

## NUTRITION REPORTS INTERNATIONAL

THE POTENTIAL NUTRITIONAL CONTRIBUTION OF OPAQUE-2 CORN<sup>1</sup>V. Valverde,<sup>2</sup> R. Martorell,<sup>3</sup> H. Delgado,<sup>2</sup> V. M. Pivaral,<sup>2</sup>L. Elías,<sup>4</sup> R. Bressani<sup>4</sup> and R. E. Klein<sup>2</sup>

## ABSTRACT

Dietary survey data (24-hour recall) on 762 women and 292 children from rural Guatemala showed that methionine and cystine were the most limiting amino acids in about 45% of the diets. Lysine was the most limiting amino acid in 31% of the diets in women and in 36% of those in children while tryptophan was most limiting in 20% and 12% of the diets in women and children respectively. If common corn (8.2% protein) were substituted by Opaque-2 corn (8.2% protein), all diets would become satisfactory in tryptophan and the percent of diets limited by lysine would decrease to 11% and 18% respectively in women and children. The percent of diets with a chemical score less than 75% would decrease from 35% to 15% in women and from 38% to 17% in children. NDpCal% values would also rise significantly, particularly if high protein Opaque-2 corn varieties, which contain as much as 11% protein, were consumed.

## INTRODUCTION

Till recently, the shortage of protein was seen as the major nutritional problem of the less developed nations. This belief in a "protein gap" led to the development of many research programs and nutritional schemes whose aims were to increase protein quantity and quality in human diets. The development and marketing of vegetable protein mixes, the cereal-breeding programs, and the amino acid fortification projects, as well as the search for new and often exotic sources of protein, are examples of the kinds of strategies pursued and sometimes implemented.

The value of such efforts has been called into question by recent developments. Reassessments of protein-energy interrelationships (1, 2), supported by analyses of dietary data (1, 3-5), suggest that what is needed for improving dietary intakes in most developing nations is not more and better protein but more energy. The new strategy for resolving the prevailing energy and protein deficits calls for an increase in consumption of cereal-based diets (6). The policy implications are great as

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<sup>1</sup>This work was supported by the U.S. Agency for International Development (AID) Contract No. 931-17-560-625-73.

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it may be easier and cheaper to increase consumption of traditional food items than to introduce new, high-protein foods. However, agreement on the merits of the new strategy is by no means universal, for the necessary data to back these claims are far from complete (7, 8) and are open to several interpretations (9).

Research on corn breeding led, in the 1960s, to the development of Opaque-2 corn, unique in being high in lysine and in tryptophan (10, 11). Efforts, which continue to this day, were then begun to introduce the new variety into the corn-eating areas of the world, such as Mesoamerica, as a measure to better protein quality (12). Unfortunately the original mutant had a soft floury endosperm which weighed less than normal corn and yielded 10% to 15% less per hectare (3). In addition, the soft endosperm was more susceptible to damage by insects, Fusarium, and ear rot. Intensive research in Mexico at the "Centro Internacional de Mejoramiento de Maíz y Trigo" (CIMMYT) overcame most of the problems of low yields and high disease susceptibility through the development of a heavier, hard endosperm Opaque-2 variety (12, 13).

By selectively mating high-protein strains, researchers have also obtained varieties which exceed the usual protein content of Opaque-2 corn (11, 14). Also, high-protein, hard-endosperm Opaque-2 corn containing around 11% protein has been experimentally produced at INCAP from Opaque-2 seeds containing 8.2% protein by fertilizing the foot of the corn stalk at the flowering stage (15).

In spite of efforts to develop varieties suitable for cultivation in corn-eating areas, surprisingly little is known about the nutritional implications for a chronically malnourished population of substituting Opaque-2 corn for common corn. While the superior nutritional quality of Opaque-2 corn over common corn has been demonstrated in rats, chicks, and pigs as well as in children and adults in the metabolic ward (16) corresponding data from field studies are scant. A small study from India suggests that children receiving Opaque-2 corn grow better than those receiving common corn (17). However, large-scale field trials with lysine and other amino acids have not shown measurable benefits in nutritional status (18).

Many doubt that the widespread adoption of Opaque-2 by Mesoamerican populations would be nutritionally beneficent. The protein quality of the diets is not as low as would be expected from the quality of corn alone, for beans, which complement corn, are also consumed by these populations (19). Corn and beans provide approximately 60% of the protein intake of Guatemalan adults (20) and foods accounting for the other 40%, like rice, wheat, and to a lesser extent meat and dairy products, tend to have better amino acid patterns than the two staples (7).

