En: Seed Protein Improvement in Cereals and Grain Legumes. Vol. 1. Vienna, Austria, International Atomic Energy Agency, 1979, p. 369-384.

Reprint from

"SEED PROTEIN IMPROVEMENT IN CEREALS AND GRAIN LEGUMES"

VOL.I

INTERNATIONAL ATOMIC ENERGY AGENCY
VIENNA, 1979

# GERM-ENDOSPERM RELATIONSHIP IN THE NUTRITIONAL IMPROVEMENT OF MAIZE GRAIN

F.R. POEY*, R. BRESSANI, A.A. GARCÍA*,
M.A. GARCÍA**, L.G. ELÍAS
Instituto de Nutrición de Centro América
y Panamá (INCAP),
Guatemala, C.A.

Abstract

GERM-ENDOSPERM RELATIONSHIP IN THE NUTRITIONAL IMPROVEMENT OF MAIZE GRAIN.

**Germ influence on the whole kernel and considerations on evolutionary, genetic, biological and nutritive effects of germ and endosperm favours the hypothesis that "it is possible to select high maize grain yield with high protein quality and quantity due to the independence of genetic and environmental factors that determine these variables". A series of four biological evaluations of 9, 12, 15, 18, 21 and 24% germ diets, obtained by adding ground germ or endosperm tissue to shelled grain, demonstrated that 21% germ represents the maximum nutritional value, equivalent to approximately 80% of casein with methionine as the most limiting amino acid as germ percentage increases. Net protein ratio values were obtained on six male weanling rats per treatment fed ad libitum for 10 days on diets balanced for protein, oil and amino acids to pursue the objectives of food and protein utilization efficiency, nutritive value of the protein and identification of limiting amino acids. After one cycle of full sib mating in a recurrent divergent selection applied to CIMMYT's maize population Tuxpeño Planta Baja germ proportion averaged 13.3 and 9.9% in the divergent subpopulations with no negative effects on kernel weight per ear. Visual selection for large germ on previously yield-selected materials, followed by whole-kernel methionine selection is suggested as a methodology to improve the yield and nutritional quality of maize.**

## INTRODUCTION

Breeding to improve the nutritional quality of the maize kernel has received special attention in the last decade, especially through the use of the endosperm mutant *opaque-2.* This approach, however, has been associated with undesirable physical characteristics of the *opaque-2* kernel that seriously limit its acceptance by farmers and consumers. Other earlier approaches with normal endosperm maize trying to increase the germ proportion were also affected by relatively lower yield potential and so were discontinued or discouraged. Furthermore, it seemed that grain yield, protein content and protein quality were negatively

---

* Instituto de Ciencia y Technología Agrícolas, ICTA, Guatemala.
** Northrop Kim and Co., México, D.F.

correlated and thus when selecting for only one of those criteria the others would be reduced.

Recent breeding research, however, in cereal grains concludes positively about the simultaneous improvement of yield and protein quality and quantity. Wheat lines previously identified for high protein demonstrated transgressive segregation for both high and low protein content and for high lysine, suggesting good possibilities for simultaneous improvement of these traits [1]. High protein and acceptable grain yields in wheat had been reported earlier [2, 3]. In oats new varieties and experimental lines averaged from 1 to 2% higher protein content than other varieties compared at similar yield levels [4]. Among cereals rice has the best amino acid balance so breeders are mainly concerned in increasing protein content while maintaining or improving yield potential. Research being carried out at IRRI in the Philippines has identified lines with 2% more protein and comparable grain yield as commercial varieties. Furthermore, higher protein brown rice tends to have higher hardness scores and whole-grain yield during milling and also better resistance to abrasive milling than rice with normal protein content [5]. In relation to maize the negative correlation between yield and protein quantity and quality has been reported repeatedly [6–9]. However, critical evaluation of those reports and further evolutionary, genetic and biological considerations justified stating the following hypothesis [10]:

"It is possible to select high maize grain yield with high protein quantity and quality due to the independence of genetic and environmental factors that determine these variables."

The hypothesis was based, principally, on the following considerations:

(1) Biological functions of germ and endosperm are different and their protein constitutions are determined through different genetic mechanisms. Anthropocentric domestication first, and conscious methodology to improve yield later, have resulted in a larger development of the endosperm portion, thus reducing the nutritional influence of the germ in the whole kernel. Therefore, the actual proportion of germ to endosperm is a consequence of selection pressure and not a functional or biological relationship required by the plant itself.

