of weight than an adult (See Tables 10 and 11 in the Appendix). The chemical score is a good indicator of protein quality because it correlates very strongly with nitrogen retention and growth tests such as biological value (BV), net protein utilization (NPU) and protein efficiency ratio (PER). The addition of soybean meal improves the quantities of the two limiting amino acids of corn (lysine and tryptophan) and therefore the chemical score, BV, NPU and PER (Serna Saldivar 1996a). Experiments performed by Bressani et al. (1974) and Serna Saldivar et al. (1988) clearly demonstrate that the addition of soybean to tortillas did not affect protein digestibility and clearly improved nitrogen retention and growth. Laboratory rats fed tortillas with 8% soybean flour grew twice as fast as animals fed control tortillas. The protein quality of fortified tortillas is slightly lower than that of animal products (Table 5), but much better than the protein quality of regular corn tortillas, flour tortillas, or corn flakes. A cost comparison among these products shows that the fortified tortillas provide the best cost-nutrition benefit. Another advantage of soybean fortification is that the fortified tortilla contains higher amounts of tryptophan, the amino acid that can be transformed into one of the most important vitamins in human nutrition: niacin, the vitamin which prevents pellagra.

Table 4. Proximate Composition of Regular and Fortified/Enriched Corn Tortillas in Comparison with Other Selected Foods (100 g)^a

Nutrient	Corn Tortilla		Bread Wheat	Wheat Tortilla	Corn Flakes	Meat	Milk	Egg
	Regular	Fortified						
Water (%)	41.9	43.3	36.9	29.3	2.6	56.3	87.8	75.5
Energy (kcal)	236	234	270	351	389	237	66	152
Protein (%)	5.4	6.8	8.3	7.2	8.1	31.1	3.2	12.0
Fat (%)	1.1	1.2	3.9	9.8	0.3	11.6	3.9	10.9
Crude Fiber (%)	0.6	0.7	0.3	0.2	0.4	---	---	--
Ash (%)	0.8	0.9	2.1	0.9	2.9	1.0	0.7	1.0
NFE (%)^b	50.2	47.5	48.5	52.6	85.7	0.0	4.4	0.6

^a Data from Serna Saldivar (1996). Values for regular and fortified corn tortillas were obtained from analysis while for the other foods were extracted from USDA (1979) nutrient composition tables.

^b Nitrogen Free Extract is an indication of total soluble carbohydrate content (starch, sugars, etc.)

Table 5. Protein Quality and Essential Amino Acids Content of Regular and Fortified/Enriched Corn Tortillas in Comparison with Other Selected Foods (100 g)^a

Amino Acid ^b	Corn Tortilla		Bread Wheat	Wheat Tortilla	Corn Flakes	Meat	Milk	Egg
	Regular	Fortified						
Phenylalanine	0.27	0.32	0.30	0.25	0.48	1.21	0.18	0.64
Histidine	0.18	0.20	0.18	---	0.23	1.06	0.10	0.29
Isoleucine	0.19	0.25	0.20	0.23	0.39	1.40	0.18	0.66
Leucine	0.73	0.76	0.41	0.40	0.69	2.46	0.34	1.04
Lysine	0.16	0.25	0.18	0.15	0.18	2.58	0.31	0.82
Methionine	0.12	0.15	0.08	0.12	0.17	0.80	0.08	0.39
Threonine	0.21	0.25	0.15	0.17	0.33	1.36	0.14	0.59
Tryptophan	0.02	0.02	0.09	0.06	0.02	0.35	0.06	0.19
Valine	0.26	0.32	---	0.26	0.49	1.51	0.23	0.79
Limiting AA	Lys/Trp	Lys/Trp	Lys	Lys	Lys	None	None	None
Chemical Score	44	72	40	41	41	>90	>90	100
Protein								
Digestibility	84	83	88	88	85	>90	>94	>95
Biological Value	50	68	48	46	45	>85	>85	>90
PER	0.9	1.9	1.1	1.0	0.7	2.3	2.6	2.8

^a Data from Serna Saldívar (1996). Values for regular and fortified corn tortillas were obtained from analysis while for the other foods extracted from USDA (1979) nutrient composition tables. The values of protein digestibility, biologic value and PER were obtained from the scientific literature.

^b Requirement of Amino acids in g/100 g protein is: 5.44 Lysine, 3.52 Methionine + Cystine, 4.0 Threonine, 7.04 Leucine, 4.0 Isoleucine, 4.96 Valine, 6.08 Phenylalanine + Tyrosine, 0.96 Tryptophan. To obtain the amino acids (g/100 g protein) divide the amino acid value between the protein percentage multiplying per 100. For example, the regular tortilla contains 0.13 % Lysine and 5.4 % protein; thus, $0.13 / 5.4 \times 100 = 2.4$ g/100 g protein. The chemical value is obtained by dividing the g/100 g protein of limiting amino acid enter the respective requirement for children $\times 100$. In a regular tortilla it would be $(2.4 / 5.44) 100 = 44.1$ %.

