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Undesirable Sensory Properties of the Dried Pea (*Pisum sativum*). The Rôle of Saponins

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ABSTRACT

The rôle of saponins in the undesirable sensory properties of the dried pea P. sativum *is reported. The sensory properties of isolated soyasaponin I are defined and described as bitter, astringent and metallic. The distribution of saponin in various air-classified pea flour fractions shows that the protein-rich fraction may contain sufficient saponin to cause undesirable tastes.*

INTRODUCTION

Vegetable protein is an increasingly important component of the human diet; however, its effective utilization in human foods depends, to a large degree, upon consumer acceptance. Good sensory characteristics are an essential requirement for such acceptance (Pattee *et al.*, 1982). For this reason, the flavour problems associated with soya products have been the subject of considerable research (Cowan *et al.*, 1973; Sessa & Rackis, 1976; Rackis *et al.*, 1979; Roozen, 1979; Huang *et al.*, 1981; Adler-Nissen & Olsen, 1982) with the undesirable taste characteristics being variously described as 'beany', 'green', 'nutty', 'astringent' and 'bitter'. The latter two terms, together with 'metallic', have also been shown in this work to describe the 'off-taste' sometimes present in the flour produced from milled, whole pea seed.

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Recent studies have shown the pea to have great potential as a source of plant protein for the UK and efforts have been made to increase the crop yield, to optimise the conditions for protein isolation and to identify and exploit other by-products of this processing, e.g. starch and fibre. The present work, part of such general studies being undertaken by the Agricultural and Food Research Council, has involved the chemical and organoleptic investigation of pea flour and its fractions with a view to isolating and identifying the natural components of pea seed responsible for such flavour defects.

EXPERIMENTAL

Materials

Pea samples were obtained from the John Innes Institute, Norwich. Soybean flour was purchased from BDH Chemicals Ltd, Poole, Malvern water was purchased from Cadbury-Schweppes, Reading. Methanol, quinine sulphate and alum were of Analar grade, the former being redistilled before use. The gel permeation study was conducted using Sephadex G-50 (Pharmacia Fine Chemicals, Milton Keynes). Soyasaponin I was isolated from soybean flour using flash chromatography, as described elsewhere (Price & Fenwick, 1984).

METHODS

Sensory

All samples were dissolved or suspended in Malvern water. Assessments on the pea flour were made on a 10 % slurry; the samples from the first stages of the fractionation were evaluated at the concentration present in the original pea flour, but slightly higher concentrations were tested in the latter stage of fractionation due to the removal of some components, shown to be bitter, in the early stages. The purified saponin was assessed at a range of concentrations, considered to be typical of that present in peas—0·03, 0·04, 0·06, 0·07, 0·09 and 0·1 %. Evaluations were performed on 5-ml samples which were swirled in the mouth for 10 s and expectorated. Assessments continued for 10 min after the introduction of the sample into the mouth.

Evaluations were carried out using two panels of assessors:

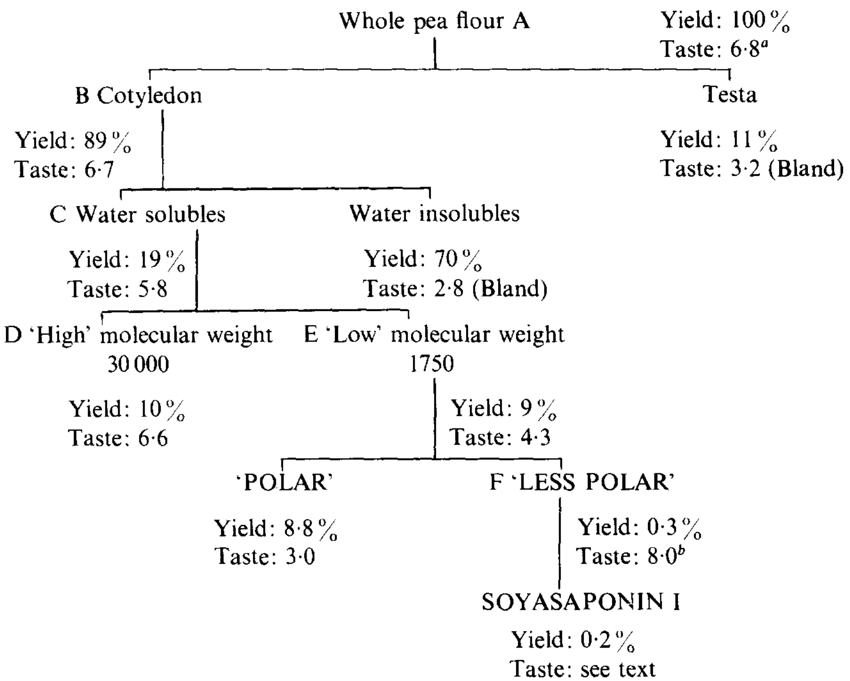
(a) Hedonic ratings were obtained on cv. 'Filby' by sixty assessors; preliminary work had suggested that this cultivar was bitter and astringent. Assessors rated the samples, 1 and 5 min after tasting, on a 9-point scale from 'like extremely' through 'neither like nor dislike' to 'dislike extremely'. They also described the sample, using their own words, and rated the intensity of the stimulus at 0·5, 1, 3, 5 and 10 min after tasting (rating scale: 0, absent; 2, very weak; 4, weak; 6, moderate; 8 strong; 10, very strong).
(b) Nine of these assessors were 'trained' by familiarising these with the main terms used to describe a number of pea samples; namely, 'pea', 'bitter' (standard, 0·004% quinine sulphate), 'astringent' (standard, 0·05% aluminium potassium sulphate, alum) and 'metallic' (standard, a clean copper coin). The intensities of these characteristics were assessed at time 0 and at each of five further times (see (a) above). Each sample was evaluated by at least six of the 'trained' assessors and the results for each characteristic are expressed as the mean panel rating of the maximum score given by each person. In addition, as assessors differed in their perception of which characteristic was most intense in a sample, the maximum undesirable quality is quoted (i.e. the panel mean for the highest rating given by each assessor, independent of characteristic).

Because quantities of material were generally limited, the pea fractions were usually only assessed on one occasion. When duplicate samples were evaluated, however, they were found to differ by less than 0·8 units on the 10-point scale. All saponin samples were assessed in duplicate; the assessments differed by less than 0·3 units.

Chemical

(i) Fractionation of pea flour
The procedure adopted is outlined in Fig. 1. Since each stage of the fractionation was monitored by sensory assessment, there were severe constraints imposed upon the techniques and reagents which could be employed.

Whole pea flour, fraction A in the Figure (100 g) was extracted three times with distilled water (3 × 100 ml) using an Ultra-turrax homogeniser for 0·5 h with cooling so that the mixture did not exceed 15 °C. After



[a] Taste: Overall undesirable score—2 = very weak; 4 = weak; 6 = moderate; 8 = strong.
[b] Concentrated fraction tasted.

Fig. 1. Fractionation procedure for the isolation of undesirable taste components in pea flour.

centrifugation at 18 000 rpm (5 °C) for 0·5 h the supernatant was freeze-dried and the residue vacuum-dried. This water-soluble portion (fraction C) was subjected to gel permeation on Sephadex G50 using water as eluent. Two fractions—one containing components with a molecular weight in excess of 30 000 (fraction D) and the other containing components with a molecular weight of less than 1750 (fraction E)—were each collected and freeze-dried. Reversed-phase chromatography and subsequent normal phase chromatography on the lower molecular weight fraction effected the isolation of soyasaponin I.

Air classification of the pea flour was carried out using an Alpine Zigzag Classifier, model A100 MZR.

(*ii*) *Chemical analysis*

The saponin content of pea flour and its fractions was determined by thin layer and gas chromatographic procedures (Curl *et al.*, 1985).