(2) Reports on a negative correlation between protein (quality and quantity) and yield can be explained on the basis of samples where only one variable received selection pressure. Correlations estimated on samples previously unselected for protein or yield did not show significant correlations [10].

## GERM INFLUENCE ON THE WHOLE KERNEL

The functional specialization of the germ, responsible for the development of the plant, demands consistency of its protoplasmic tissue to ensure the

TABLE I. MEAN AND VARIABILITY IN KERNEL WEIGHT AND GERM PERCENTAGE OF 12 TROPICAL MAIZE VARIETIES [11]

| | Kernel weight (g) | | | % germ | | |
|---|---|---|---|---|---|---|
| | Within varieties | | Among varieties | Within varieties | | Among varieties |
| | Min. | Max. | | Min. | Max. | |
| Mean | 0.20 | 0.62 | 0.32 | 10.51 | 15.21 | 12.90 |
| Standard deviation | 0.04 | 0.08 | 0.12 | 1.57 | 6.35 | 1.65 |
| Coefficient of variation (%) | 12.38 | 25.37 | 36.38 | 11.19 | 51.96 | 12.86 |

primary cell division and differentiation according to the genetic code of its nuclear material. Natural and/or induced genetic modifications of its chemical components would interfere with its metabolic process causing lethality or abnormal development of the plant. Changes in the chemical composition of the endosperm, on the other hand, would not have such drastic effects on the normal development of the plant. It should be remembered also that the size of the endosperm is larger than what is functionally required for the germination process of the kernel.

In spite of its chemical consistency, the germ does influence the overall chemical components of whole kernel through its relative contribution to the grain weight. Moreover, within the germ the protein and oil contents offer variability for selection.

Ample variation for kernel weight and germ:endosperm ratio was demonstrated in 12 varieties from Mexico and Guatemala. Mean weight, standard deviations and coefficient of variation for kernel weight and germ percentage in the kernel indicated that kernel weight within populations varies less than among populations, while the germ percentage showed an opposite tendency. Table I summarizes this information [11].

Correlation measured between kernel weight and germ percentage resulted in a non-significant value of 0.005. Considering kernel weight as a major yield component, these results suggest good possibilities for improving the germ proportion without affecting the yield potential. Bjarnason and Pollmer [12] found also a non-significant correlation value of – 0.16 in a study of 92 ears of $S_1$ and $S_2$ lines from five groups of translucent, semitranslucent, opaque, IHP backcrosses to adapted inbreds and IHO synthetics. The germ contribution to the whole grain ranged from 9.5 to 18.5% of the weight on individual kernels and from 11.89 to 16.21% on a group basis.

Johnson and Vasal reported in 1972 [13] on 43 materials from very diverse origins, finding ranges of 3.1 to 23.6% of the total kernel protein contributed by the germ with a mean of 10.3%. The *opaque-2* versions of the same genotypes more than doubled that value with a mean of 22.4%, confirming an enhancement of the germ contribution resulting from pleiotropic effects of that mutant.

The variability in germ protein depends, to a high degree, upon the oil content of the germ [12, 14]. Oil is highly correlated to germ weight [15], therefore selection for larger germ may result in a relative reduction of its protein content. These and other correlations between oil and protein, however, generally involve samples selected for high oil with no simultaneous selection criterion applied for protein content.

## NUTRITIVE VALUE OF THE GERM AND ENDOSPERM

Reported values indicate between two and three times higher concentration of protein in germ than in endosperm; practically all the oil and three to four times higher lysine and tryptophan content are found in the germ while the sulphur amino acids content is lower than in endosperm [16, 17]. Besides increasing protein quantity and quality, more germ tissue in corn kernels will increase the caloric value as well. Table II includes typical values for protein, oil and amino acids for germ and endosperm [18–21].

The biological value of germ, endosperm and whole grain on four materials from Guatemala, including an *opaque-2* variety, demonstrated 50–100% higher efficiency of the germ than the endosperm as measured by net protein ratio (NPR). These results are reported in Table III as a percentage of the casein value.

The better biological values of germ over the endosperm and whole grain justify the possibility of improving the nutritional value of the maize kernel by breeding for higher germ proportion. Yield selection, however, should be considered simultaneously in order to avoid an indirect selection of smaller endosperm.

Alexander et al. [22] reported gain from two cycles of selection in an improved Lancaster population involving an index of yield and protein. In Iowa Stiff Stalk Synthetics (SSS) the correlation between yield and protein was – 0.20 in 1975 and + 0.34 in 1976. Realized genetic correlations were positive in both years in SSS, indicating that protein and yield may be positively correlated. Performance of $C_0 \times C_0$ and $C_1 \times C_1$ hybrids between the Lancaster and SSS populations confirmed the gains made in the parent populations. They also reported performance of hybrids with 80% higher oil content (8.2 versus 4.6%) than ordinary corn with yields that approach those of standard hybrids. Presumably those hybrids have larger germs than the standard hybrids.