Minerals. As expected, there are significantly improved amounts of iron in the fortified tortilla (Table 6). The amount of iron present in the fortified tortilla was almost four times that in the regular tortilla (Serna Saldivar 1996a). Calcium and iron are the most relevant minerals in practical human nutrition. The first due to its high requirement for the formation and maintenance of bones (prevention of osteoporosis and osteomalacia) and teeth (see Table 12 in the Appendix), and the second because it is essential for production of hemoglobin and myoglobin (prevention of anemia and infectious diseases). Cereal-based diets are lacking in these two minerals (Serna Saldivar et al. 1996b). The calcium- and iron-rich foods are almost exclusively limited to dairy products and other non-dairy animal products, respectively. Tortillas contain moderate to high quantities of calcium due to the lime-cooking and steeping. Recent studies conducted by Serna Saldivar et al. (1991, 1992) demonstrated that the calcium in tortillas is well absorbed and highly bioavailable for growing rats, and also that the calcium of protein-fortified tortillas was better utilized than the calcium from regular tortillas. Serna Saldivar et al. (1991) estimated that around 50% of the calcium consumed by an average Mexican comes from nixtamalized products.

Vitamins. The addition of the enrichment premix increased the content of vitamin A (not specifically considered in this report), thiamine (B₁), riboflavin (B₂), niacin and folic acid (also not considered in this report), as was planned and expected (Serna Saldivar 1996a). The enriched nixtamalized flour contained approximately three times as much thiamine (B₁), riboflavin (B₂) and niacin as the regular flour (Table 7). Table 13 in the Appendix shows the average requirements for these vitamins. The addition of these B vitamins is undoubtedly beneficial for groups of people who depend on tortillas as their main staple. These vitamins are partially lost during the nixtamalization process and are critically important to prevent pellagra and other related disorders.

The addition of vitamin A to provide 100% of the daily requirement was considered necessary in this Mexican study, because tortillas have very low levels of this nutrient. According to the Instituto Nacional de Nutrición Salvador Zubirán, vitamin A deficiency is very common among infants and children in Mexico and Central America, especially those living in rural and urban marginal areas.

Table 6. Mineral Composition of Regular and Fortified/Enriched Corn Tortillas in Comparison with Other Selected Foods (100 g)^a

Nutrient	Corn Tortilla		Bread Wheat	Wheat Tortilla	Corn Flakes	Meat	Milk	Egg
	Regular	Fortified						
Minerals:								
Ca (mg)	68	63	126	42	3	13	115.0	53
Fe (mg)	0.7	2.9	2.8	1.5	6	3.7	0.1	2.0
Mg (mg)	65	71	22	85	12	23	11	12
P (mg)	182	186	100	77	63	235	92	202
K (mg)	201	281	110	99	92	263	140	135
Na (mg)	21	19	510	573	1238	71	---	---
Zn (mg)	1.3	1.4	0.6	0.6	0.3	10.2	0.4	1.3
Cu (mg)	0.6	0.6	0.1	0.1	0.7	0.1	---	0.1

^a Data from Serna Saldivar (1996). Values for corn tortillas were obtained from analysis while for other foods were extracted from USDA (1979) nutrient composition tables (per 100 g whole food).

Table 7. B-Vitamin Composition of Regular and Fortified/Enriched Corn Tortillas in Comparison with Other Selected Foods (100 g)^a

Nutrient	Corn Tortilla		Bread Wheat	Wheat Tortilla	Corn Flakes	Meat	Milk	Egg
	Regular	Fortified						
Vitamins								
B1 (mg)^b	0.23	0.56	0.47	---	1.3	0.1	0.0	0.1
B2 (mg)^c	0.07	0.24	0.31	---	1.5	0.3	0.2	0.1
Niacin (mg)^d	0.86	2.63	3.75	---	17.6	2.67	0.08	0.08

^a Data from Serna Saldívar (1996). Values for corn tortillas were obtained from analysis while for other foods extracted from USDA (1979) nutrient composition tables.

^b Vitamin B1 or Thiamin has high susceptibility to light, low to oxygen, high to heat and high to metals presence.

^c Vitamin B2 or Riboflavin has very high susceptibility to light, low to oxygen, low to heat and high to metal presence.

^d Niacin has low susceptibility to light, oxygen, heat and metal presence

However, the fortification of sugar with vitamin A in Central America has given excellent results, so the addition of vitamin A to tortilla flour in Central America would not be recommended because people can consume large quantities causing hypervitaminosis. Excess amounts of vitamin A are stored in the liver and can become toxic.