The present paper is an attempt to estimate the potential dietary impact of Opaque-2 corn in Mesoamerican populations like those of rural Guatemala. A large sample of 24-hour recall histories were analyzed and indicators of protein quality and utilization of Opaque-2 and regular corn were compared to assess the potential effect of substituting one for the other.

## MATERIAL AND METHODS

The dietary data come from a continuing longitudinal study in coffee plantations on mountain slopes facing the Pacific Coast of Guatemala. The settlements are permanent, the heads of household being salaried agricultural workers employed throughout the year. Most inhabitants of the communities are Indian, though all speak Spanish reasonably well. The study area is known for its high prevalence of protein-calorie malnutrition. For example, anthropometric surveys reveal that 45% of the children under five years of age suffer from second and third degree malnutrition as defined by Gómez et al. (21).

Dietary surveys were available for women and for children less than five years of age. For the purposes of these analyses, diets during a two-month period of women and on children 24 to 47 months of age were selected for study. As one of the purposes of the analyses was to assess the protein quality in each meal relative to the total 24-hour intake, the few individuals not reporting data for all three traditional meals were excluded from the sample. The resulting sample consisted of 762 diets of women and 292 diets of children.

A modified 24-hour recall survey was used. All information was provided by the female head of the household and in local weights and measures. As the interfamily variance was large, family-specific weights and measures were used in assessing the amounts of foods consumed. Food composition tables developed for Central America were utilized to estimate nutrient intake (22). The protein score of each meal and of the total day were estimated using the amino acid pattern suggested by FAO/WHO (7). The amino acid patterns of Opaque-2 varieties were derived from Bressani (16). NDpCal% was calculated using protein scores (23) and appropriate corrections were made for protein concentration in diets (24, 25).

Three sets of values were generated, corresponding to the reported patterns of food intake but assuming that different kinds of corn were used. The corn varieties were as follows:

- (a) the corn actually consumed (common corn)
- (b) regular Opaque-2 corn (OC), and
- (c) high-protein Opaque-2 corn (HPOC).

Opaque-2 corn is the currently available hard-endosperm variety which contains 8.2% protein. High-protein Opaque-2 corn has been successfully produced from regular Opaque-2 seed at INCAP's experimental farm by fertilizing the foot of the corn stalk at the flowering stage and contains 11% protein of quality similar to that of its parent seed. The feasibility of producing HPOC in Mesoamerican farms is untested, and the HPOC is included in the analyses solely to estimate a maximum potential impact of the high-lysine corn. Indices of protein quality (score) and utilization (NDpCal%) corresponding to each type of corn were compared. For the purposes of this paper, the NDpCal% and protein score values were interpreted as if protein were the limiting nutrient, although energy may be limiting.

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Women weighed 47 kg and consumed 53.4 g of protein and 1764 calories daily. Children weighed 11.4 kg and consumed 20.2 g of protein and 734 calories. Women consumed 269 g of corn per day and children 78 g (20).

## RESULTS

The scores for those amino acids most likely to be limiting, lysine, methionine + cystine, and tryptophan, as well as NDpCal% values are shown in Table I for diets corresponding to the three corn varieties. Data are presented separately for women and children for each meal and for the total 24-hour period. As expected, the concentration of lysine and tryptophan as well as NDpCal% values increased when either regular Opaque-2 corn (OC) or high protein Opaque-2 corn (HPOC) were substituted for common corn; all appropriate paired t-test comparisons of these aspects between either of the Opaque-2 corn varieties and common corn were statistically significant ( $p < .05$ ). Also, as expected, the methionine-cystine concentration changed little ( $p > .05$ ) as a result of the substitutions. In fact, the sulphur-containing amino acids, methionine and cystine, became the limiting amino acids in the Opaque-2 diets. Diets for the two Opaque-2 corn varieties had similar mean scores ( $p > .05$ ) but because of the greater protein concentration, HPOC diets had larger NDpCal% values than OC diets ( $p < .05$ ).

The limiting amino acids generally associated with corn-based diets are lysine and tryptophan. The data in Table I, however, show that in the common corn diet, the limiting amino acids for breakfast as well as for the total 24-hour intake were methionine and cystine. It is also interesting to note that the NDpCal% values for the total 24-hour period were significantly greater than the values for each specific meal ( $p < .05$ ). Finally, in all of the patterns described above, no differences were observed between the diets of women and children.