TABLE II. REPRESENTATIVE VALUES OF NUTRITIONAL COMPONENTS OF GERM AND ENDOSPERM MAIZE TISSUE

| Component | Germ [18] | Endosperm [19] |
|---|---|---|
| Protein (%) | 23.2 [20] | 9.5 [20] |
| Oil (%) | 34.5 [21] | 0.8 [21] |
| *Amino acid* (g/100 g protein) | | |
| Lysine | 6.1 | 1.6 |
| Tryptophan | 1.2 [20] | 0.3 |
| Ammonia | 2.2 | – |
| Histidine | 2.9 | 2.9 |
| Arginine | 9.1 | 3.4 |
| Aspartic acid | 8.2 | 7.0 |
| Glutamic acid | 13.1 | 26.0 |
| Threonine | 3.9 | 3.5 |
| Serine | 5.5 | 5.6 |
| Proline | 4.8 | 8.6 |
| Glycine | 5.4 | 3.0 |
| Alanine | 6.0 | 10.1 |
| Valine | 5.3 | 5.4 |
| Cystine | 1.0 | 1.8 |
| Methionine | 1.7 | 2.0 |
| Isoleucine | 3.1 | 4.5 |
| Leucine | 6.5 | 18.8 |
| Tyrosine | 2.9 | 5.3 |
| Phenylalanine | 4.1 | 6.5 |

Bjarnason and Pollmer [12] also confirmed a wide variation in the proportion of germ and germ protein, but suggested that some compensatory effects may retard the improving of protein quality and quantity by selecting maize seeds with high germ proportion because of the observed trend to a decrease in the protein content of the germ.

High oil corn selection has shown better feed conversion ratios in finishing swine than ordinary corn [23]. It was found also that less protein supplement was required when the high-oil corn diet was used, confirming similar earlier reports [24].

TABLE III. NET PROTEIN RATIOS (NPR) OF WHOLE GRAIN GERM AND ENDOSPERM OF GUATEMALAN MAIZE VARIETIES
Expressed as percentage of casein.

| | Variety | | | |
|---|---|---|---|---|
| | Amarillo | Azotea | Cuarenteño | Opaco-2 |
| Whole grain | 42.5 | 44.3 | 65.4 | 81.4 |
| Germ | 65.7 | 80.4 | 90.6 | 85.0 |
| Endosperm | 40.9 | 42.0 | 46.4 | 77.0 |
| Casein | 100.0 | 100.0 | 100.0 | 100.0 |

## BREEDING CONSIDERATIONS

Breeding for larger germ and grain yield simultaneously represents added production of protein and oil than just breeding for yield alone.

The energy cost to produce a protein or lipid molecule is higher than for a carbohydrate one. These costs have been estimated for plants to be 1 g of glucose to produce 0.83 g of carbohydrates, or 0.40 g of proteins or 0.33 g of lipids. Furthermore, added protein production requires an increment in the nitrogen input to the plant [25].

This argument may be interpreted as a selection dilemma where some grain component should be sacrificed on behalf of others. This would require, first, a plant that is producing grain at an optimum level of efficiency of available energy and nitrogen input. This optimum situation depends on genetic and environmental factors so complex in nature that it can be assumed that some intermediate factor or process becomes limiting before the energy and nitrogen availability to the final protein, oil and carbohydrate production occurs.

If this consideration is valid, much progress can still be made by improving the metabolic efficiency and environmental condition of the plant.

The energy utilization by the plant, particularly in tropical maize, is considered very inefficient [26, 27], suggesting that upon improving the plant efficiency by increasing the sink, energy that is not used could be channelled to added kernel production and kernel components under proper input of needed ingredients. The harvest index (grain/stover) for tropical maize has been reported as being between 0.26 and 0.36 with experimental yields rarely above 5 t/ha, while temperate materials show harvest indices between 0.50 and 0.60 with yield potential above 9 t/ha [28]. It is evident that the energy cost in tropical maize is quite high and that within these limits it should be improved.