RESULTS AND DISCUSSION

(i) Sensory studies on pea flour

The descriptions of the tastes perceived by sixty assessors in panel (a) are shown in Table 1, whilst the distributions of the frequency of the hedonic response to the whole flour of cv. 'Filby' seed, determined after 1 and 5 min, are given in Table 2. Seventy-three per cent of the assessors described the taste as bitter, astringent or metallic. The sample was disliked by 63% and 55% of the panel 1 and 5 min, respectively, after tasting. The time/sensory response curves for the different taste characteristics rated by panel (b) (Fig. 2) show that the pea flavour was detected immediately, the bitter and metallic characteristics were

TABLE 1
Qualitative Assessment of Undesirable Taste in Pea Flour

Description of taste	*Per cent of assessors using description*[a]
Pea	100
Bitter	73
Astringent	70
Metallic	12
Others (16)	32

[a] Total number of assessors, 60.

TABLE 2
Hedonic Response to Flavour of Pea Slurry (cv. 'Filby')

Hedonic rating	*Frequency of response*[a]	
	t = 1 min	*t = 5 min*
Like extremely/very much	2	2
Like moderately	5	1
Like slightly	7	2
Neither like/dislike	8	14
Dislike slightly	11	20
Dislike moderately	24	11
Dislike very much	3	2

[a] Total number of assessors, 60.

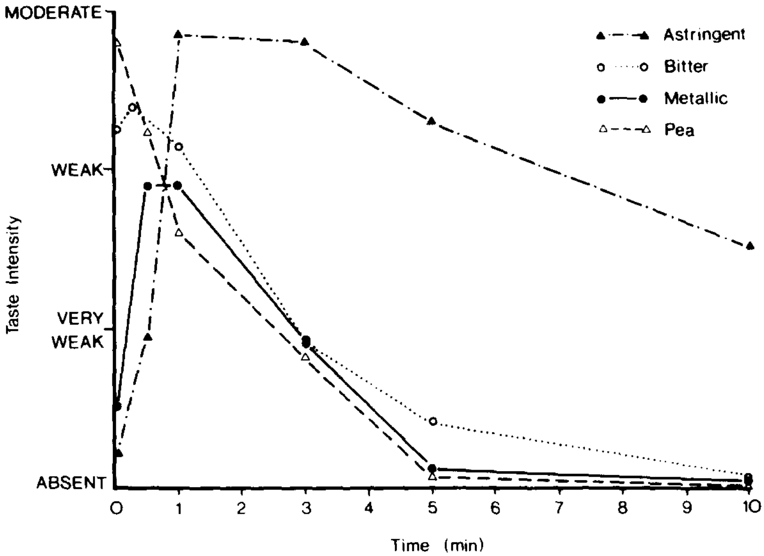


Fig. 2. Plot of time and taste intensity for pea flour.

maximal after 30 s and the astringency was slower to develop, reaching a maximum between 1 and 3 min after tasting and thereafter decreasing slowly. It is therefore likely that this astringent characteristic is the major contributor to the unpleasant taste noted after 5 min (Table 2 and Fig. 2). To examine the variability of the important taste characteristics in dried pea, flours were obtained from thirty-six varieties (supplied from the breeding programme of the John Innes Institute) and were assessed by the trained panel for bitter, astringent and metallic notes. The results were: bitterness (mean, 4·1; maximum, 7·3), astringency (mean, 3·9; maximum, 6·5), metallic (mean, 1·9; maximum, 4·0), maximum undesirable note (mean, 5·6; maximum, 7·3).

It has been reported in the literature that tannins are responsible for astringency in pasture and other legumes. As part of the studies being conducted elsewhere, the tannin contents of the thirty-six pea samples reported above were determined. However, no relationship was observed between these tannin contents (determined using three methods, Stickland, 1984) and the above sensory characteristics. It should be mentioned that tannins are a diverse family of polyphenolic compounds and this finding would not necessarily exclude a correlation between a specific phenolic compound and the results of the taste panel. Griffiths (1981) has reported a correlation between tannin content in pea cultivars and flower colour; in particular, those containing low levels of tannins possess white flowers. In order to reduce the possibility of tannins contributing to the undesirable flavours of pea, a detailed fractionation study was initiated using flour from the white flowering cv. 'Filby' which had been found to possess bitterness and astringency.

