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CHANGES IN GLUCOSINOLATES AND PHENOLICS DURING EXTRUSION COOKING OF RAPESEED–LEGUME MIXTURES

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During the extrusion cooking of mixtures of dehulled faba beans or dehulled peas with full-fat rapeseed or extracted rapeseed meal, respectively, native glucosinolates, sinapine, tannins and other phenolics are partially destroyed. The initial water content (12 or 18%) and the maximum extrusion temperature (180 or 200 °C) had no great effect on degradation.

rapeseed; extracted rapeseed meal; faba beans; peas; glucosinolates; sinapines; tannins; extrusion cooking

INTRODUCTION

Both full-fat rapeseed and rapeseed extracted meal are good sources of protein produced in Central Europe (M á t r a i , 1992). They are suitable substitutes for imported soybean, especially in mixture with legumes, such as faba beans, peas or lupins.

The disadvantage of rapeseed or extracted rapeseed meal as raw material is relatively high content of antinutritional or even toxic components: (1) glucosinolates and their degradation products (isothiocyanates, thiocyanates, nitriles, oxazolidinethiones and other substances); (2) phenolics (particularly, sinapines and tannins); (3) lipidic substances (higher fatty acids, such as erucic and eicosenoic fatty acids, which have been, however, almost totally eliminated by selection in modern cultivars, and thio-acids); (4) essential fatty acid decomposing enzymes (lipoxygenases and hydroperoxide lyases); (5) phytates, and other compounds. Antinutritional products are formed as a result of browning reactions during rapeseed processing.

The extrusion cooking is a convenient method for the preparation of feed mixes for broilers. In the extrusion process, the material is heated to high temperatures under high pressure, even when the treatment is relatively short. Some toxic substances may be decomposed during the process, but very little is known on their degradation in rapeseed. Therefore, changes of glucosinolates and phenolics will be discussed as they are the most important substances deteriorating the quality of rapeseed, even in case of low-glucosinolate cultivars.

MATERIAL AND METHODS

The following materials were used for the extrusion cooking (their compositions are given in Tab. I):

(a) Faba beans: Polish variety Nadwiślański, cultivated and harvested in central Poland in 1993, dehulled by mechanical and pneumatical processing;
(b) Peas: Polish variety Kaliski, cultivated and harvested in central Poland in 1993, dehulled by mechanical and pneumatical treatment;
(c) Full-fat rapeseed: The Polish variety Bolko „00" rapeseed, cultivated and harvested in central Poland in 1993;
(d) Rapeseed extracted meal: Obtained from Bolko „00" rape variety, after thermal and moisture adaptation, then processed under conventional combined expeller-press solvent-extraction procedure in an oil processing plant.

I. Composition of raw materials

Analyzed component		Dehulled faba beans	Dehulled peas	Full-fat rapeseed	Rapeseed extracted meal
Dry matter	(%)	87.03	88.26	93.74	90.44
Water	(%)	12.97	11.74	6.36	9.56
Oil	(% d. s.)	0.75	0.82	41.44	1.00
Protein	(N x 6.25; %)	31.1	21.0	20.3	33.2
Crude fibre	(%)	1.4	1.1	6.5	12.5
N-free extract	(%)	50.1	62.7	21.4	36.9
Ash	(%)	3.7	2.7	4.1	6.8
Glucosinolates	(%)	0	0	0.89	0.57
Phenolics	(g/kg)	0.29	0.20	13.41	5.85
Sinapines	(g/kg)	0.00	0.00	7.68	3.37

Mixtures used for the extrusion contained 25.0%, 50.0%, and 75.0% rapeseed or rapeseed extracted meal in mixtures with 75.0%, 50.0%, and 25.0% of dehulled faba beans or dehulled peas, respectively. For the extrusion, the material was ground, and the water content adjusted to either 12.0% (lower water content) or to 18.0% (higher water content) in another series of experiments.

The mixtures were processed in a twin-screw extrusion cooker, type 2S 9/5, produced by Z. M. Ch. Metalchem, Gliwice, Poland. During the processing of rapeseed–legume mixtures, the residence time in the extruder was 50 seconds, the maximum pressure 7 MPa, and the maximum capacity: 200 kg/h. The extrusion parameters differed in the water content (either 12.0% or 18.0 % water) and in the extrusion temperature. The following temperatures were reached in the respective consecutive segments of the extruder: (a) Lower-temperature profile: 100 – 120 – 150 – 180 – 120 °C; (b) Higher-temperature profile: 120 – 140 – 170 – 200 – 140 °C. The samples were then packed into plastic bags, and stored in a refrigerator until the analysis.