TABLE IV. CHANGES IN GERM PERCENTAGE AND YIELD COMPONENTS AFTER ONE CYCLE OF DIVERGENT RECURRENT SELECTIONS FOR GERM:ENDOSPERM RATIO

| Selection | % germ | Weight per kernel (g) | Kernels/ear | Kernel ear weight (g) |
|---|---|---|---|---|
| High germ | 13.3 | 0.27 | 337 | 91.0 |
| Low germ | 9.9 | 0.31 | 269 | 83.9 |

TABLE V. EFFICIENCY OF FOOD UTILIZATION MEASURED AS NPR, WEIGHT GAIN, AND FEED EFFICIENCY OF DIETS CONTAINING VARYING GERM:ENDOSPERM RATIOS[a] [30]

| % germ | NPR | Weight gain (g) | Feed efficiency[b] |
|---|---|---|---|
| 12 | 2.27 ± 0.12 | 3.8 ± 0.50 | 17.18 ± 0.92 |
| 15 | 2.87 ± 0.16 | 6.5 ± 0.48 | 10.30 ± 0.89 |
| 18 | 3.09 ± 0.17 | 10.2 ± 1.47 | 7.92 ± 1.09 |
| 21 | 3.24 ± 0.17 | 11.7 ± 1.58 | 6.98 ± 1.18 |
| 24 | 3.01 ± 0.17 | 9.3 ± 1.69 | 8.22 ± 0.18 |
| Casein | 4.10 ± 0.18 | 27.5 ± 1.73 | 3.14 ± 0.18 |

[a] Diets were not standardized for protein or oil content.
[b] Feed intake/weight gain (FE).

Breeding research currently conducted at Chapingo, Mexico, selecting divergently for high and low germ proportion and for high yield in both subpopulations should contribute information on the feasibility of improving yield and nutritive value in normal endosperm tropical maize population. CIMMYT's maize population Tuxpeño Planta Baja is being subjected to recurrent divergent selection for high and low germ proportion in the kernel and also for yield. Two divergent subpopulations were formed, respectively, with the highest and lowest 13% families on basis of germ percentage from 400 original selfed ears. After one cycle of full-sib mating on each subpopulation germ proportion averages

TABLE VI. NUTRITIVE VALUE OF PROTEIN MEASURED AS NPR, WEIGHT GAIN, AND FEED EFFICIENCY FOR DIETS CONTAINING VARYING GERM:ENDOSPERM RATIOS[a]

| % germ | Protein content (%) | NPR | Average weight gain (g) | FE |
|---|---|---|---|---|
| 9 | 7.6 | 2.25 ± 0.10 | 3.5 ± 1.83 | 21.77 ± 0.64 |
| 12 | 7.9 | 2.76 ± 0.12 | 6.3 ± 0.87 | 11.51 ± 0.30 |
| 15 | 7.8 | 3.32 ± 0.08 | 10.0 ± 0.62 | 7.53 ± 0.37 |
| 18 | 8.1 | 2.98 ± 0.18 | 11.0 ± 0.91 | 7.71 ± 0.35 |
| 21 | 8.4 | 2.96 ± 0.16 | 11.8 ± 0.54 | 7.25 ± 0.24 |
| Casein | 10.4 | 4.20 ± 0.15 | 34.3 ± 1.37 | 2.77 ± 0.25 |

[a] Isocaloric diets, variable protein content.

were 13.3 and 9.9%, while kernel weight per ear averages 91.5 and 84.3 g respectively for the high and low germ selections. Table IV summarizes these data. Results of this research, so far, suggest no negative effects on yield upon selecting for high germ proportion assuming that kernel ear weight estimates field grain yield.

## BIOLOGICAL EVALUATION OF VARYING GERM:ENDOSPERM RATIO

The theoretically better nutritive value of the higher germ proportion in maize requires biological confirmation to determine a realistic and justified goal for breeding programmes. A series of biological evaluation trials of maize with varying germ:endosperm ratio were conducted at the Instituto de Nutrición de Centro América y Panamá (INCAP) in Guatemala.

Varying proportions of germ were obtained by the addition of ground germ or endosperm tissue to ordinary maize to simulate 9, 12, 15, 18, 21 and 24% germ in whole grain. In each experiment six male weanling rats were fed 10 days and the net protein ratio (NPR) measured and compared to a casein diet as reference. All diets were supplemented with a complete vitamin and mineral mixture to supply the rat's requirements for these nutrients. A white endosperm maize variety from the Guatemala highlands, Compuesto Blanco, was used in all the experiments and a tropical white endosperm variety, ICTA B-1,