(ii) Fractionation studies on pea flour

The fractionation study was conducted as shown in Fig. 1. The whole pea flour obtained from cv. 'Filby' (A) possessed a maximum undesirable rating of 6·8 (Fig. 1). The undesirable components were found in the cotyledon fraction (B) and were readily extracted by water (C). The latter fraction, corresponding to 19 % of the original flour, was divided into two fractions by gel permeation chromatography on Sephadex G50. Both high and low molecular weight fractions (D and E, respectively) possessed the undesirable sensory characteristics of the original pea flour. Preliminary investigation of the latter fraction revealed that the bitterness and astringency were associated with what appeared to be a single component. Fraction E was thus subjected to reversed phase flash chromatography and a cut was isolated (F, corresponding to 0·3 % of the original flour), which possessed astringency and bitterness and was homogeneous, by thin layer chromatography. Subsequent isolation, purification and chemical investigations revealed this component to be soyasaponin I (Price & Fenwick, 1984).

It should be pointed out that, under certain adverse circumstances, other low molecular weight components may also contribute to the above undesirable flavours. A bitter taste was found to be associated with a phospholipid-rich fraction obtained from pea flour stored for 1 year at room temperature, but no such 'off-taste' was apparent in a similar fraction obtained from the freshly milled sample. It is assumed that the observed bitterness originates from lipid oxidation, in a manner similar to that reported by Sessa & Rackis (1976) for soya flour. The possibility that ethyl-α-D-galactopyranoside, formed during extraction if ethanol is used, could contribute to the observed bitterness of the pea fractions studied here (Honig *et al.*, 1971) was excluded by the use of methanol. Whilst it is possible that the analogous methyl α-D-galactopyranoside may be formed under these circumstances, this does not possess the undesirable flavour characteristics of the higher homologue (K. R. Price, unpublished).

Saponins, like tannins, are generally referred to as being bitter

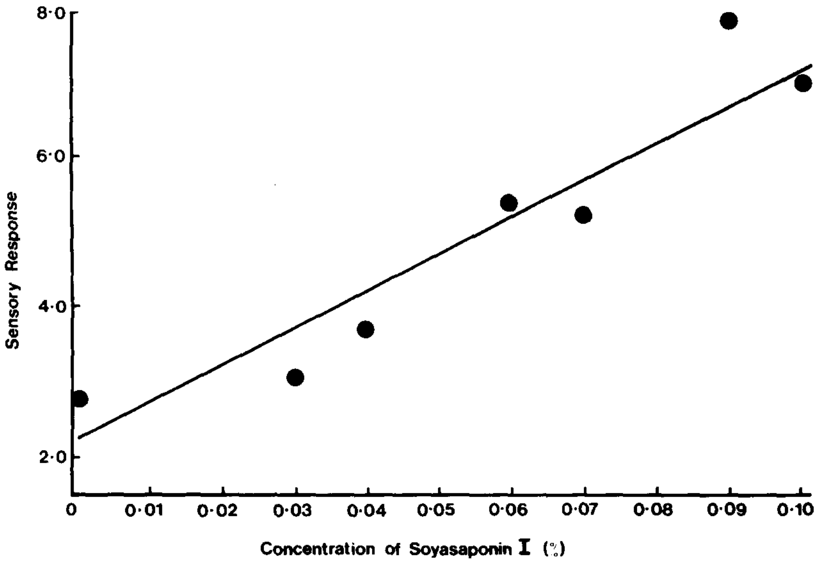


Fig. 3. Plot of concentration of soyasaponin I and sensory response.

