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Relationship between theoretical and experimentally detected tannin content of common beans (*Phaseolus vulgaris* L.)

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The present study was undertaken to determine the degree of any relationship between the theoretical and the experimentally determined tannin content in common beans. With this objective, theoretical tannin content was calculated as a function of seed coat tannin content and seed coat percentage weight in 19 varieties of common beans, grown in two localities of the Mexican highlands. The percentage of undetected tannins was calculated by the difference for all cultivars. The theoretical tannin content was correlated with the tannins detected experimentally in whole bean flour. The undetectable tannins ranged from 9.9 to 66.4% for varieties grown in Aguascalientes, and from 4.1 to 69.7% for cultivars harvested in Durango. A significant positive correlation was observed between the values of the theoretical tannin content and tannins detected experimentally in cultivars from Aguascalientes ($r=0.81$, $P<0.01$) and Durango ($r=0.72$, $P<0.01$). Nevertheless, there were many examples that showed the real content of tannins to have been underestimated. In accordance with this study, it is propose that, in order to obtain the real content of tannins of beans, these compounds should be determined in the seed coat flour. Copyright © 1996 Elsevier Science Ltd.

INTRODUCTION

Dry bean tannins have received considerable attention, largely as a result of their possible influence on the nutritional and aesthetic qualities of foods. Anti-nutritional effects of dry bean tannins have been reported to differ with animal species (Reddy *et al.*, 1985). Biological effects of tannins in humans vary considerably (Hughes, 1991; Morrow, 1991; Deshpande, 1992; Reyes-Moreno & Paredes-López, 1993).

Major amounts of bean tannins are located in the seed coat with low or negligible amounts in the cotyledons (Deshpande *et al.*, 1982). Tannins form complexes with cotyledon proteins, carbohydrates and other polymers in food. Although protein–tannin complexes are not detected by routine methods (Bressani *et al.*, 1982; Butler *et al.*, 1980), tannin content is usually determined in whole grain flour within 6 (Deshpande & Cheryan, 1987) to 24 h (Reyes-Moreno, 1992) of grinding. This may cause the tannins, from the seed coat, to form such complexes with proteins of the cotyledons. A probable consequence is an underestimation of the real tannin content, so making it difficult to identify varieties with high or low tannin contents.

The present research was undertaken to study the relationship between the theoretical tannin content based on that of the seed coat and the tannin content detected experimentally in whole grain flour, and to determine the degree to which tannins remain undetected owing to the presence of the proteins of cotyledons in whole grain flour.

MATERIALS AND METHODS

Dry beans

Nineteen varieties of common beans grown under rain-fed conditions in two different growing locations of the semi-arid highlands of México, Aguascalientes and Durango, were used in this study. Three replicates of every genotype were established in both growing locations.

Tannin content detected experimentally

Tannins were assayed according to the modified vanillin–HCl method of Price *et al.* (1978). Tannins were determined

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Table 1. Theoretical and experimentally detected tannin content in whole grain flour and percentage of undetected tannins in 19 varieties of beans grown in Aguascalientes*

Variety	Colour	Theoretical content of tannins (mg cat. eq./g)	Tannins detected experimentally (mg cat. eq./g)	Undetected tannins (%)
Ara 18	Yellow	9.6^{j}	8.7^{hi}	9.9
Feb 174	Brown	29.5^{cd}	26.5^{b}	10.0
Ara 797	Beige	37.8^{a}	32.4^{a}	14.2
MAM 42	Yellow	29.7^{bcd}	24.5^{c}	17.4
L-1213-2	Pinto	18.1^{h}	13.6^{f}	25.9
MAM 29	Yellow	18.8^{gh}	13.4^{f}	28.6
URG-4516	Brown	31.4^{b}	21.0^{d}	33.3
URG-3252	Pink	28.0^{d}	17.7^{e}	36.7
CIAT-326/85	Yellow	11.0^{j}	6.9^{j}	37.3
Río Grande	Yellow	14.3^{i}	8.75^{ghi}	38.6
Bayo Zacatecas	Beige	23.2^{f}	14.0^{f}	39.7
POB-2-SELL	Pinto	24.1^{ef}	14.1^{f}	41.4
1127-DM-2M	Pinto	23.4^{f}	13.6^{f}	42.1
San Luis	Black	23.6^{f}	13.2^{f}	43.9
G-13748	Pink	25.9^{e}	14.1^{f}	45.3
MAM 45	Yellow	14.2^{i}	7.3^{ij}	48.3
MAM 28	Beige	20.3^{g}	8.6^{hi}	57.7
Puebla 493	Black	22.4^{f}	8.9^{gh}	60.1
G-4523	Red	30.3^{bc}	10.2^{g}	66.4

*Mean of three replicate analyses, expressed on dry weight basis.
$^{a-j}$Means in the same column with different superscripts are significantly different ($P < 0.01$).

in the seed coat, whole grain flour, and cotyledons in triplicate immediately after grinding (80 mesh). (+)Catechin was used as the reference standard, and tannins were expressed in mg of catechin equivalent per g of dry sample (mg cat. eq./g).