Table 8 compares the nutrients supplied by 150 g portions of tortillas and of meat, as a percentage of the daily nutrient requirements for infants. As mentioned before, the fortification with soybean meal increased significantly the protein and the two most limiting amino acids of corn: lysine and tryptophan. The consumption of 5 fortified tortillas (150 g) provides one half of the protein, and of the lysine and tryptophan required by a growing infant. The consumption of 300 g of fortified tortillas (less than 200 g of fortified masa flour) is sufficient to meet the protein and protein quality requirements in full. The fortified tortillas are a better source of calcium than meat and provide only slightly lower amounts of phosphorus, iron and riboflavin. It is noteworthy to mention that at current costs, one kilogram of meat is about as expensive as twenty kilograms of fortified tortillas (Serna Saldivar 1996).

The fortified/enriched tortillas are an excellent provider of calories, protein, calcium, iron, vitamin A and the B-vitamins, thiamine, riboflavin, niacin, and folic acid, nutrients that are consumed far below the recommended amounts by low-income 1 socioeconomic groups (see Table 13 in the Appendix). At the current prices of nixtamalized flours, soybean flour and enrichment premix, one kilogram of fortified/enriched tortilla flour would cost only 10-12 Mexican cents (1.2 to 1.4 US cents) more than one kg of regular tortilla flour.

HUMAN TRIALS CONDUCTED BY INSTITUTO NACIONAL DE NUTRICIÓN SALVADOR ZUBIRÁN

The nutritional value of regular and fortified/enriched tortillas is being tested during a two-year study (1996 and 1997) in two neighboring rural Otomi (indigenous Mexican) communities located in the state of Queretaro, México: the community of El Rincon (the experimental group that receives the fortified tortilla flour) and the community of Yosphi (the control group that receives the regular tortilla flour; Muñoz de Chavez and Chavez 1997).

Table 8. Nutrient Contribution from 150 g portions of Regular and Fortified/Enriched Tortillas in Comparison with Meat^a.

Nutrient	Daily Requirement for Infants	Regular Tortilla	Fortified/Enriched Tortilla	Meat
Calories	1300	27.5 %	26.7 %	27.3 %
Protein	26 g	31.2 %	47.9 %	115.0 %
Amino acids				
Lysine	0.832 g	28.8 %	45.1 %	465.1 %
Tryptophan	0.163 g	18.4 %	22.1 %	322.1 %
Minerals				
Calcium	800 mg	12.7 %	11.8 %	2.4 %
Phosphorus	800 mg	34.2 %	34.8 %	44.0 %
Iron	15 mg	7.4 %	28.5 %	37.0 %
Vitamins				
Thiamin	0.7 mg	49.3 %	120.0 %	21.4 %
Riboflavin	0.8 mg	13.1 %	45.0 %	56.3 %
Niacin	9.0 mg	14.3 %	43.8 %	45.5 %

^a Data from Serna Saldivar (1996).

Families having at least one child 5 years of age or less were selected and given either the regular or the fortified dry masa flour. A total of 125 families from the El Rincon community and 145 families from the Yosphi community are taking part in the study. The study was categorized as blind because these families do not know about the treatments. Both neighboring communities are very poor, with Otomi background. Some of the subjects do not even speak Spanish. The communities practice subsistence agriculture and do not even harvest enough food to fulfill their needs for the entire year. These communities were chosen because of their high consumption of tortillas (645 g in El Rincon and 539 g in Yosphi) and because they represent a good model for all the ethnic groups of Mexico. Moreover, previous field studies had showed that the total caloric or energy consumption was low (1714 kcal/capita/day for El Rincon and 1980 kcal/capita/day in Yosphi). Tortillas and related products (atole) are the staple food, providing more than 80% of the total caloric intake. In addition, they consume beans on 76% of the days and animal foods very rarely (less than on 8% of the days).

Dry masa flour was provided depending on the number of family members. The flour was distributed in lots of 2.5 to 3.5 kg/day corresponding to 350 to 400 g/day/person, a quantity very similar to the average consumption found in these communities (Muñoz de Chavez and Chavez 1997). The masa flour was mainly used to prepare tortillas although it was also consumed roasted, in the form of atole (a thin porridge).

Both types of flours were well accepted, and there was not any significant difference in the organoleptic acceptability of the products (Muñoz de Chavez and Chavez 1997).

The fortified/enriched tortilla samples obtained from the households had a protein and micronutrient content slightly lower than expected. The average protein content (dry basis) ranged from 7.9 to 11.3% (around 10% lower than expected). The available lysine was also slightly lower than expected (242 ± 52). Nevertheless, these values were higher than the ones corresponding to regular tortillas ($7.7 \pm 0.8\%$ protein and 139 ± 36 available lysine (Muñoz de Chavez and Chavez 1997)).

The study monitored weight gains and height for all infants and children every other month. Also a clinical evaluation was performed by specialists from Texas A&M University and Universidad de Querétaro. The data presented only includes values for the first year (march 1996 to march 1997). It is important to mention that during the months of May and June, at the start of the rainy season, both communities had epidemics of diarrhea and varicella or chicken pox (Muñoz de Chavez ,and Chavez, 1997).