The dry matter content was determined by drying at 105 °C, and the water content was calculated from the weight loss. The content of oil was determined after Soxhlet using hexane as a solvent. Crude protein, crude fibre, nitrogen-free extract, and ash were determined after the standard AOCS procedures.

For the determination of glucosinolates 0.5 g of defatted sample were weighed out, and sample was heated to 90 °C for 15 min. The sample was mixed with 10 ml of boiling water, and shaken 1 min. After the addition of the clarifying solution: lead acetate (0.1 mol/l) and barium acetate (0.1 mol/l), the suspension was shaken for a short time, and then heated for 10 min in a boiling water bath (the suspension was shaken twice during the period). The suspension was then cooled to the ambient temperature, and centrifuged at 85 Hz for 5 min. Clear solution were used for the HPLC analysis. The method using the reversed-phase high performance liquid chromatography with linear solvent gradient (Björkqvist, Hase, 1988) was slightly modified. Apparatus: Hewlett Packard 1050, equipped with the autosampler and the integrator HP 3396 Series II; column: 200 mm x 4.6 mm, packed with Hypersil ODS (C18), particle size 0.005 mm, provided with a precolumn (20 mm x 4 mm) packed with the same sorbent (both produced by Hewlett Packard); mobile phases: A = aqueous solution of ammonium acetate (0.1 mol/l), B = acetonitrile; gradient: 0 min: 0% B and 100% A, 9 min: 3% B and 97% A, 25 min: 20% B and 80% A and 30 min: 0 % B and 100% A; flow rate: 2.0 ml/min; injected volume: 0.100 ml, detection at 235 nm. All glucosinolate peaks were integrated, and the sum reported as total glucosinolates.

Vinyloxazolidinethione was determined by UV spectrophotometry (Wetter, 1957).

Total extractable phenolic substances were determined by spectrophotometry in defatted samples (Kolovrat, 1990). About 200 mg of sample was extracted 4 times, each time with 5 ml methanol in an ultrasonic bath for 5 min. The extract was filled up to 25.0 ml, diluted 25 times with methanol, and the content measured at 330 nm with methanol in the reference cell. The content of phenolics is expressed in mg of sinapine thiocyanate in 1 g of the defatted sample. The standard sinapine thiocyanate was isolated from rapeseed extracted meal (Kolovrat, 1990).

The sinapine content was determined using ion-pair high performance liquid chromatography (Henning, 1982) under the following conditions: apparatus Hewlett Packard 1050, equipped with the autosampler and the integrator HP 3396 Series II; column 250 mm x 4 mm, packed with Separon SGX C18, particle size 0.010 mm (Tessek, Prague); column temperature: 50 °C; flow rate: 1 ml/min; mobile phase: acetonitrile : water (400 : 600 v/v), lauryl sulphate (4 mmol/l), potassium dihydrogen phosphate (8 mmol/l), and the mixture was adjusted with 85% phosphoric acid to pH = 3.5; sample: undiluted extract prepared for the spectrophotometric determination of sinapine; injected volume: 0.020 ml; UV/VIS detector HP 1050 at 325 nm; the sinapine content was expressed in mg of sinapine thiocyanate in 1 g of defatted sample.

RESULTS AND DISCUSSION

Water content in the extruded products

Main part of the original water content is lost during extrusion process by evaporation. The residual water content decreases with the increasing rapeseed fraction. The water content was very slightly higher in mixtures with seeds than with extracted meal (Tab. II) and mixtures containing dehulled peas had slightly more water than those with dehulled faba beans.

The extrusion temperature had no significant effect on the content of residual water in extruded products (Tab. III). On the contrary, the water content in the original mixture before extrusion was very important. The average water content in extruded products produced from low-water mixtures was 7.18% while that from high-water mixtures was 9.35%. This difference affects both the texture and the stability on storage.

