

# A NOTE ON THE CORRELATION BETWEEN LYSINE AND TRYPTOPHAN CONTENT IN MAIZE KERNEL ENDOSPERMS<sup>1</sup>

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## ABSTRACT

In maize kernel endosperm populations segregating for the *opaque-2* (high-lysine) mutant, the correlation between the lysine and tryptophan content is positive and highly significant in mixed kernel samples. However, in samples restricted to either *opaque-2* or normal endosperm types, the strength of this association is invariably reduced as indicated by correlation and regression coefficients. This anomaly arises from the fact that in respect to lysine and tryptophan content the *opaque-2* mutant separates segregating populations into

high and low subpopulations presumably with distinct means and variances, thus invalidating the correlations and regressions based on mixed kernel samples. The regression of lysine on tryptophan (as an indirect screening method for lysine content) is therefore incorrectly based if done on mixed kernel data and considerably reduced if based on pure kernel types. Critical comparisons and evaluations should therefore be based on more accurate and direct analytical methods.

The important discovery by Mertz et al. (1) that the *opaque-2* mutant brings about considerable increases in the lysine and tryptophan content of maize endosperm proteins brought with it analytical problems as efficient selection and progress in maize breeding programs would depend on an accurate and rapid method of screening for either or both of these amino acids.

Lysine determinations made by amino acid analysis, although accurate, are time-consuming and expensive so that alternative methods such as tryptophan evaluations were considered by various research workers (2,3). Colorimetric tryptophan analyses, it has been argued, could be done in much greater numbers than lysine analyses and would therefore be more suitable for screening, particularly if it could be shown that this method is reasonably accurate and that the tryptophan content is highly correlated with the lysine content in the endosperm proteins.

Hernandez and Bates (2) obtained a highly significant correlation of  $r = 0.85$  between the lysine and tryptophan content (expressed as a percentage of crude endosperm protein) in a mixed kernel sample of *opaque-2* and normal endosperms. On the basis of this result, it was recommended that the more rapid and simpler tryptophan method be adopted in protein laboratories and that at least in screening programs the co-variate lysine be estimated indirectly from the regression equation  $y = 0.3601 + 4.0745x$ , where  $y$  and  $x$  represent the lysine and tryptophan content, respectively. Consequently, this method, with minor modifications, has been adopted by both the protein laboratory of the International Maize and Wheat Improvement Center (CIMMYT) in Mexico and the Biochemistry department of Purdue University in the U.S. (3), with the provision that the more accurate actual lysine evaluations may still be required for purposes of monitoring in previously selected samples.

<sup>1</sup>Paper read at the Fifth Genetics Congress, Stellenbosch, Republic of South Africa, Feb., 1974.

## MATERIALS AND METHODS

In the course of the routine analysis of maize samples in the Biochemistry department of the University of Natal, Pietermaritzburg, numerous *opaque-2* and normal endosperm kernel types derived from segregating populations have been evaluated for tryptophan content by the method of Spies and Chambers (4), and for lysine content by the conventional amino acid analysis and later modifications of this standard procedure by Dennison (5).

For comparison with the results of Hernandez and Bates (2) the correlation between the tryptophan and lysine content as well as the regressions, regression equations and F-tests were calculated for a total mixed kernel sample of 110, as well as the constituent 71 *opaque-2* and 39 normal kernel types. These data together with a similar analysis of the data of Hernandez and Bates (2) are given in Table I. Scatter diagrams of the lysine and tryptophan content in all these samples are presented in Figs. 1 and 2.

TABLE I  
Estimated Correlation (r) and Regression (b) Coefficients Together with  
Regression Equations for Lysine Content (y) Derived from  
Tryptophan and Lysine Values in Local and Overseas Maize Endosperm Material

| Kernel Type                          | Sample Size | r     | b     | y              | F  |
|--------------------------------------|-------------|-------|-------|----------------|----|
| Local data                           |             |       |       |                |    |
| Mixed                                | 110         | 0.76  | 3.40  | $1.33 + 3.40x$ | ** |
| <i>Opaque-2</i>                      | 71          | 0.52  | 1.80  | $2.14 + 1.80x$ | ** |
| Normal                               | 39          | 0.25  | 0.96  | $1.49 + 0.96x$ | NS |
| Hernandez and Bates (2) <sup>a</sup> |             |       |       |                |    |
| Mixed                                | 55          | 0.85  | 4.07  | $0.36 + 4.07x$ | ** |
| <i>Opaque-2</i>                      | 7           | -0.29 | -1.71 | $5.29 - 1.71x$ | NS |
| Normal                               | 48          | 0.57  | 3.14  | $0.70 + 3.14x$ | ** |

<sup>a</sup>Data reduced to two decimals.