The growth data is summarized in Fig. 2. Infants and children receiving the fortified masa flour grew 49% more than their counterparts fed the regular flour. The weight gains in the subjects fed the fortified/enriched mix were 92% of the expected for normal subjects, notwithstanding that at the beginning of the experiment the community suffered bouts of diarrhea, varicella and other common diseases. It is important to mention that the control subjects suffered a higher incidence of infectious diseases and in some instances weight losses were observed. In Fig. 3 it can be easily observed that subjects fed with the fortified tortillas had a lower incidence of moderate and severe malnutrition and had a higher proportion of individuals categorized as normal.

The protein fortification and enrichment with selected vitamins and iron also improved the nutritional status of adults (Muñoz de Chavez and Chavez 1997). At the beginning of the experiment, the body mass index classified 13.1% of the adult female members of the El Rincón community as malnourished (less than 17 kg/m²) and 28.3% underweight (17 to 20 kg/m²). In contrast, in the control community there were no subjects classified as malnourished and only 23.2% were underweight. After only five months of supplementation, there were no malnourished subjects in the experimental community and after 10 months, the percentages of underweight subjects were practically the same in both communities (around 21%). For males, there was a significant increase in the percentage of overweight subjects in the control community (from 22.3% to 31.5%) whereas in the experimental community the percentage did not change (15%).

The clinical observations showed that the condition of hair, nails and skin improved in the individuals consuming the fortified/enriched tortillas. In this group it was observed that abnormal hair, nails and skin decreased from 82% to 33%, 71% to 39%, and 65% to 21%, respectively.

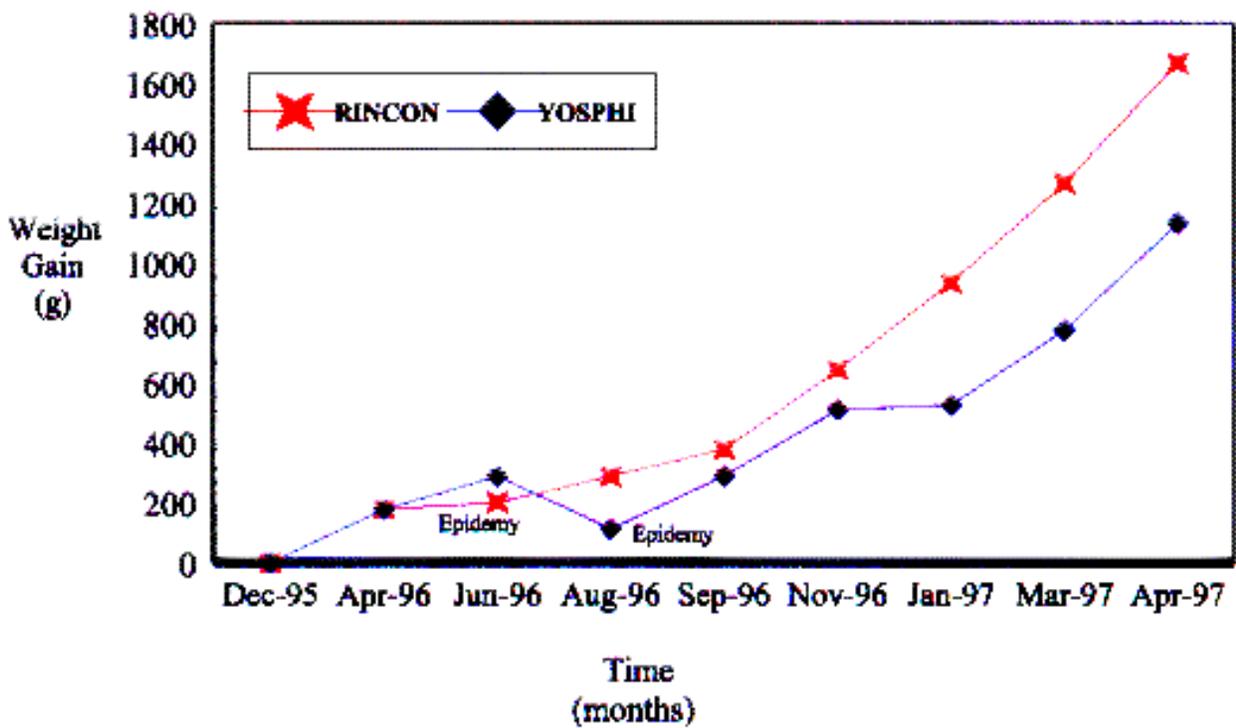
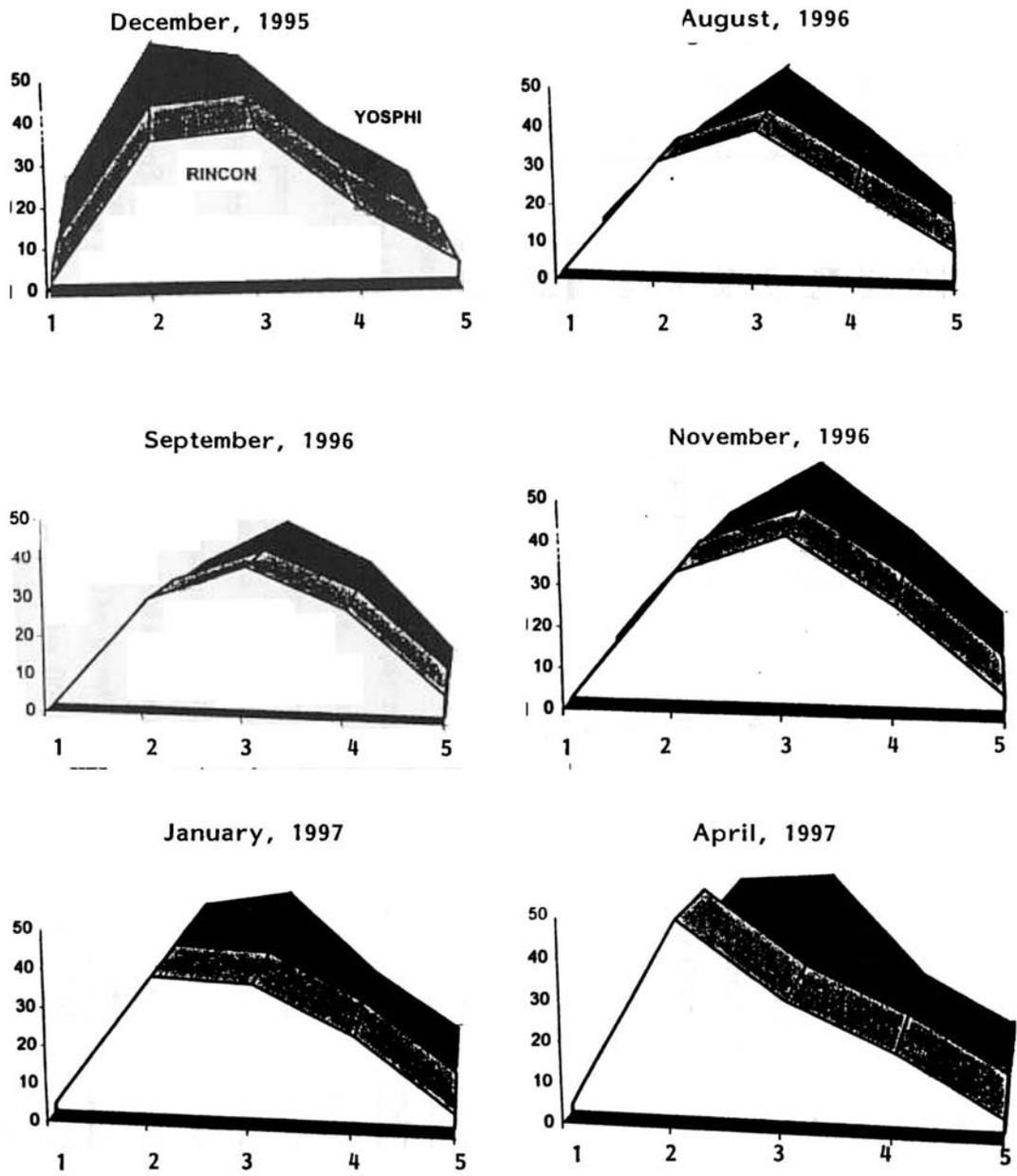