(Daubert, 1950), although there have been few detailed sensory studies conducted. It was of particular interest in the present study to obtain organoleptic data on the purified pea saponin and to relate these to its effect in the pea flour. Accordingly, soyasaponin I was isolated from both pea and soya flours in sufficient quantities to facilitate such study. The relationship between the concentration of soyasaponin I and sensory response is illustrated in Fig. 3 and shows the effect of testing the purified glycoside at concentrations up to 0·1 % in Malvern water. A linear fit on the data gave a slope of 4·963 with a standard error of 0·783 and a *T* value of 6·34 being significant at the 1 % level; the correlation coefficient was 0·931. The concentration of soyasaponin I at which the undesirable bitter and astringent tastes were rated as moderate (score 6·0) by the panel was approximately 0·08 %. If the sole cause of bitterness and astringency in pea flour were to be soyasaponin I then the corresponding concentration in the flour would need to be 0·8 %. The possibility that other factors in pea flour might modify (enhance or reduce) the intensity of the sensory characteristics of the saponin observed in aqueous solution was examined. However, when the assessors were presented with samples of pea flour containing additional purified soyasaponin I, no such effects were observed.

Inspection of the literature initially suggests that such levels may be achieved, as Fenwick & Oakenfull (1983) have reported a saponin content of 1·1% (dry weight) in green, immature peas and 4·3% (dry weight) in soybean flour. There is considerable evidence to indicate that soyasaponin I is the major saponin in soyabean flour and if this is the case, then the sensory data presented here would suggest that the flours examined by Oakenfull and Fenwick would be organoleptically unacceptable. In order to examine this in more detail, the method of analysis used by the Australian workers was improved by the addition of a reversed phase chromatography step and with the use of pure soyasaponin I as standard (Curl *et al.*, 1985). The saponin content of the original pea flour was observed to be of the order of 0·2%—considerably less than that reported earlier, but in agreement with the yields of the compound chemically isolated. Using this method (tlc), a saponin content of 0·46% was measured for commercial soya flour; once again, much lower than that reported by Oakenfull and Fenwick but more in agreement with levels found recently in several varieties of soya by Kitagawa *et al.* (1984*a*) using gas chromatography (0·22–0·32%) and by Kitigawa *et al.* (1984*b*) using high performance liquid chromatography (0·20–0·26%).

Despite the fact that the saponin content of the whole flour of cv. 'Filby' was much less than that considered necessary for undesirable sensory characteristics, processing of the whole flour was found to have a significant effect on saponin content. When the various air-classified

TABLE 3
Effect of Saponin Levels on the Sensory Properties of Some Air-Classified Fractions Obtained from Pea Flour[a]

Fraction	*Distribution of saponins*[c]	*Bitter*	*Sensory ratings* *Astringent*	*Metallic*	*MUR*[d]
Protein-rich (16·2)[b]	61·9	5·8	6·0	2·6	6·7
Starch-rich (72·9)[b]	33·3	4·0	5·4	2·0	5·8
Hull-rich (10·9)[b]	4·8	1·7	3·2	2·0	4·0
Whole flour (100)[b]	100	6·8	6·5	3·0	6·8

[a] cv. 'Filby'.
[b] Per cent of whole flour.
[c] Measured by thin layer chromatography (Curl *et al.*, 1984) as per cent of total in each fraction.
[d] Maximum undesirable rating (see 'Experimental' section).

fractions were examined (Table 3), the saponin content of the protein-, starch- and hull-enriched material was found to be distributed in the ratio of 13:7:1, respectively, with the protein-rich fraction exhibiting an approximately fourfold increase in saponin content over that present in the original flour. The starch- and hull-enriched fractions showed corresponding reductions of a half and a third, respectively. This distribution of saponin content on processing is in agreement with the general findings of Elkowicz & Sosulski (1982), although, because of the limitations of the haemolytic assay for saponin content used, these workers were unable to detect this effect in pea and soya.

Thus, the level of saponin present in the protein-rich fraction of cv. 'Filby' is such as to adversely affect its sensory characteristics. From the data presented here it would be expected that a whole pea (or soya) flour possessing a saponin content in excess of 0·2% would, on air classification, yield a protein-rich fraction possessing undesirable sensory characteristics. On the other hand, traditional wet methods for protein concentration would seem to remove most of the saponins.