TABLE VII. EFFICIENCY OF PROTEIN UTILIZATION OF TWO VARIETIES IN DIETS WITH VARYING MAIZE GERM:ENDOSPERM RATIO MEASURED BY NPR[a]

| % germ | NPR | | |
|---|---|---|---|
| | C.B.[b] | | B-1[c] |
| 12 | 2.87 ± 0.19 | | 2.92 ± 0.15 |
| 15 | 3.00 ± 0.24 | | 3.26 ± 0.19 |
| 18 | 3.66 ± 0.16 | | 3.34 ± 0.24 |
| 21 | 3.60 ± 0.10 | | 3.29 ± 0.22 |
| 24 | 3.61 ± 0.06 | | 2.89 ± 0.16 |
| Casein | | 4.91± 0.22 | |

[a] Isoproteic-isocaloric diets.
[b] Guatemala highland variety Compuesto Blanco (CB).
[c] Guatemala lowland variety ICTA B-1 (B1).

was also included in one of the experiments. The trials were planned to define the following objectives:

(1) Efficiency of food utilization
(2) Nutritive value of protein
(3) Efficiency of protein utilization
(4) Identification of limiting amino acid.

## 1. Efficiency of food utilization

Diets not standardized for oil and protein content enhance the effect of the increments of these components as they are supplied by the varying germ:endosperm ratios studied. NPR and feed efficiency were calculated. The casein reference diet was standardized at 8.2% protein and 5% oil content. Table V shows that 21% germ has the best NPR values, resulting in 79% efficiency compared with the casein control [29]. The decrease in NPR observed on the 24% germ diet was attributed to a deficiency in essential amino acids, probably methionine, rather than to protein level, since both weight gain and feed efficiency also decreased. The possibility also existed that a high oil content resulting from a high amount of germ may have reduced food intake, thus resulting in lower weight gain, NPR and feed efficiency.

TABLE VIII. LYSINE, TRYPTOPHAN AND METHIONINE CONTENT IN VARYING GERM:ENDOSPERM RATIOS OF TWO MAIZE VARIETIES

| % germ | Lysine | | Tryptophan | | Methionine | |
|---|---|---|---|---|---|---|
| | Comp B1 | ICTA B-1 | Comp B1 | ICTA B-1 | Comp B1 | ICTA B-1 |
| | | | (g/100 g protein) | | | |
| 12 | 2.2 | 1.7 | 0.6 | 0.5 | 2.5 | 2.0 |
| 15 | 2.9 | 2.4 | 0.8 | 0.7 | 2.2 | 2.0 |
| 18 | 3.4 | 2.6 | 0.9 | 0.7 | 1.9 | 1.8 |
| 21 | 3.5 | 2.7 | 0.9 | 0.7 | 1.7 | 1.5 |
| 24 | 4.0 | 3.2 | 0.9 | 0.9 | 1.5 | 1.4 |

## 2. Nutritive value of protein

Similar diets were prepared with the same material standardized for oil content in order to study the effect of the total protein as it increases in the varying germ:endosperm samples. The control diet of casein was standardized at 10% protein in addition to a similar oil content.

Table VI shows that 15% germ gave the best NPR value and 21% germ the best weight gain. On the other hand, feed efficiency improved as germ proportion increased. In this particular situation the decrease in NPR was probably due to the increased level of protein in the diets as well as to a deficiency in essential amino acids. A common finding for all protein quality evaluation methods is that at low levels of protein intake there is a linear response which becomes quadratic as protein content in the diet increases.

## 3. Efficiency of protein utilization

Similar diets were standardized at 5% oil and 7.4% total protein in order to allow the quality of the protein to be the only variable. Compuesto Blanco (CB) and ICTA B-1 were the maize varieties used in this experiment. The casein control was standardized at 10% protein and same levels of oil. Results shown in Table VII suggest a limiting factor to the nutritional efficiency of protein utilization around the 18–21% germ proportion. As indicated previously, this could be due to diminishing sulphur amino acid content.

TABLE IX. LYSINE, TRYPTOPHAN AND METHIONINE SUPPLEMENTATION OF DIETS WITH VARYING GERM:ENDOSPERM RATIOS

| % germ | NPR | |
|---|---|---|
| | Supplemented[a] | Control |
| 13 | 3.78 ± 0.25 | 2.35 ± 0.16 |
| 15 | 3.89 ± 0.17 | 2.76 ± 0.24 |
| 18 | 3.30 ± 0.13 | 3.17 ± 0.05 |
| 21 | 3.12 ± 0.09 | 3.28 ± 0.13 |
| 24 | 3.18 ± 0.11 | 2.79 ± 0.13 |
| Casein | 3.94 ± 0.19 | |

[a] Isocaloric and isoproteic diets.