Oil content in the extruded products

During the extrusion, the content of extractable lipids increased (Tab. IV), in the average, by 4.26% in mixtures with seeds, and by 2.76% in mixtures with extracted meal, respectively. The increase of extractable lipids was caused by release of lipids bound in lipoproteins and cell membranes during

II. Effect of composition on the water content in extruded products (average values of four experiments)

Rapeseed in the mixture	With faba beans % water	With peas % water	Average value
Full-fat seed:			
25%	8.56	8.98	8.77
50%	8.16	8.56	8.36
75%	7.79	8.20	8.00
Extracted meal:			
25%	8.43	8.56	8.50
50%	7.58	8.70	8.14
75%	7.48	8.20	7.82

III. Effect of processing conditions on the residual water content in extruded products

Analyzed mixtures	Processing conditions	With full-fat rapeseed water (%)	With extracted meal water (%)
With faba beans	temperature:		
	lower	8.26	7.66
	higher	8.08	7.99
	water content:		
	lower	7.34	6.60
	higher	9.00	9.06
With peas	temperature:		
	lower	8.44	8.47
	higher	8.72	8.51
	water content:		
	lower	7.57	7.23
	higher	9.59	9.75

the heat treatment in presence of water, when the protein fraction denaturated. The extrusion temperature and the initial water content had no significant effect on the lipid release.

Degradation of glucosinolates during the process

Rapeseed glucosinolates are partially decomposed under catalytic action of myrosinases (thioglucoside glucosidases) even during the storage of seeds

IV. Changes of oil extract during the extrusion of rapeseed—legume mixtures (average values of four experiments)

Analyzed mixture	Oil (%) before the extrusion	Oil (%) after the extrusion	Difference (%) due to extrusion
25% R + 75% F	10.84	15.56	4.72
50% R + 50% F	21.04	25.34	4.30
75% R + 25% F	31.24	34.74	3.50
25% R + 75% P	10.90	15.32	4.42
50% R + 50% P	21.08	25.29	4.21
75% R + 25% P	31.26	35.64	4.38
25% M + 75% F	0.71	2.79	2.08
50% M + 50% F	0.78	3.06	2.28
75% M + 25% F	0.85	6.08	5.23
25% M + 75% P	0.77	3.74	2.97
50% M + 50% P	0.82	2.67	1.85
75% M + 25% P	0.87	2.93	2.06

Notes: R = full-fat rapeseed, F = dehulled faba beans, P = dehulled peas, M = extracted rapeseed meal

(James, Rossiter, 1991), especially, if they have been damaged and the enzyme got into contact with glucosinolates. However, the hydrolysis is extremely slow at the natural water content of 12% or lower. It may become much faster at the water content of 18% used in the extrusion experiments, particularly, if rapeseeds have been ground.

A set of experiments was carried out at 50% water and at ambient temperature. Only 1.05% of the original rapeseeds glucosinolates were decomposed during 3 hours. The degree of glucosinolate decomposition using the incubation time of 20 h (overnight storage) is shown in Tab. V. Losses were neither high, nor negligible. They occurred in fresh seeds, but to lesser extent, even in extracted meals and in the products after extrusion, where active myrosinases may be hardly expected. It seems that some non-enzymic decomposition could proceed as well (MacLeod, Rossiter, 1986).

The formation of vinyloxazolidinethione could not be detected, except in the experiment with the incubation of ground rapeseed (not in case of extracted meal) with 18% water and 20-h incubation, but progoitrin could be decomposed after another mechanism than into vinyloxazolidinethione under conditions of extrusion cooking (Gronowitz et al., 1978)

Changes in the glucosinolate content during the extrusion are shown in Tab. VI. In mixtures with full-fat rapeseed, they varied between 14–36% of

V. Losses of glucosinolates during the 20-h incubation at ambient temperature (50% water)

Incubated material	Original content before the incubation (%)	Losses of the original content (%)
Rapeseed	100.00	9.94
Extracted meal	63.57	7.13
Extruded products:		
50% seed + 50% beans	32.89	5.77
75% seed + 25% beans	50.72	4.74
50% seed + 50% peas	42.63	3.23
50% meal + 50% beans	28.29	3.15
75% meal + 25% beans	43.49	4.08
25% meal + 75% peas	15.58	4.42
75% meal + 25% peas	45.99	5.56

