

## Flatulence-causing galactooligosaccharides of *Phaseolus coccineus* L. and *Phaseolus vulgaris* L.

RYSZARD KOSSON

Department of Processing and Freezing, Research Institute of Vegetable Crops, 22 Lipca 1/3,  
96-00 Skierniewice, Poland

(Received: March 15, 1988. Accepted: May 18, 1988)

### Abstract

*Phaseolus coccineus* beans were assayed for the presence of raffinose and stachyose in whole seeds and in the morphological parts of the seed: cotyledon, hypocotyl and hull. The quantitative composition of the galactooligosaccharides of *Ph. coccineus* was compared with *Ph. vulgaris* seeds. The major galactooligosaccharide of *Ph. coccineus* seeds is stachyose. Another galactooligosaccharide, verbascose, was present in *Ph. coccineus* seeds in very low amounts (0.04%). The highest concentrations of raffinose and stachyose were found in the hypocotyl, 1.11% and 7.48%, respectively. The hull contained the lowest quantities of stachyose and only traces of raffinose.

**Key words:** *Phaseolus coccineus*, *Phaseolus vulgaris*, seed, cotyledon, hypocotyl, hull, galactooligosaccharide content

### INTRODUCTION

The galactooligosaccharides of the raffinose family of sugars (raffinose, stachyose and verbascose) from legume seeds have been known as one of the important contributors to flatus in humans when ingested (Murphy et al. 1972, Cristofaro et al. 1973). This is primarily due to the lack of the hydrolytic enzyme, alpha-galactosidase in the human gastrointestinal tract (Gitzelmann and Auricchio 1965).

The predominance of a particular oligosaccharide seems to depend on the type of legume. For example, verbascose is the major oligosaccharide in the seeds of *Phaseolus mungo*, *Cicer arietinum*, *Cajanus cajan*, *Phaseolus aureus* and *Vicia faba*, whereas stachyose is the major oligosaccharide in *Pisum sativum*, *Phaseolus vulgaris*, *Lens esculenta*, *Glycine max* and *Dolichos sinensis*. Raffinose is present in moderate to low amounts in most legumes. Ajugose is the other higher molecular weight oligosaccharide of the raffinose family of

sugars, which is present in small amounts in *Pisum sativum* and *Lupinus albus* (Reddy et al. 1984).

Most species of the *Phaseolus* genus have already been studied for the presence of those sugars in seeds; for example, *Phaseolus vulgaris* (Iyer et al. 1980), *Phaseolus aureus* (Naivikul and D'Appolonia 1978), *Phaseolus mungo* (Rao and Belavady 1978), *Phaseolus lunatus* (Hardinge et al. 1965), *Phaseolus chrysanthos* (Tanusi et al. 1972). However, no papers were found in the available literature on the sugars of the raffinose family in the seeds of *Phaseolus coccineus* (*Phaseolus multiflorus*).

For this reason, the present paper describes the extraction and quantitative determination of raffinose and stachyose in whole seeds of *Phaseolus coccineus*. The distribution of these sugars in the cotyledon, hypocotyl and hull was estimated as well. The contents of raffinose and stachyose in *Phaseolus coccineus* seeds were compared with *Phaseolus vulgaris* seeds.

## MATERIALS AND METHODS

### PLANT MATERIAL

The seeds of the runner bean cultivar Piękny Jaś (*Ph. coccineus*) and 4 local populations of semi-runner bean (*Ph. coccineus*), originating from SHRO Snowidza, Kasilan, Tyszowce and Kraśnik, were assayed for the presence of raffinose and stachyose. Verbascose was determined in semi-runner bean populations from SHRO Snowidza. Cotyledons, hypocotyls and hulls were isolated from seeds of Piękny Jaś cultivar and an analysis of galactooligosaccharides was performed. Four cultivars and 4 breeding lines of *Phaseolus vulgaris* were selected for raffinose and stachyose analysis in seeds.