Analyses of the amino acids lysine, tryptophan and methionine conducted on the varying G/E ratios of the above samples shown in Table VIII clearly suggested that methionine is becoming the limiting one of the three, as its relative content is reduced as the germ proportion increases. It may also be seen that both lysine and tryptophan increase but methionine decreases as the germ ratio increases. The NPR of CB at 24% germ was higher and contained more lysine and methionine (Tables VII and VIII) than the NPR of the comparable value of Bl with lower levels of the two amino acids. These results suggest, therefore, that lysine may be still limiting after methionine.

## 4. Identification of limiting amino acids

Based on the lysine, tryptophan and methionine levels of the varying germ samples reported in Table VIII, diets were corrected to reach the higher levels of lysine and tryptophan of the 24% germ treatment and the higher methionine level of the 12% germ sample, considered at 4.0, 0.9 and 2.5%, respectively. Other treatments included additional supplementation with 0.1, 0.2 and 0.3% methionine to the 24% germ treatment, without further lysine or tryptophan supplementation to give confirmation on the supposed limiting sulphur amino acid. The casein reference diet was standardized at 7.3% protein and 7% oil. A control set of diets without protein, oil or amino acid supplementation was also included to provide a critical comparison of the supplemented diets. Results in Table IX show higher gains with lysine and tryptophan supplementation than with methionine at the lower germ percentage diet (which had higher supplementation of lysine and tryptophan). Methionine addition to the 24%

TABLE X. EFFECT OF METHIONINE SUPPLEMENTATION TO A 24/76, GERM/ENDOSPERM RATIO DIET AS MEASURED BY NPR

| Methionine supplementation % addition | NPR |
|---|---|
| 0 | 2.79 ± 0.24 |
| + 0.1 | 2.82 ± 0.12 |
| + 0.2 | 3.05 ± 0.17 |
| + 0.3 | 3.22 ± 0.12 |
| + 0.32 | 3.18 ± 0.11 |

germ diet increased protein quality from a NPR of 2.74 to 3.18; this increase was significantly lower than the NPR of natural corn supplemented with lysine and tryptophan. These results suggest, as previously indicated, that in the 24% germ diet other amino acids were limiting after methionine. The significance of methionine in partially improving the protein quality of germ is shown in Table X. Progressive supplementation with methionine in the 24% germ diets clearly shows the limitation of this amino acid at the higher germ levels. It is important to indicate, however, that endosperm tissue was also present, which even at a contribution of 76% provided more protein than germ tissue of the whole kernel. Thus it is possible to still have a secondary deficiency in lysine.

## BIOLOGICAL CONSIDERATION

The results of the present study confirm the thesis that the nutritional quality of whole maize kernels can be improved by increasing the proportion of germ in relation to endosperm in the kernel. Germ proteins are good sources of lysine and tryptophan, amino acids deficient in endosperm protein. On the other hand, germ tissue is deficient in methionine, which is present in a higher amount in endosperm proteins. Therefore, the proteins of the two tissues should complement each other.

The results of methionine supplementation to the 24% germ ratio, however, are not as outstanding as expected. No explanation is available at present, but it is possible that at this ratio of germ:endosperm lysine becomes also a limiting amino acid, second to methionine. Some evidence to support this contention was provided. Likewise, it is possible for germ tissue to be high in trypsin inhibitors, which have been reported to be present in such material [30, 31]. Since these substances are heat labile, cooked samples of 24% germ should give higher NPR values than those found in the present report. Such information has been reported for wheat germ [32].

Finally, the quality of germ protein is not as constant as reported by various workers, based on amino acid analysis. In the present study protein germ quality varied from about 66 to 91% of the protein quality of casein and it contributed significantly to the quality of the whole kernel. These results suggest, therefore, that besides selection for a higher germ ratio, selection for methionine content in whole kernel should be recommended for breeding programmes. Obviously, more work is needed both from the breeding aspects of the problem as well as from the chemical and nutritional characterization of the germ:endosperm relationship.

## CONCLUSIONS AND RECOMMENDATIONS

Biological evaluation of a varying germ:endosperm ratio demonstrated that 21% germ results in maximum nutritional value compared with approximately 80% of casein.

Selection for large germ on previously yield-selected material followed by whole-kernel methionine selection is suggested for the improvement of yield and nutritional quality of maize.

## REFERENCES

[1] MATTERN, P.J., JOHNSON, V.A., STROIKE, J.E., SCHMIDT, J.W., KLEPPER, L., ULMER, R.L., "Status of protein quality improvement in wheat", High Quality Protein in Maize, Dowden, Hutchinson and Russ, Stroudsburg (1975) 387.