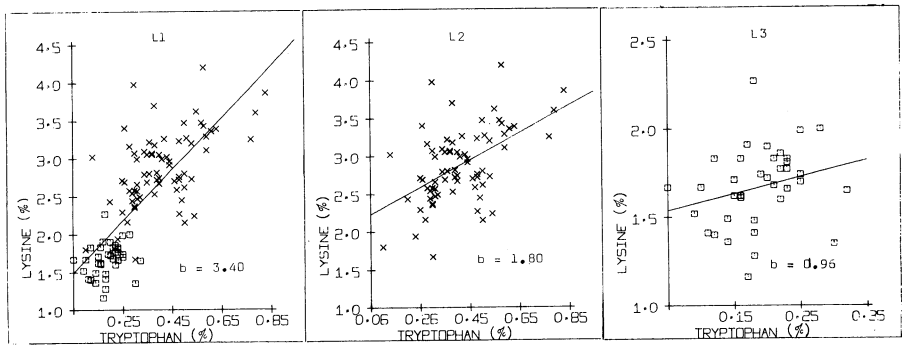


Fig. 1. Relationship between the lysine and tryptophan content (expressed as a percentage of crude protein) in local samples of mixed (L1), *opaque-2* (L2), and normal (L3) maize endosperms.

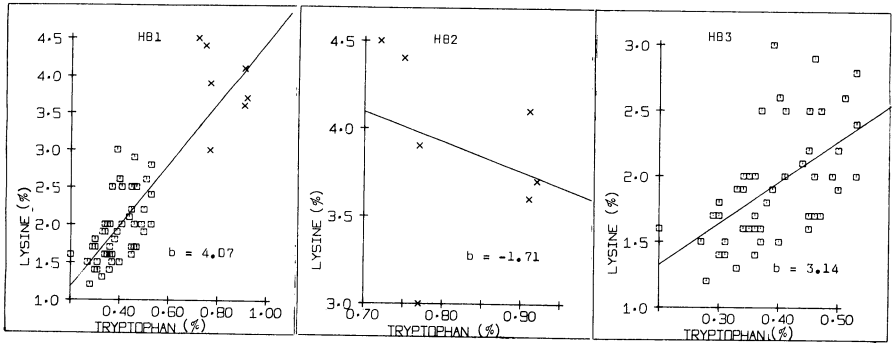


Fig. 2. Relationship between the lysine and tryptophan content (expressed as a percentage of crude protein) in samples of mixed (HB1), *opaque-2* (HB2), and normal (HB3) maize endosperms as derived by Hernandez and Bates (2).

## DISCUSSION AND CONCLUSIONS

Data in Table I show that the correlation between the lysine and tryptophan content is high and significant in mixed samples. However, the magnitude of the correlations, and hence, also, the regression coefficients and equations, change in various ways when only single kernel endosperm types are considered. The most noteworthy result is that the magnitude of these statistics is usually reduced while, in some cases, there does not appear to be a significant correlation between the amino acid values. In fact, the association is negative though not significantly so in the small *opaque-2* sample of Hernandez and Bates (2) (see Table I and Fig. 2).

An examination of the scatter diagrams (Figs. 1 and 2) shows that for mixed samples the *opaque-2* and normal components of these populations are at different extremities of the regression line. In terms of lysine and tryptophan content, segregation of the *opaque-2* mutant has therefore separated the sample into two distinct populations, presumably with different means and variances, thus invalidating any but intrapopulation comparisons.

The local data still show a reasonably high and significant correlation ( $r = 0.52$ ; 69 d.f.) between the amino acid values in the *opaque-2* sample but a nonsignificant figure in normal material. The data of Hernandez and Bates (2), again, show a relatively high correlation in normal material and a negative value in the *opaque-2* sample although the latter could possibly be due to an unusually small sample.

Although it must therefore be accepted that a high tryptophan value will with reasonable certainty also indicate a relatively high lysine value, one must view with some suspicion the high correlations and predictive values obtained with mixed samples. Likewise, these findings also suggest that the associations found between these two amino acids in pure sample types are of a magnitude that cannot warrant any but the most general screening procedure for the co-variate lysine which should, at least in elite, preselected samples, always be evaluated by more precise methods.

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