### EXTRACTION AND SILYLATION

The air-dry bean seeds were homogenized in a Tecator blender to obtain flour of 0.42-0.25 mm. The galactooligosaccharides were extracted from the beans by refluxing a 10 g sample for half an hour with 100 cm<sup>3</sup> of 80% ethanol. Extracts were cooled, centrifuged and transferred to 100 cm<sup>3</sup> volumetric flasks. A sample of 100-200 mm<sup>3</sup> of supernatant was evaporated to dryness using a stream of air. Sugars were converted to their trimethylsilyl derivatives (TMS) by an injection of 100 mm<sup>3</sup> of silylating mixture containing trimethylsilylimidazole and pyridine according to a previously described method (Kosson 1988).

### GAS LIQUID CHROMATOGRAPHY

A Jeol JGC-1100 gas chromatograph with a flame ionization detector (FID) was used for the separation of the TMS derivatives of raffinose and stachyose. Glass columns (1.5 m × 3 mm) were packed with 3% SE-30 on

60/80 mesh Gas Chrom Q. Gas chromatography operating conditions were: carrier gas — argon; injector temperature 305°C, detector oven temperature 305°C, column temperature 270°C, with programmed increase at 2°C min<sup>-1</sup> to 305°C.

## RESULTS AND DISCUSSION

The results given in Table 1 show that *Phaseolus coccineus* seeds contained 0.29-0.51% raffinose and 2.29-2.50% stachyose. Both runner bean and semi-runner local populations belonging to the same species, *Ph. coccineus*, did not differ significantly in the stachyose contents, although the total galactooligosaccharides content was found to be higher in runner bean.

Table 1

Raffinose and stachyose content in *Phaseolus coccineus* whole seeds (%)

<i>Ph. coccineus</i>	Raffinose (R)	Stachyose (S)	Sum (R+S)
Runner bean cultivar Piękny Jaś	0.51 ± 0.01*	2.50 ± 0.06	3.01
Semi-runner local population from:			
Snowidza SHRO	0.29 ± 0.01	2.42 ± 0.10	2.71
Kasiian	0.33 ± 0.00	2.34 ± 0.21	2.67
Tyszowiec	0.30 ± 0.00	2.29 ± 0.20	2.59
Kraśnik	0.30 ± 0.01	2.50 ± 0.23	2.80

\* Averages from five or three independent replicates ± confidence limits at  $p = 0.95$  evaluated according to Dean and Dixon test.

There are no publications in the available literature concerning the galactooligosaccharides in *Ph. coccineus* seeds; therefore, our results were compared to *Ph. vulgaris* beans (Table 2). The raffinose and stachyose contents in both *Ph. vulgaris* and *Ph. coccineus* species were similar. Differences in the raffinose content between the analysed *Ph. vulgaris* cultivars were nearly double and triple. The results obtained concerning to the raffinose and stachyose content in *Ph. vulgaris* seeds are in agreement with those published in literature (Iyer et al. 1980, Reddy et al. 1984).

According to the data presented in Table 3, raffinose and stachyose are not regularly distributed in all parts of the *Ph. coccineus* seed, the cotyledon, hypocotyl and hull. The highest concentration of raffinose and stachyose was found in the hypocotyl, 1.11% and 7.49 % respectively. The hull contained only traces of raffinose and small quantity of stachyose. Cerning et al. (1975)

Table 2

Raffinose and stachyose contents in *Phaseolus vulgaris* whole seeds (%)

<i>Ph. vulgaris</i>	Raffinose (R)	Stachyose (S)	Sum (R+S)
Cultivar			
Janina	0.20±0.03	1.85±0.26	2.05
Wiejska	0.19±0.01	1.94±0.08	2.13
Atut	0.35±0.01	2.57±0.21	2.92
Biała Wyborowa	0.42±0.03	2.04±0.15	2.46
Breeding line			
IGE 900/82	0.20±0.00	1.94±0.20	2.14
IGE 29/83	0.15±0.01	1.94±0.06	2.09
IGE 1168/83	0.15±0.01	2.16±0.16	2.31
IGE 1446/83	0.45±0.01	1.86±0.34	2.31

investigated oligosaccharides in cotyledons (including germ) and hulls of *Vicia faba* seeds and stated that the hulls contained much less oligosaccharides than cotyledons; only traces of raffinose were present in the hulls. *Vicia faba* and *Ph. coccineus* belong to the same family, *Papilionaceae*.