A more detailed examination of the pattern of limiting amino acids is shown in Table II. As before, data are provided for women and children and for diets corresponding to the three varieties of corn. An analysis of the values generated for common corn reveals that methionine and cystine were the limiting amino acids in 47% of the diets in women and in 43% of those in children. Also lysine was limiting in about a third of the diets of both women and children while tryptophan was limiting in 20% of the diets of women and 12% of those in children. When regular Opaque-2 corn was substituted for common corn, the percentage of diets in which lysine and tryptophan were limiting decreased significantly ( $p < .05$ ). Conversely, the percent of diets in which methionine + cystine were limiting increased markedly ( $p < .05$ ). The percentage of diets with protein scores of 100 also increased from 2.5% to 9.0% for women and from 8.2% to 16.4% for children. Data for the HPOC variety are not appreciably different from those described above for regular Opaque-2 corn.

The frequency distribution of the protein scores of the diets of women is shown in Figure 1. Protein scores lower than 75% occur in 35% of the diets with common corn, 15% with OC, and 10% with HPOC. The calculations of children's diets show a similar pattern, 38% below a score of 75% with common corn, 17% with OC, and 14% with HPOC. For both

Table 1.--Amino acid scores ( $\bar{X}$ +S.D.) and NDpCal% ( $\bar{X}$ +S.D.) by meal in women and children for diets containing three types of corn

	Women (n=762)			Children 24-47 months (n=292)		
	Common corn	Regular Opaque-2 corn	High-protein Opaque-2 corn	Common corn	Regular Opaque-2 corn	High-protein Opaque-2 corn
<b>Breakfast</b>						
Lysine	85 $\pm$ 17	96 $\pm$ 9	96 $\pm$ 9	85 $\pm$ 18	94 $\pm$ 10	95 $\pm$ 9
Methionine + cystine	82 $\pm$ 13*	83 $\pm$ 13*	84 $\pm$ 12*	84 $\pm$ 14*	85 $\pm$ 14*	86 $\pm$ 13*
Tryptophan	85 $\pm$ 10	100 $\pm$ 0	100 $\pm$ 0	88 $\pm$ 11	100 $\pm$ 0	100 $\pm$ 0
NDpCal%	7.0 $\pm$ 2.0	7.6 $\pm$ 1.8	8.5 $\pm$ 1.7	6.8 $\pm$ 2.3	7.5 $\pm$ 2.0	8.2 $\pm$ 1.9
<b>Lunch</b>						
Lysine	80 $\pm$ 18*	94 $\pm$ 10	93 $\pm$ 9	82 $\pm$ 18*	93 $\pm$ 12	93 $\pm$ 11
Methionine + cystine	86 $\pm$ 13	87 $\pm$ 12*	88 $\pm$ 11*	86 $\pm$ 14	87 $\pm$ 14*	88 $\pm$ 13*
Tryptophan	84 $\pm$ 11	100 $\pm$ 1	100 $\pm$ 1	87 $\pm$ 11	100 $\pm$ 1	100 $\pm$ 1
NDpCal%	6.8 $\pm$ 2.4	7.9 $\pm$ 2.0	8.8 $\pm$ 1.9	6.9 $\pm$ 2.6	7.6 $\pm$ 2.4	8.4 $\pm$ 2.4
<b>Dinner</b>						
Lysine	80 $\pm$ 19*	93 $\pm$ 12	93 $\pm$ 11	82 $\pm$ 20*	92 $\pm$ 13	92 $\pm$ 13
Methionine + cystine	84 $\pm$ 14	85 $\pm$ 14*	86 $\pm$ 13*	84 $\pm$ 14	85 $\pm$ 14*	86 $\pm$ 13*
Tryptophan	85 $\pm$ 11	100 $\pm$ 1	100 $\pm$ 1	88 $\pm$ 11	100 $\pm$ 1	100 $\pm$ 1
NDpCal%	6.6 $\pm$ 2.4	7.4 $\pm$ 2.2	8.3 $\pm$ 2.2	6.5 $\pm$ 2.6	7.1 $\pm$ 2.4	7.8 $\pm$ 2.4
<b>Total 24-hour intake</b>						
Lysine	87 $\pm$ 13	98 $\pm$ 5	97 $\pm$ 5	88 $\pm$ 14	97 $\pm$ 10	96 $\pm$ 7
Methionine + cystine	85 $\pm$ 10*	86 $\pm$ 10*	87 $\pm$ 9*	87 $\pm$ 11*	87 $\pm$ 11*	88 $\pm$ 10*
Tryptophan	87 $\pm$ 8	100 $\pm$ 0	100 $\pm$ 0	90 $\pm$ 9	100 $\pm$ 0	100 $\pm$ 0
NDpCal%	7.5 $\pm$ 1.6	8.1 $\pm$ 1.5	8.9 $\pm$ 1.4	7.4 $\pm$ 2.0	7.9 $\pm$ 1.7	8.6 $\pm$ 1.6