VI. Changes of glucosinolates during the extrusion of rapeseed–legume mixtures (average values of four experiments)

Analyzed mixture	GLS (%) before the extrusion	GLS (%) after the extrusion	Difference (%) due to the extrusion
25% R + 75% F	25.00	19.10	5.90
50% R + 50% F	50.00	38.03	11.97
75% R + 25% F	75.00	46.82	28.18
25% R + 75% P	25.00	16.23	8.77
50% R + 50% P	50.00	39.83	10.17
75% R + 25% P	75.00	64.35	10.65
25% M + 75% F	15.89	15.14	0.75
50% M + 50% F	31.78	30.56	1.22
75% M + 25% F	47.67	44.20	3.45
25% M + 75% P	15.89	17.18	–1.29
50% M + 50% P	31.78	33.02	–1.24
75% M + 25% P	47.67	46.56	1.11

Notes: GLS = glucosinolates, R = full-fat rapeseed, F = dehulled faba beans, M = rapeseed extracted meal, P = dehulled peas

the original amount (25.21% in the average), but they were negligible in mixtures with extracted meal. Obviously, short time of extrusion processing does not affect the glucosinolate content like longer exposition to lower temperature (Velíšek et al., 1991).

VII. Effect of the conditions of extrusion cooking on the content of glucosinolates, sinapines and tannins in extruded rapeseed-legume mixtures (average values of six experiments)

Substances studied	Factor studied	Full-fat seed	Extracted meal
Glucosinolates (% of the content in seeds)	temperature:		
	lower	38.74	30.93
	higher	39.46	31.28
	water content:		
	lower	38.03	31.52
	higher	40.10	30.69
Phenolics (g/kg)	temperature:		
	lower	6.18	3.68
	higher	5.63	3.86
	water content:		
	lower	6.39	3.81
	higher	5.51	3.73
Sinapines (g/kg)	temperature:		
	lower	3.03	2.46
	higher	2.76	2.51
	water content:		
	lower	3.12	2.51
	higher	2.67	2.46

The temperature profile during the extrusion had no effect on degradation of glucosinolates. The effects of the initial water content were neither systematic, nor substantial (Tab. VII).

Changes in sinapine and other phenolics during extrusion

Among rapeseed phenolics, sinapine is the most important group of substances (Shahidi, Naczk, 1992). Sinapine (choline esters of sinapic acid and related phenolic acids) imparts both seeds and extracted meal an objectionable bitter and astringent taste (Ismail et al., 1981). Tannins are objectionable because they decrease the digestibility of proteins (Clandinin, Heard, 1968).

Changes in sinapine during the extrusion cooking are shown in Tab. VIII. The original sinapine content was relatively low compared with the literature (Blair, Reichert, 1984), but still within the scope as reported in the literature (Clausen et al., 1985). In mixtures with full-fat rapeseed, losses

VIII. Changes in the content of sinapines during the extrusion cooking of rapeseed-legume mixtures (averages of four experiments)

Analyzed mixture	Sinapines (g/kg) before extrusion	Sinapines (g/kg) after extrusion	Difference (g/kg) due to extrusion
25% R + 75% F	1.92	0.91	–1.01
50% R + 50% F	3.84	2.56	–1.28
75% R + 25% F	5.76	4.43	–1.33
25% R + 75% P	1.92	1.25	–0.67
50% R + 50% P	3.84	3.03	–0.81
75% R + 25% P	5.76	5.18	–0.58
25% M + 75% F	0.84	0.72	–0.12
50% M + 50% F	1.68	2.19	+0.51
75% M + 25% F	2.52	4.46	+1.94
25% M + 75% P	0.84	1.09	+0.25
50% M + 50% P	1.68	2.46	+0.78
75% M + 25% P	2.52	3.99	+1.47

Notes: R = full-fat rapeseed, F = dehulled faba beans, P = dehulled peas, M = rapeseed extracted meal

of sinapine are substantial, especially in mixtures containing excess legumes and in mixtures containing faba beans. In mixtures with extracted meal, the apparent content of sinapine rather increases. It is, however, less probably due to release of sinapines from insoluble forms than to formation of some interfering substances.

Higher extrusion temperature increased slightly losses of sinapine in mixtures containing full-fat rapeseed. Higher initial water content had a similar effect (Tab. VII).