Fig. 2. Growth Rates of Children Fed Regular (Yosphi Community) and Fortified/ Enriched Tortillas (El Rincon Community; Muñoz de Chavez and Chavez 1997).

NUTRITIONAL STATE



1=Overweight; 2=Normal; 3=Slight Malnutrition; 4=Moderate Malnutrition; 5=Severe Malnutrition

Fig. 3. Comparison in Degrees of Malnutrition Between Subjects Fed Regular (Yosphi Community) and Fortified/Enriched Tortillas (El Rincon Community; Muñoz de Chavez and Chavez 1997).

In the control group subjects no significant clinical changes were observed, and at the end of the year they showed higher incidences of abnormal hair, nails and skin than subjects fed the fortified/enriched tortillas (Muñoz de Chavez and Chavez 1997).

For the experimental group, iron levels as measured by blood plasma hemoglobin in preschool- and school-age children showed a significant increase. Hemoglobin (iron) levels increased from 11.7 to 12.7 g/100 mL in preschool- and from 12.6 to 14.2 g/100 mL in school-age children. In contrast, children in the control community showed a slight, non-significant decrease in blood hemoglobin levels, especially school-age children (Muñoz de Chavez and Chavez 1997).

The folic acid (2.7 to 5.2 ng/ml) and vitamin B₁₂ (108.6 to 209.2 pg/ml) plasma values almost doubled after one year in the subjects fed the fortified tortillas, whereas the subjects in the control community maintained the same initial folic acid and vitamin B₁₂ values. Plasma levels of thiamine, riboflavin and pyridoxine at the beginning, middle, and end of the first year of the study for both communities are presented in Fig. 4. It can be easily observed that the plasma concentration of these vitamins stayed about the same for the subjects fed the control tortilla whereas those fed fortified tortillas almost tripled their serum thiamine and doubled their riboflavin and pyridoxine levels. This clearly demonstrates an acute deficiency of these essential vitamins, which can impair carbohydrate metabolism.