[2] MIDDLETON, G.K., BODE, C.E., BAYLES, B.B., A comparison of the quality and quantity of protein in certain varieties of soft wheat, Agron. J. 46 (1954) 500.

[3] JOHNSON, V.A., MATTERN, P.J., SCHMIDT, J.W., "Genetic studies of wheat protein", Symposium Seed Proteins (INGLETT, G.E., Ed.), Avi, Westport (1972) 126.

[4] SCHRICKEL, D.J., CLARK, W.L., "Status of protein quality improvement in oats", High Quality Protein in Maize, Dowden, Hutchinson and Russ, Stroudsburg (1975) 400.

[5] JULIANO, B.O., BEACHELL, H.M., "Status of rice improvement", High Quality Protein in Maize", Dowden, Hutchinson and Russ, Stroudsburg (1975) 457.

[6] EAST, E.M., JONES, A.F., Genetic studies on the protein of maize, Genetics 6 (1920) 543.

[7] HAMILTON, T.S., "Nutritive value of corn proteins", Proc. 3rd Hybrid Seed Corn Industry-Research Conf. Am. Seed Traders Assoc. 3 (1948) 96.

[8] FREY, K.J., BRIMBALL, B., SPRAGUE, G.F., The effect of selection upon protein quality in the corn kernel, Agron. J. 41 (1949) 399.

[9] HOPKINS, C.G., SMITH, L.H., EAST, E.M., "The structure of the corn kernel and the composition of its different parts", Seventy Generations of Selection for Oil and Protein in Maize (DUDLEY, J.W., Ed.), Crop Science Society of America (1974) 33.

[10] POEY, F.R., El mejoramiento integral del maíz, Thesis, Colegio de Postgraduados, ENA, Chapingo, Mexico (1975).
[11] GARCIA, M.A., CONDE, A., BRESSANI, R., POEY, F.R., Kernel characteristics and measurement of the germ-endosperm ratio in maize, Agronomy Abstracts, Los Angeles (1977) 115.
[12] BJARNASON, M., POLLMER, W.G., The maize germ, its role as a contributing factor to protein quantity and quality, Z. Pflanzenzuecht. 68 (1972) 83.
[13] JOHNSON, E.C., VASAL, S.K., "Estudios para el mejoramiento del índice de proteína en maíz", Simp. desarrollo y utilización de maíces de alto valor nutritivo, Colegio de Postgraduados, ENA, Chapingo, Mexico (1973) 113.
[14] BRUNSON, A.M., ESRLE, F.R., CURTIS, J.J., Interrelations among factors influencing the oil content of corn, J. Am. Soc. Agron. 40 (1948) 180.
[15] DUDLEY, J.W., LAMBERT, R.J., ALEXANDER, D.E., "Seventy generations of selection for oil and protein concentration in the maize kernel", Seventy Generations of Selection for Oil and Protein in Maize (DUDLEY, J.W., Ed.), Crop Science Society of America (1974) 181.
[16] DUTRA DE OLIVEIRA, J.E., DE SOUZA, N., "El valor nutritivo de productos de la molienda del maíz, de la suplementación con aminoácidos y de mezclas de maíz común y de opaco-2", Mejoramiento Nutricional del Maíz (BRESSANI, R., BRAHAM, J.E., BEHAR, M., Eds), INCAP, Publicación L-1, Guatemala (1972) 209.
[17] ELIAS, L.G., "Utilización de subproductos de trigo y de maíz en la elaboración de alimentos ricos en proteína", Récursos Proteínicos en América Latina (BEHAR, M., BRESSANI, R., Eds), INCAP Publicación L-1, Guatemala (1971) 396.
[18] MERTZ, E.T., NELSON, O.E., BATES, L.S., VERON, O.A., Better protein quality in maize, Adv. Chem. 57 (1966) 228.
[19] NELSON, O.E., MERTZ, E.T., BATES, L.S., Second mutant gene affecting the amino acid pattern of maize endosperm proteins, Science 150 (1965) 1949.
[20] VILLASIS, J.E., Comportamiento de *Sitrotoga cerealella,* Oliver (Lepidoptera: Gelechiidae) y de *Sitophilus zea maiz,* Motschulsky (Coleoptera curculionidas) en diez tipos de maíz con características contrastantes, Thesis, Colegio de Postgraduados, ENA, Chapingo, Mexico (1971).
[21] EARLE, F.R., CURTIS, J.J., HUBBARD, J.E., Composition of the component parts of the corn kernels, Cereal Chem. 23 (1946)·504.
[22] ALEXANDER, D.E., LAMBERT, R.J., DUDLEY, S.W., "Breeding maize for oil and protein", Proc. VI Reunión de Maiceros de la Zona Andina, Lima, 1978 (in press).
[23] NORDSTROM, J.W., BEHRENDS, B.R., HEADE, R.J., THOMPSON, E.H., Effects of feeding high oil corns to growing-finishing swine, J. Anim. Sci. 35 2, 357.
[24] TERRILL, S.W., MOFFIT, J.G., KRIDER, J.L., EARLEY, E.B., BULL, S., Value of low and high fat corns for fattening hogs, J. Anim. Sci. 10 (1951) 902.
[25] BATHIA, C.R., RABSON, R., Energetic considerations in cereal breeding for protein improvement, Science 194 (1976) 1418.
[26] GOLDSWORTHY, P.R., COLGROVE, M.L., Growth and yield of highland maize in Mexico, J. Agric. Sci. 83 (1974) 213.
[27] GOLDSWORTHY, P.R., PALMER, A.E., SPERLING, D.W., Growth and yield of lowland tropical maize in Mexico, J. Agric. Sci. 83 (1974) 223.
[28] YAMAGUCHI, J., Varietal traits limiting the grain yield of tropical maize. II. The growth and yield of tall and short varieties, Soil Sci. Plant Nutr. 20 (1974) 145.
[29] GARCIA, A., POEY, F.R., BRESSANI, R., "Avances en el estudio del valor biológico del grano de maíz con variables proporciones de germen:endospermo", Proc. XXIV Reunión Anual del PCCMCA, San Salvador, El Salvador (1978)'M16/1.