Changes in total phenolics (Tab. IX) depend again on the composition of extruded mixtures. Moderate decrease was observed in mixtures with rapeseed, especially in presence of faba beans. Two opposed processes obviously proceed in mixtures with extracted meal; the phenolics content is decreased by the heat treatment, but some phenolics or some substances interfering with the determination are produced.

In mixtures with seeds, lower phenolics losses were obtained at higher extrusion temperatures or at higher initial water contents.

The difference between total phenolics and sinapines may be considered as free phenolic acids, their esters, and tannins (Leung et al., 1979). The content changes were irregular during the extrusion. On an average, it de-

IX. Changes in the content of total phenolics during the extrusion cooking of rapeseed-legume mixtures (averages of four experiments)

Analyzed mixture	Phenolics (g/kg) before extrusion	Phenolics (g/kg) after extrusion	Difference (g/kg) due to extrusion
25% R + 75% F	3.57	2.15	−1.42
50% R + 50% F	6.85	5.40	−1.45
75% R + 25% F	10.13	9.28	−0.85
25% R + 75% P	3.50	3.11	−0.39
50% R + 50% P	6.80	6.19	−0.61
75% R + 25% P	10.10	9.56	−0.54
25% M + 75% F	1.69	0.72	−0.97
50% M + 50% F	3.13	3.08	−0.05
75% M + 25% F	4.55	6.28	+1.73
25% M + 75% P	1.64	1.44	−0.20
50% M + 50% P	3.08	3.12	+0.04
75% M + 25% P	4.52	7.02	+2.50

Notes: R = full-fat rapeseed, F = dehulled faba beans, P = dehulled peas, M = rapeseed extracted meal

creased by 0.36 g/kg in case of mixtures with seeds, and by 0.29 g/kg in case of mixtures with extracted meal. The difference observed in the content of sinapine did not occur in the case of tannins.

CONCLUSIONS

The extrusion cooking decreased moderately the content of glucosinolates and sinapines, which is advantageous for the nutritional value of extruded feed mixes. The extrusion should not be considered, however, as a procedure for rapeseed detoxication. It is in agreement with the opinion, that the biological value of rapeseed is better improved by breeding than by any detoxication processing (B i l l e et al., 1983). Nevertheless, recent investigations with animal feed application of faba bean–rapeseed extrudate (M o ś c i c k i et al., 1994), generally, showed positive influence of extrusion cooking on the nutritional value of faba bean–rapeseed concentrate used in the chicken diet. It means that the extrusion process can be taken into account during selection of thermal treatment for feed components.

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Změny glukosinolátů a fenolických látek během zpracování směsí řepkových semen s luskovinami extruzní technologií.

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Řepková semena (nebo řepkový extrahovaný šrot) byla smísena s odslupkovanými semeny bobu obecného nebo odslupkovaným hrachem v hmotnostních poměrech 3 : 1, 1 : 1 a 1 : 3 a obsah vody ve směsi byl nastaven na hodnotu 12 % nebo 18 %. Směsi byly extrudovány na dvoušnekovém extrudéru Metalchem 25 9/5 při maximálních teplotách 180 a 200 °C. Změny obsahu glukosinolátů a fenolických látek byly stanoveny kapalinovou chromatografií s vysokou účinností. Obsah vody v extrudovaných výrobcích nezáležel na maximální teplotě, ale byl ovlivněn původním obsahem vody. Během záhřevu se uvolnila část lipidů vázaná v lipoproteinech. Glukosinoláty se při extruzi rozložily v rozsahu kolem 25 % původního obsahu; vliv podmínek extruze byl nepodstatný. Z fenolických látek byly nejvíce zastoupeny sinapiny, jejichž obsah byl relativně nízký. Ve směsích obsahujících plnotučnou řepku byly ztráty sinapinu při extruzi podstatné, zvláště při vyšších teplotách extruzního procesu. Ve směsích s extrahovaným řepkovým šrotem se při záhřevu tvořily barevné produkty, rušící při stanovení. Celkem se ztráty glukosinolátů a sinapinů ukázaly menší než při toastingu extrahovaných šrotů.

řepková semena; řepkový extrahovaný šrot; bob obecný; hrách; glukosinoláty; sinapiny; taniny; extruzní technologie

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