The liposoluble vitamins, A, E (tocopherol), and D were also measured, but the results were inconclusive. Perhaps the enrichment premix did not contain the specified quantities, or the amounts added were too small. As measured, both communities had similar blood retinol values. However, the group that received the fortified/enriched tortillas showed less clinical vitamin A deficiency (Muñoz de Chavez and Chavez 1997). This could be due to a synergistic effect with the other vitamins provided and especially with the better protein quality in the fortified tortillas. The retinol is transported in the blood bound to a protein, and in severe malnutrition this transport protein can be more critical than the concentration of the vitamin itself. Due to the importance of vitamin A for the adequate growth and health of children, it is recommended its bioavailability and quantities present in the enrichment premix be verified.

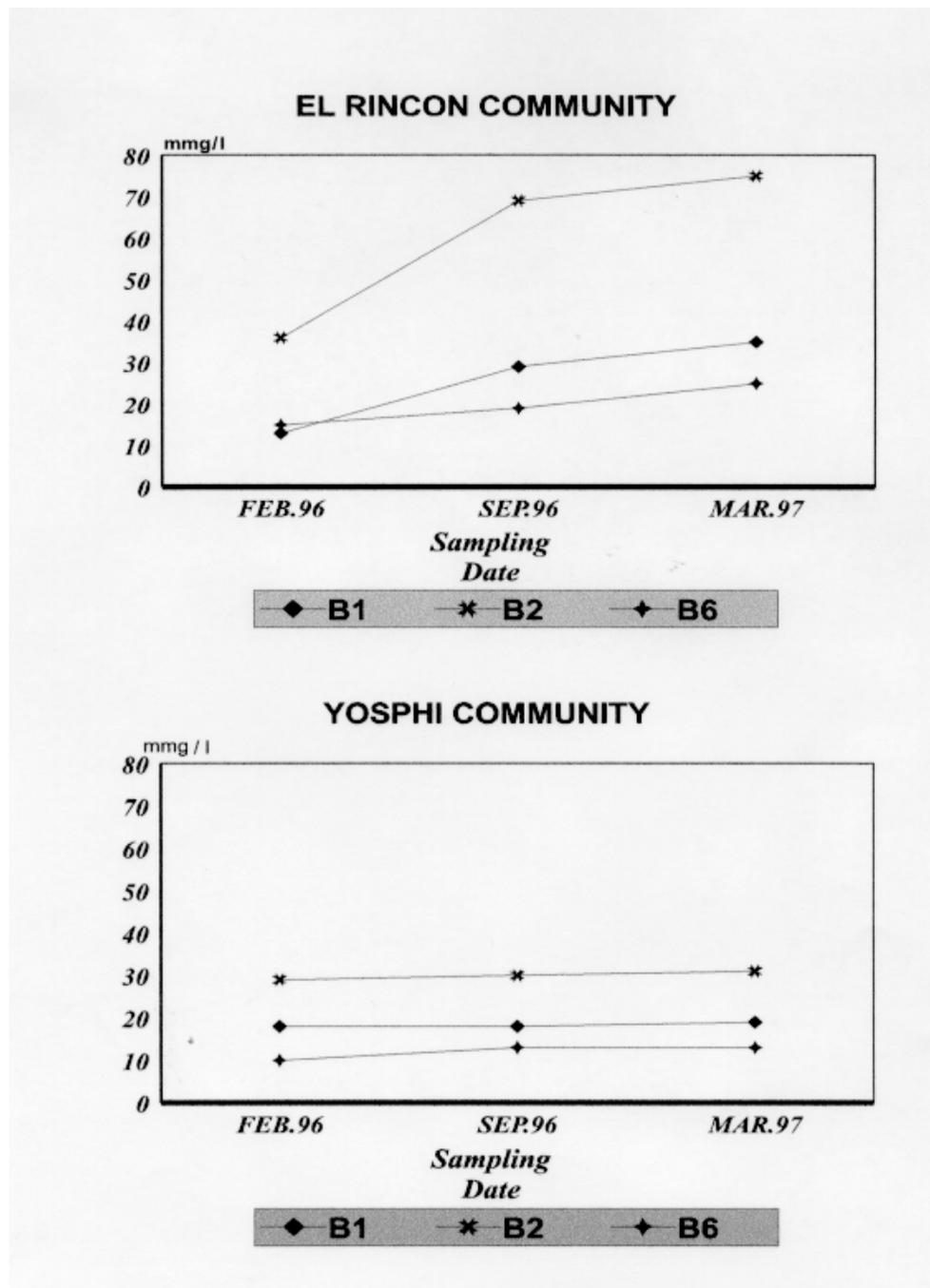


Fig. 4. Plasma Levels of Selected B vitamins for Human Subjects fed Regular (Yosphi Community) and Fortified/Enriched Tortillas (El Rincon Community; Muñoz de Chavez, and Chavez 1997).