[30] HALIM, A.H., WASSON, E.E., MITCHELL, A.L., Trypsin inhibitor in corn (*Zea mays* L.) as influenced by genotypes and moisture stress, Crop Sci. 13 (1973) 405.
[31] SWARTZ, M.J., MITCHELL, H.L., COX, D.J., REECK, G.R., Isolation and characterization of trypsin inhibitor from Opaque-2 corn seeds, J. Biol. Chem. 252 (1977) 8105.
[32] MORAN, T.T., Jr., SUMMERS, J.D., BASS, E.J., Heat processing of wheat germ meal and its effect on utilization and protein quality for the growing chick. Toasting and autoclaving, Cereal Chem. 45 (1968) 304.

## DISCUSSION

F.U. ZINK: Since nearly all the phytin is located in the germ, an increase in the proportion of germ in maize will involve an increase in phytin, and since this has a negative effect on the availability of micro-elements, particularly Zn, in the intestinal tract, an increase in the proportion of germ with a view to improving protein quality might give rise to problems with the uptake of Zn by man or animals.

R. BRESSANI: Biological tests with rats have shown increments in the nutritional value of maize samples up to about 18% germ, and a levelling-off above 20%; this limitation of nutritional value requires further study. Your point concerning micro-element availability deserves attention.

M. DENIĆ: Since there are some maize genotypes with more than one germ per kernel, would it not be appropriate to breed for this character as well?

R. BRESSANI: Nutritionally speaking, this might be of great interest, although it would give rise to some agronomic problems, such as determining an appropriate plant density or pruning the seedlings to allow for only one stalk. It should be remembered that a commercial yield is necessary as well as the extra nutritive value.

A. BOYAT: How did the yield behave in your selection for a higher percentage of germ?

R. BRESSANI: So far we can only infer yield from the grain produced per plant. The high-germ selection has not shown less weight than the low-germ selection. Yield trials are being conducted now with these selections.

J.R. JIMENEZ: Did the four Mexican varieties whose nutritional values were compared with *opaque-2* values carry the *opaque-2* gene? As you know, the *opaque-2* gene increases the size of the embryo.

R. BRESSANI: No, the four Mexican varieties were of the normal-endosperm type. The *opaque-2* material was also a Mexican variety (Tuxpeño), converted to *opaque-2*.

C.R. BHATIA: In general, it is true for most cereals that protein and lysine concentrations in the embryo are much higher than in the endosperm.

In wheat embryos, for instance, the major protein fractions are very different from those in the endosperm. The embryo contains mainly the water and salt-soluble proteins, albumins and globulins. In fact, embryo proteins are similar to grain legume seed proteins in their amino acid composition. If the approach outlined by you is successful, it would provide a combination of cereal and complementary legume-type proteins.