\* Limiting amino acid.

Table 1.--Amino acid scores ( $\bar{X}$ ±S.D.) and NDpCal% ( $\bar{X}$ ±S.D.) by meal in women and children for diets containing three types of corn

	Women (n=762)			Children 24-47 months (n=292)		
	Common corn	Regular Opaque-2 corn	High-protein Opaque-2 corn	Common corn	Regular Opaque-2 corn	High-protein Opaque-2 corn
<b>Breakfast</b>						
Lysine	85 ± 17	96 ± 9	96 ± 9	85 ± 18	94 ± 10	95 ± 9
Methionine + cystine	82 ± 13*	83 ± 13*	84 ± 12*	84 ± 14*	85 ± 14*	86 ± 13*
Tryptophan	85 ± 10	100 ± 0	100 ± 0	88 ± 11	100 ± 0	100 ± 0
NDpCal%	7.0 ± 2.0	7.6 ± 1.8	8.5 ± 1.7	6.8 ± 2.3	7.5 ± 2.0	8.2 ± 1.9
<b>Lunch</b>						
Lysine	80 ± 18*	94 ± 10	93 ± 9	82 ± 18*	93 ± 12	93 ± 11
Methionine + cystine	86 ± 13	87 ± 12*	88 ± 11*	86 ± 14	87 ± 14*	88 ± 13*
Tryptophan	84 ± 11	100 ± 1	100 ± 1	87 ± 11	100 ± 1	100 ± 1
NDpCal%	6.8 ± 2.4	7.9 ± 2.0	8.8 ± 1.9	6.9 ± 2.6	7.6 ± 2.4	8.4 ± 2.4
<b>Dinner</b>						
Lysine	80 ± 19*	93 ± 12	93 ± 11	82 ± 20*	92 ± 13	92 ± 13
Methionine + cystine	84 ± 14	85 ± 14*	86 ± 13*	84 ± 14	85 ± 14*	86 ± 13*
Tryptophan	85 ± 11	100 ± 1	100 ± 1	88 ± 11	100 ± 1	100 ± 1
NDpCal%	6.6 ± 2.4	7.4 ± 2.2	8.3 ± 2.2	6.5 ± 2.6	7.1 ± 2.4	7.8 ± 2.4
<b>Total 24-hour intake</b>						
Lysine	87 ± 13	98 ± 5	97 ± 5	88 ± 14	97 ± 10	96 ± 7
Methionine + cystine	85 ± 10*	86 ± 10*	87 ± 9*	87 ± 11*	87 ± 11*	88 ± 10*
Tryptophan	87 ± 8	100 ± 0	100 ± 0	90 ± 9	100 ± 0	100 ± 0
NDpCal%	7.5 ± 1.6	8.1 ± 1.5	8.9 ± 1.4	7.4 ± 2.0	7.9 ± 1.7	8.6 ± 1.6

\*Limiting amino acid.

Table II

Percent of diets by limiting amino acid<sup>a</sup>

Limiting amino acid	Women (n=762)			Children (n=262)		
	Common corn	Regular Opaque-2 corn	High protein Opaque-2 corn	Common corn	Regular Opaque-2 corn	High protein Opaque-2 corn
None	2.5	9.0	8.3	8.2	16.4	15.1
Lysine	30.7	11.0	13.0	36.3	18.2	19.9
Methionine + cystine	46.7	80.0	78.7	43.2	65.4	65.1
Tryptophan	20.1	0.0	0.0	12.3	0.0	0.0

<sup>a</sup>The lowest score for any of the essential amino acids designates the limiting amino acid.

women and children, the distribution of protein scores is similar for OC and HPOC varieties.