In order to correct for interference from natural pigments in dry beans, a blank sample was prepared by subjecting the original extract to the same conditions of the reaction, but without vanillin reagent.

Theoretical tannin content

The theoretical content of tannins (TCT) of the samples was calculated as follows:

$$TCT = (SCT) \times (SC)/100,$$

where SCT is the seed coat tannin content (mg cat. eq./g) and SC is the seed coats percentage of the whole common beans.

Statistical analysis

Results were statistically treated by analysis of variance and Tukey's test (Cochran & Cox, 1975). Correlation coefficients were calculated where appropriated.

RESULTS AND DISCUSSION

Tannin detection

The undetected tannins of varieties grown in Aguascalientes ranges from 9.9 to 66.4% (Table 1). Varieties Ara 18, Feb 174, Ara 797 and MAM 42 exhibited less than 20% of undetectable tannins, 12 varieties gave 20–50%, and only cultivars MAM 28, Puebla 493 and G-4523 led to more than 50% of undetected tannins.

It can be seen in Table 1 that varieties with the highest theoretical tannin content (more than 20 mg cat. eq/g) did not give the highest values of tannins detected experimentally. In fact, there were no statistical differences ($P < 0.01$) between varieties MAM 42, URG 4516 and G-4523 in their theoretical tannin content, yet there were statistical differences ($P < 0.01$) between their content of tannins detected experimentally.

For cultivars grown in Durango, the undetected tannins ranged from 4.1 to 69.7% (Table 2). In this locality, seven varieties exhibited less than 20% of undetected tannins, 10 varieties gave 20–50%, and only Puebla 493 and CIAT-326/85 led to more than 50% of undetected tannins.

If values of the theoretical and experimentally detected tannins are compared, some examples bring out the incongruity clearly: varieties Ara 797 and San Luis did not present statistical differences between their values of theoretical tannin content, but there was a significant statistical difference in their contents of tannins determined experimentally. Varieties L-1213-2, G-4523 and MAM 29 gave identical results. There are other similar examples in both localities. No tannins were detected in the cotyledons in either case.

In spite of the observed significant positive correlation between the values of the theoretical tannin content and the tannins detected experimentally for varieties from Aguascalientes ($r = 0.81$, $P < 0.01$) and Durango ($r = 0.72$ $P < 0.01$), it is proposed on the basis of the results presented here that tannins be determined

Table 2. Theoretical and experimentally detected tannin content in whole grain flour and percentage of undetected tannins in 19 varieties of beans grown in Durango*

Variety	Colour	Theoretical content of tannins (mg cat. eq./g)	Tannins detected experimentally (mg cat. eq./g)	Undetected tannins (%)
URG-3252	Pink	20.5^{e}	19.6^{c}	4.1
L-1213-2	Pinto	12.6^{ij}	12.0^{ef}	4.4
G-13748	Pink	11.6^{j}	10.9^{gh}	6.0
Ara 797	Beige	28.2^{b}	26.0^{a}	7.9
G-4523	Red	12.3^{ij}	10.6^{gh}	13.9
MAM 45	Yellow	9.4^{k}	8.0^{jkl}	14.3
URG-4516	Brown	13.2^{i}	11.0^{gh}	16.7
Ara 18	Yellow	9.65^{k}	7.3^{kl}	24.2
MAM 29	Yellow	12.2^{ij}	9.3^{ijk}	24.7
Bayo Zacatecas	Beige	15.5^{h}	10.4^{gh}	32.9
MAM 28	Beige	17.8^{g}	11.3^{fg}	36.5
MAM 42	Yellow	37.4^{a}	22.0^{b}	41.1
Río Grande	Yellow	14.9^{h}	8.4^{jk}	43.7
San Luis	Black	28.5^{b}	15.7^{d}	44.7
1127-DM-2M	Pinto	17.6^{g}	9.5^{hij}	46.1
Feb 174	Brown	19.0^{f}	10.2^{gh}	46.5
POB-2SELL-11	Pinto	26.6^{c}	13.8^{e}	48.8
Puebla 493	Black	22.2^{d}	9.8^{ghi}	55.8
CIAT-326/85	Yellow	22.1^{d}	6.7^{l}	69.7

*Mean of three replicate analyses, expressed in dry weight basis.

$^{a-l}$Means in the same column with different superscripts are significantly different ($P < 0.01$).

in seed coat flour in order to avoid their underestimation in common beans.

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