The saponin content of pea flour possessing a strong bitterness rating (7.8) was double that of one possessing a weaker bitterness rating (4.3) whilst the saponin content of a commercially debittered pea flour (bitterness rating, 3·0) was half that of the original pea isolate (bitterness rating, 6·8)—a likely consequence of the steaming process.

It has been emphasised above that the undesirable sensory characteristics of certain samples of pea flour are due to a variety of factors, of which saponins are but one. Proper processing and storage would reduce the possibility of off-flavours due to artefact formation and lipid oxidation. From the data presented here, however, it is clear that a significant part of the overall taste defects of certain pea flours is due to the presence of bitter, astringent and metallic-tasting components with a fraction (D, Fig. 1) possessing a high molecular weight. Present and future work is directed towards the isolation and identification of these interesting components.

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REFERENCES

Adler-Nissen, J. & Olsen, H. S. (1982). Taste and taste evaluation of soy protein hydrolyzates. *Chem. Fds Beverages Recent Dev.* **149**.

Cowan, J. C., Rackiss, J. J. & Wolf, W. J. (1973). *J. Am. Oil Chemists Soc.*, **50**, 426A–44A.

Curl, C. L., Price, K. R. & Fenwick, G. R. (1985). *Food Chemistry* (In prep).

Daubert, B. F. (1950). *Soybeans and soybean products.* Vol. 1. (Markley, K. S. (Ed.)), Interscience Publishers Inc., NY., 374–5.

Elkowicz, K. & Sosulski, F. W. (1982). Anti-nutritive factors in eleven legumes and their air-classified protein and starch fractions. *J. Food Sci.*, **47**, 1301–4.

Fenwick, D. E. & Oakenfull, D. (1983). Saponin content of food plants and some prepared foods. *J. Sci. Food Agric.*, **34**, 186–91.

Griffiths, D. W. (1981). The polyphenolic content and enzyme inhibitory activity of testas from bean and pea varieties. *J. Sci. Food Agric.*, **32**, 797–804.

Honig, D. H., Rackiss, J. J. & Sessa, D. J. (1971). Isolation of ethyl α-D-galactopyranoside and pinitol from hexane–ethanol extracted soybean flakes. *J. Agr. Food Chem.*, **19**, 543–6.

Huang, A-S., Hsieh, A-L. & Chang, S. S. (1981). Characterisation of the non-volatile minor constituents responsible for the objectionable taste of defatted soybean flour. *J. Food Sci.*, **47**, 19–23.

Kitagawa, I., Yoshikawa, M., Hayashi, T. & Tanayama, T. (1984*a*). Characterisation of saponin constituents in soybeans of various origins and quantitative analysis of soyasaponins by gas–liquid chromatography, *Yak Zasshi.*, **104**, 162–8.

Kitagawa, I., Yoshikawa, M., Hayashi, T. & Tanayama, T. (1984*b*). Quantitative determination of soyasaponins of various origins and soybean products by means of high performance liquid chromatography. *Yak. Zasshi.*, **104**, 275–9.

Pattee, H. E., Salunkhe, D. K., Sathe, S. K. & Reddy, N. R. (1982). Legume lipids. *CRC Crit. Reviews in Food Sci. and Nutr.*, **17**, 97–139.

Price, K. R. & Fenwick, G. R. (1984). Soyasaponin I. A compound possessing undesirable taste characteristics isolated from the dried pea. *J. Sci. Fd Agr.*, **35**, 887–93.

Rackiss, J. J., Sessa, D. J. & Honig, D. H. (1979). Flavor problems of vegetable food proteins. *J. Am. Oil Chemists Soc.*, **56**, 262–71.

Roozen, J. P. (1979). The bitterness of protein hydrolysates. *Progress in flavour research.* Chapter 28, 321–6.

Sessa, D. J. & Rackiss, J. J. (1976). Lipid-derived flavours of legumes. *J. Am. Oil Chemists Soc.*, **53**, 145A.

Stickland, R. G. (1984). Condensed tannins of pea seeds. *Plant Sci. Lett.*, **34**, 403–410.