Figure 2 shows the cumulative distribution of NDpCal% values for the diets in women. Important effects of OC over common corn and of HPOC over OC are observed. Fifteen percent of the diets have an NDpCal% value lower than 6 for common corn while the corresponding percentages are 3% for OC and less than 1% for HPOC. Differences between the two Opaque-2 corn varieties are especially important for NDpCal% values lower than 7. As before, the pattern in children is similar. The percent of diets with an NDpCal% value lower than 6 is 23% for common corn, 8% for OC, and less than 5% for HPOC.

#### DISCUSSION

Lysine and to a lesser extent tryptophan are generally regarded as the most limiting amino acids in the corn-eating areas of the world because of their low concentration in corn. However, the present study shows that methionine and cystine are the most limiting amino acids in about 45% of the diets of women and in 36% of those of children while tryptophan is most limiting in 20% and 12% of the diets in women and children respectively. In Costa Rica, where diets include a wider variety of foods than in the population here studied, 35% of the diets in children had no limiting acid, 55% were poorest in methionine and cystine and only 10% were lowest in lysine (5). Both the Guatemalan and the Costa Rican study support the views of Miller (26) and Swaminathan (14) that the sulphur amino acids are the most likely to be limiting in developing countries.

Substitution of Opaque-2 corn varieties for common corn improved the availability of lysine and tryptophan and resulted in better protein scores and NDpCal% values. While the NDpCal% values for common corn diets were 7.5 and 7.4 respectively for women and children, the corresponding values for regular Opaque-2 corn were 8.1% and 7.9% and those