Creatinine is a good indicator of the nutritional status. The concentration of this compound in urine is proportional to the lean body mass (muscle). It is notable that the people from the El Rincon community showed a 44% increment in creatinine, while in the Yosphi or control community the creatinine values stayed low throughout the study. This indicates that the addition of the soybean protein produced a quite significant increase in the metabolic lean body mass (Fig. 5).

CONCLUSIONS

Fortification of corn tortillas with soybean protein and enrichment with retinol, iron, thiamine, riboflavin, niacin and folic acid, produces a food that provides the nutrients in which the Mexican and Central American diets are most deficient. The best vehicle for these nutrients is industrially-produced dry nixtamalized corn masa flour. The production of this new mix should not cause industry any logistic problems because the industrial plants have already the equipment required to process and incorporate ingredients and enrichment premixes.

The nutritional study performed in the Otomi communities has clearly shown the benefits of soybean protein fortification and vitamin/mineral enrichment, especially for infants and children. The children fed the fortified/enriched mix grew almost 50% more than their counterparts fed regular tortillas. This is important, as previous experience has shown that it is very difficult to recuperate lost growth in infants malnourished during the first 20 months of life. More important is the fact that a high proportion of the malnourished subjects changed their nutritional status to normal. The behavior and attitude of the infants fed the fortified tortillas improved; they were happier and more self-confident.

Similarly, malnutrition cases among women from El Rincon practically disappeared. On the other hand, some of the adults from Yosphi ended the first year of the study with excess weight. This probably happened because the daily calorie intake increased due to the fact that the flour was given at no cost, and because the low protein quality did not enhance deposition of lean body mass.

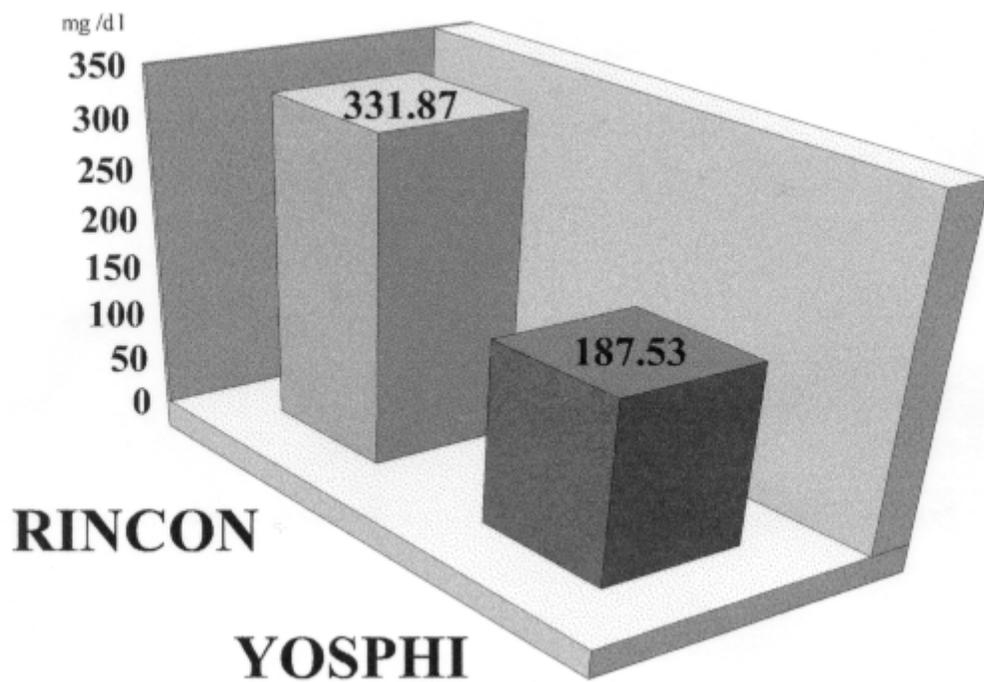


Fig. 5. Urine Creatinine Levels Levels for Human Subjects Fed Regular (Yosphi Community) and Fortified/Enriched Tortillas (El Rincon Community; Muñoz de Chavez, and Chavez 1997).

The fortification and enrichment increased the serum B-vitamin levels to normal values. The changes in hemoglobin levels were slight, despite the fact that these communities are located 3,000 m above sea level. The cases of acute anemia observed at the beginning of the experiment disappeared, but more data has to be collected to conclusively prove that the intervention with iron enrichment has given beneficial results.

The chemical and nutritional results clearly show that the fortification and enrichment was very positive and cost-effective. In practical terms, the production cost of the fortified/enriched nixtamalized flour is only 7% higher than the regular tortilla flour.

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APPENDIX

Table 9. Recommended Energy Consumption Throughout the Life Cycle and According to the Physiological State.