Table 3

Galactooligosaccharide content in cotyledon, hypocotyl and hull of *Phaseolus coccineus* seeds (%)

Part of seed	Raffinose (R)	Stachyose (S)	Sum (R+S)
Cotyledon	0.40±0.03	3.10±0.33	3.50
Hypocotyl	1.11±0.18	7.49±0.70	8.60
Hull	0.06±0.00	0.37±0.13	0.43

Only 0.04% of verbascose was found in *Ph. coccineus* seeds of semi-runner bean (not shown in tables). A similar level of verbascose — 0.1% — was reported by Iyer et al. (1980) and Reddy et al. (1984) in *Phaseolus vulgaris* seeds.

## REFERENCES

- Cerning J., Saposnik A., Guilbot A., 1975. Carbohydrate composition of horsebeans (*Vicia faba* L.) of different origins. *Cereal Chem.* 52: 125-138.
- Cristofaro E., Mottu F., Wuhrmann J. J., 1973. Study of the effect on flatulence of leguminous seeds oligosaccharides. *Nestle Res. News* 102-104.
- Gitzelmann R., Aurricchio S., 1965. The handling of soy alpha-galactosides by a normal and galactosemic child. *Pediatrics* 36: 231-235.

- Hardinge M. G., Swarner J. B., Crooks H., 1965. Carbohydrates in foods. J. Amer. Diet. Ass. 46: 197-204.
- Iyer V., Salunkhe D. K., Sathe S. K., Rockland L. B., 1980. Quick cooking of beans (*Phaseolus vulgaris* L.): II. Phytates, oligosaccharides antienzymes. Qual. Plant. — Plant Foods Human Nutr. 30: 45-52.
- Kosson R., 1988. Optymalizacja niektórych parametrów analizy galaktocukrów gazotwórczych w fasoli metodą chromatografii gazowej. Bromat. Chem. Toksykol. (w druku).
- Murphy E. L., Horsley H., Burr H. K., 1972. Fractionation of dry bean extracts which increase carbon dioxide egestion in human flatul. J. Agric. Food Chem. 20: 813-817.
- Naivikul O., D'Appolonia B. L., 1978. Comparison of legume and wheat flour carbohydrates: I. Sugar analysis. Cereal Chem. 55: 913-918.
- Rao P. V., Belavady B., 1978. Oligosaccharides in pulses, varietal differences and effect of cooking and germination. J. Agric. Food Chem. 26: 316-319.
- Reddy N. R., Pierson M. D., Sathe S. K., Salunkhe D. K., 1984. Chemical, nutritional and physiological aspects of dry bean carbohydrates. Food Chem. 13: 25-68.
- Tanusi S., Kawamura S., Kasai T., 1972. Methods of determining carbohydrates of mature legume seeds. J. Jap. Food Nutr. 25: 38-41.

### *Galaktooligosacharydy gazotwórcze w nasionach Phaseolus coccineus L. i Phaseolus vulgaris L.*

#### Streszczenie

Oznaczono skład ilościowy galaktooligosacharydów: rafinozy i stachiozy w całych nasionach oraz w liścieniach, hypokotylu i łusce nasion fasoli wielokwiatowej *Phaseolus coccineus*. Dla porównania oznaczono zawartość tych cukrów w nasionach odmian i linii hodowlanych *Phaseolus vulgaris*. Wykazano, że podobnie jak w nasionach *Ph. vulgaris*, głównym galaktooligosacharydem w nasionach *Ph. coccineus* jest stachioza. Inny z galaktocukrów — werbaskoza — jest obecny w nasionach *Ph. coccineus* w bardzo małych ilościach (0.04%). Największe stężenie stachiozy i rafinozy stwierdzono w hypokotylu, odpowiednio 7.48% i 1.11%. Okrywa nasienna fasoli zawiera najmniejsze ilości stachiozy — w porównaniu z innymi częściami nasion — oraz tylko śladowe ilości rafinozy.