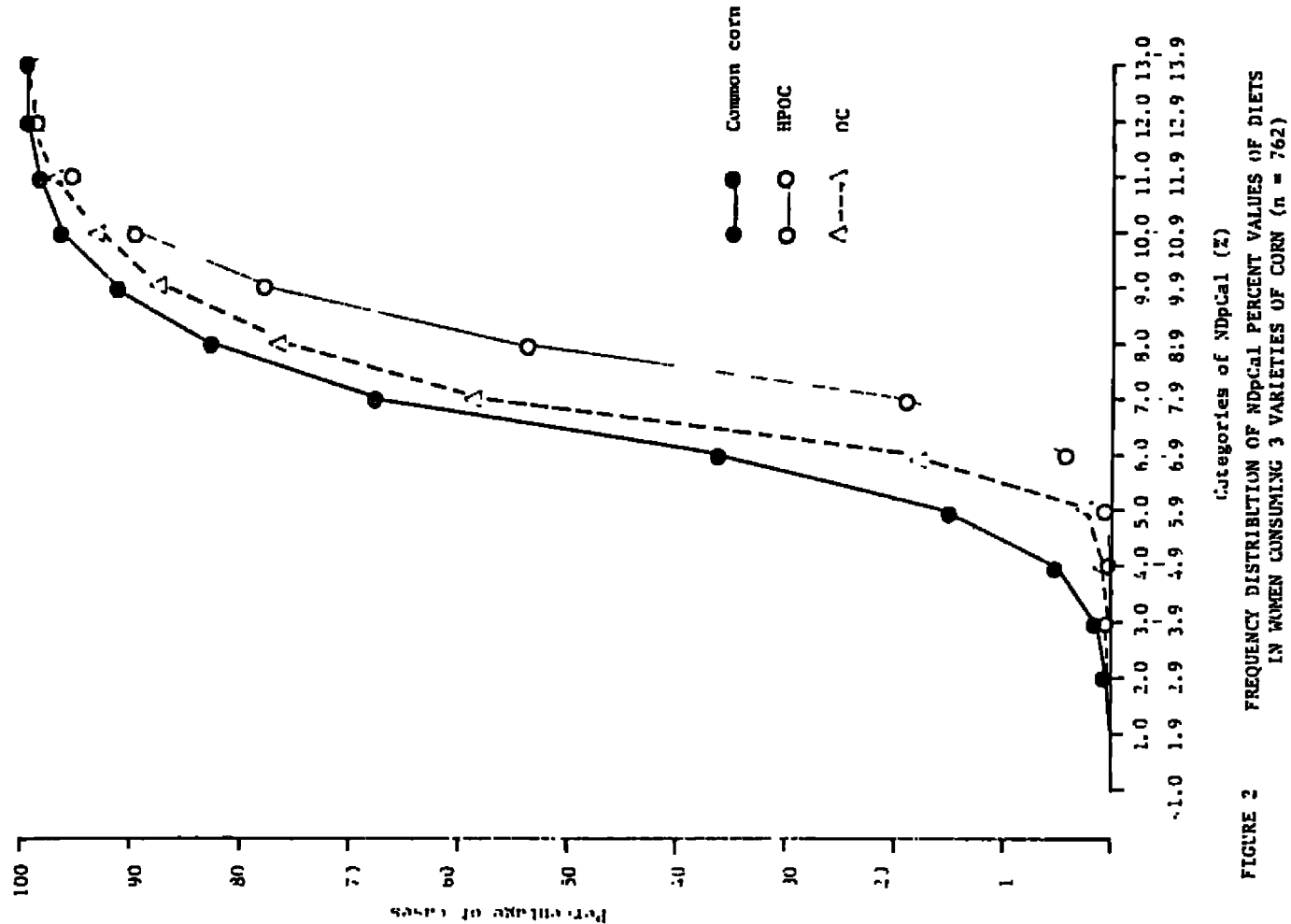


FIGURE 2 FREQUENCY DISTRIBUTION OF NDpCal PERCENT VALUES OF DIETS IN WOMEN CONSUMING 3 VARIETIES OF CORN (n = 762)