Description	Age or Physiological Condition (years)	Weight (kg)	Height (cm)	Basal	----- Energy Requirement -----		
					Factor	Per kg Kcal	PerDay
Infants	0.0-0.5	6	60	320	---	108	650
	0.5-1.0	9	71	500	---	98	850
Children	1-3	13	90	740	---	102	1300
	4-6	20	112	950	---	90	1800
	7-10	28	132	1130	---	70	2000
Men	11-14	45	157	1440	1.70	55	2500
	15-18	66	176	1760	1.67	45	3000
	19-24	72	177	1780	1.67	40	2900
	25-50	79	176	1800	1.60	37	2900
	51+	77	173	1530	1.50	30	2300
Women	11-14	46	157	1310	1.67	47	2200
	15-18	55	163	1370	1.60	40	2200
	19-24	58	164	1350	1.60	38	2200
	25-50	63	163	1380	1.55	36	2200
	51+	65	160	1280	1.50	30	1900
Pregnancy	0-3 months						0
	3-6 months						+300
	6-9 months						+300
Lactation						+500	

Source: RDA (1989)

Table 10. Essential Amino Acids Requirement (mg/kg live weight) Throughout the Life Cycle

Stage Age Weight	Baby 3-4 Months 6 kg		Infants 2 Years 13 kg		Children 10-12 Years 45 kg		Adults + 20 Years 75 kg	
	mg/kg/day	mg/day	mg/kg/day	mg/day	mg/kg/day	mg/day	mg/kg/day	mg/day
Histidine	28	168	--	--	--	--	10	750
Isoleucine	70	420	31	403	28	1260	10	750
Leucine	161	966	73	949	42	1890	14	1050
Lysine	103	618	64	832	44	572	12	900
Methionine + Cystine	58	348	27	351	22	990	13	975
Phenylalanine + Tyrosine	125	750	69	897	22	990	14	1050
Threonine	87	522	37	481	28	1260	7	525
Tryptophan	17	102	12.5	163	3.3	149	3.5	263
Valine	93	558	38	494	25	1125	10	750

Source: RDA (1989).

Table 11. Essential Amino Acids Requirement (g/100 g dietary protein) Throughout the Life Cycle.

Amino Acid	Suggested Requirement (g AA/100 g protein)			
	Infant	Children		Adult
		2 - 5 Years	10 -12 Years	
Phenylalanine + Tyrosine	7.2	6.3	2.2	1.9
Histidine	2.6	1.9	1.9	1.6
Isoleucine	4.6	2.8	2.8	1.3
Leucine	9.3	6.6	4.4	1.9
Lysine	6.6	5.8	4.4	1.6
Methionine + Cystine	4.2	2.5	2.2	1.7
Threonine	4.3	3.4	2.8	0.9
Tryptophan	1.7	1.1	0.9	0.5
Valine	5.5	3.5	2.5	1.3

Source: FAO/WHO (1990).

Table 12. Essential Macro and Micro Minerals Daily Requirements (mg/day) Throughout the Life Cycle

Stage	Infants	Children	Teenagers		Adults	
Age	0 -1 Year	1-3 Years	11-14 Years		23 -50 Years	
Sex			Female	Male	Male	Female
Weight	6 kg	13 kg	45 kg	46 kg	70 kg	55 kg
Macrominerals						
Calcium	450	800	1200	1200	800	800
Phosphorus	300	800	1200	1200	800	800
Magnesium	60	150	350	300	350	300
Microminerals						
Iron	12.5	15.0	18.0	18.0	10.0	18.0
Zinc	4.0	10	15.0	15.0	15.0	15.0
Copper	0.8	1.3	2.5	2.5	2.5	2.5
Manganese	0.8	1.3	3.8	3.8	3.8	3.8

Source: RDA (1989).

Table 13. B-Vitamins Daily Requirements Throughout the Life Cycle.

Stage	Infants	Children	Teenagers		Adults	
Age	0 -1 Year	1-3 Years	11-14 Years		23 -50 Years	
Sex			Female	Male	Male	Female
Weight	6 kg	13 kg	45 kg	46 kg	70 kg	55 kg
Vitamin B1 ^a (mg)	0.4	0.7	1.4	1.1	1.4	1.0
Vitamin B2 ^b (mg)	0.5	0.8	1.6	1.3	1.6	1.2
Vitamin B6 (mg)	0.5	0.9	1.8	1.8	2.2	2.0
Vit. B12 (µg)	1.0	2.0	3.0	3.0	3.0	3.0
Niacin ^c (mg)	7.0	9.0	15	15	18	13
Folacin (µg)	38	100	400	400	400	400
Biotine (µg)	43	65	150	150	150	150

Source: RDA (1989).

^a Vitamin B1 or Thiamin has high susceptibility to light, low to oxygen, high to heat and high to metals presence

^b Vitamin B2 or Riboflavin has very high susceptibility to light, low to oxygen, low to heat and high to metal presence

^c Niacin has low susceptibility to light, oxygen, heat and metals presence.