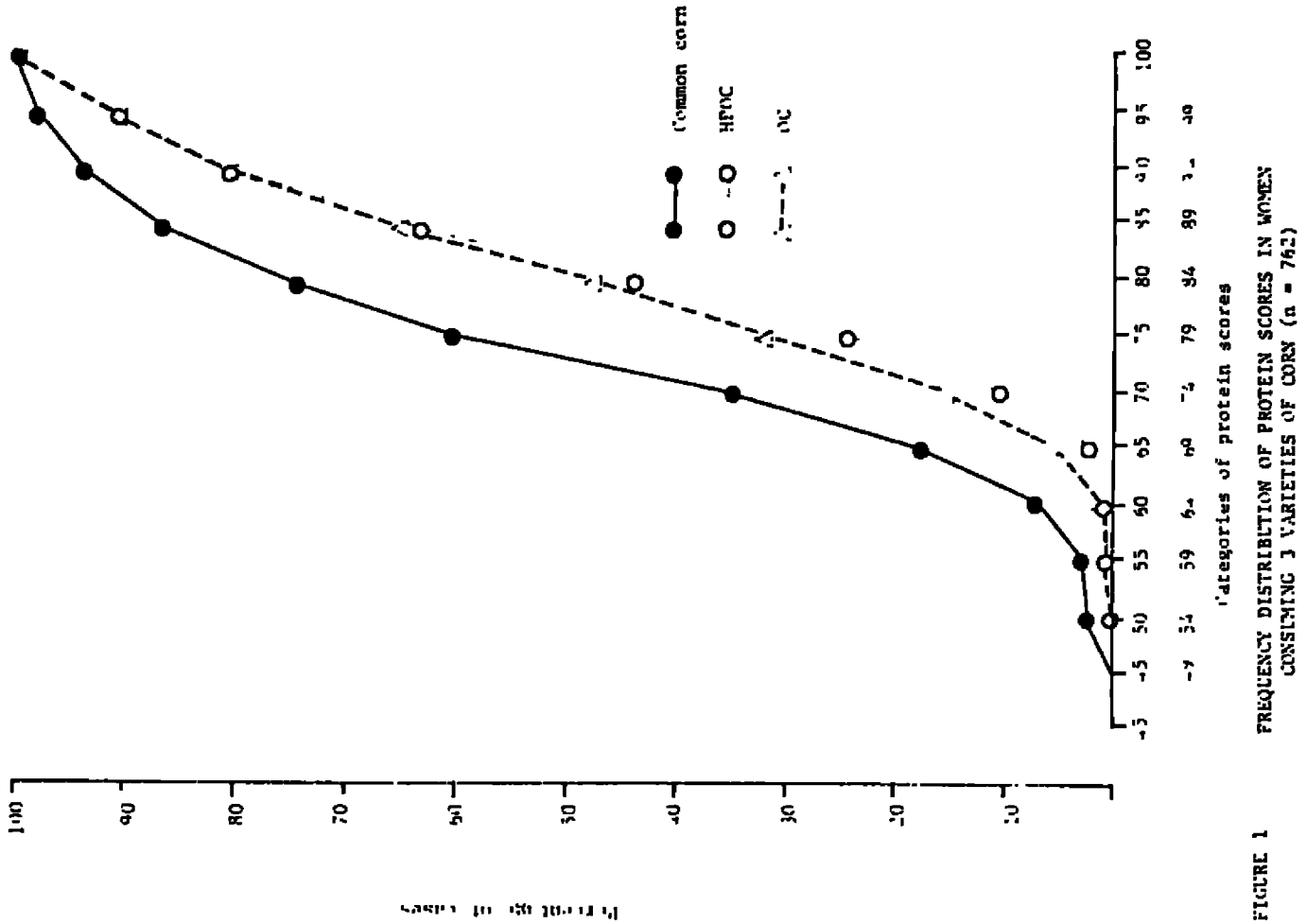


FIGURE 1 FREQUENCY DISTRIBUTION OF PROTEIN SCORES IN WOMEN CONSUMING 3 VARIETIES OF CORN (n = 762)



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for high-protein Opaque-2 corn were 8.9% and 8.6%. These improvements in protein utilization were shown to be statistically significant.

The results must be interpreted with caution on several accounts. The research model used assumes that food consumption will remain unchanged if Opaque-2 corn is substituted for common corn. It has been shown that the addition of lysine to wheat-based diets results in increased food consumption in rats (27). The situation in humans is difficult to predict for nonbiological factors such as food availability and purchasing power would also play an important role. No field study has, to our knowledge, evaluated the effects of improved protein quality on appetite and food intake.

The biological significance of improvements in protein scores and in NDpCal% values in the presence of energy deficits is currently unclear (28, 29, 30). It may be appropriate to distinguish among various possible situations in developing countries. For example, in subsistence farmers energy expenditure has been shown to be greater than energy intake during the hungry months which sometimes precede harvests (31). This may represent a situation in which significant portions of the ingested protein are broken down to provide energy and in which protein improvements may not be of value. On the other hand, during and after the harvest energy balance is positive as reflected by weight gains, a situation where additional protein is likely to be used for anabolic processes if the need exists. Still another situation is one where chronic limitations in energy intake lead to "adaptations" in the form of retarded growth and diminished physical activity causing energy input to equal energy output though at less than optimal energy levels. It is conceivable that in such a situation of energy equilibrium improvements in the amount of utilizable protein will enhance nitrogen retention rates.

While a single 24-hour recall dietary survey may, if well carried out, correctly estimate the population mean, it is a poor measure of the usual intake of individuals (32). The 24-hour recall survey will overestimate the true variance as well as the proportion of individuals falling below a "minimum" value denoting high risk. Payne (30) proposed that diets with an NDpCal% value higher than 5 present a low risk of protein malnutrition if energy needs are met. Our data show that for common corn, 5% of the diets in women (Figure 2) and 10% of those in children fall below this value. Because these are likely to be overestimates, it is clear that few cases will have, to begin with, low NDpCal% values as defined by Payne (2). This would clearly label the changes observed as biologically insignificant were it not for the fact that there is no agreement regarding protein-energy needs; indeed, Scrimshaw (9) would argue for much higher critical values of NDpCal%.

In characterizing the mean effects observed, the question arises as to which data should be used: the values for the various meals or the values for the total 24-hour period which were higher. Data on humans on the maximum period of time for which amino acids from different protein sources will still complement each other are scant. The absence of protein at breakfast or lunch has been shown to impair nitrogen utilization in young women (33).

For the reasons described above, the biological significance of the effects which may result from the substitution of Opaque-2 corn varieties for common corn cannot be fully determined with existing data. Definite answers may only come from field nutrition experiments designed to assess changes in nutritional status resulting from the introduction of Opaque-2 corn.

Discussions of the potential nutritional effects of Opaque-2 corn will remain academic until the factors limiting the widespread adoption of Opaque-2 corn are overcome. The problem lies in that the task of promotion is handicapped by the fact that there are no visible benefits to the farmer. Perhaps, the development of high-yield Opaque-2 varieties may be the factor to facilitate widespread adoption. But is it worth the effort to develop such a variety? Is it worth it to promote Opaque-2 corn in the meantime? It is agreed that these issues will be resolved as we increase our knowledge of protein-energy requirements and as we evaluate the biological impacts of field nutrition experiments.

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Accepted for publication: December 18, 1980.