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Study of Agricultural By-products. Extractability and Amino Acid Composition of Grape (*Vitis vinifera*) Skin Proteins from cv Palomino

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Extractability in various solvents of grape skin proteins from cv Palomino, the most abundant and characteristic grape in the sherry zone, was studied. The greatest extractability was obtained in NaOH solutions.

Amino acid analysis of the various protein extracts showed that the most abundant amino acid was glutamic acid and the limiting ones were tryptophan and the sulphur-containing amino acids, methionine and cystine.

Grape skin represents about 3–5% of the whole weight of pomace, and the protein content of skin is about 13–15% (Massanet *et al* 1987). Grape skin also contains about 4% lipids, 13% available sugars, 40% dietary fibre and about 13% polyphenols (Galán *et al* 1986; Igartuburu *et al* 1987). This content of polyphenols is much lower than that in the seeds and could indicate a better quality of skin proteins for their use in food and feedstuffs. There are no references in the literature that apply to the protein composition of grape skin, information essential for the evaluation of its potential use for human or animal consumption. The present paper describes a study of the extractability of the grape skin proteins of cv Palomino, the most abundant and characteristic grape in the sherry zone, and of their amino acid composition, using the procedures already described (Igartuburu *et al* 1991).

The results of the study of protein extractability are reported in Table 1 and

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show that the most efficient extracting solution was aqueous NaOH, as was so with the proteins from grapeseed cv Palomino (Igartuburu *et al* 1991).

Extractability in NaOH solution when compared with the other solutions used was much superior and the difference was greater than that observed for grapeseed proteins. De-ionised water, shown to be an excellent extracting agent for the proteins of the seeds, was inferior to $MgCl_2$ solutions for skin proteins.

TABLE 1
Influence of the main variables[a] on protein extractability (g kg^{-1} skin[b])

	Extraction with:			
	NaOH	*NaCl*	*$MgCl_2$*	*H_2O*
Concentration (M)				
0·01	114·6	9·6	21·5	
0·05	138·5	9·4	23·4	
0·10	141·9	9·1	14·0	
0·25	98·6	13·2	10·0	
0·50	84·4	7·0	9·4	
0·75	64·1	6·2	8·8	
1·00	62·1	4·7	8·3	
Temperature (°C)				
15	138·5	13·2	23·4	17·2
50	141·0	32·2	25·4	31·2
75	140·7	39·0	26·0	35·6
Time (min)				
5	69·6	4·0	21·5	16·5
15	84·4	7·5	23·3	16·9
30	138·5	13·2	23·4	17·2
60	138·0	23·7	23·5	22·5
Solid/liquid ratio				
1:100	42·7	7·1	7·1	9·3
1:250	81·5	10·1	10·4	10·2
1:500	118·1	17·1	14·7	13·0
1:1000	138·5	13·2	23·4	17·2
1:2000	144·8	14·8	39·4	30·0
pH				
1		19·1	20·2	
3		20·2	19·5	
5		20·0	20·7	
7		26·3	24·2	
9		24·2	09·0	
11		33·0	ND	

[a] Experimental conditions: solid/liquid ratio 1:1000 (0·5 g of milled dry skin and 500 ml of solvent), solute concentration 0·05 M, stirred for 30 min at 15°C, except for those assays where the influence of a specific variable was studied.
[b] Total protein: 147·4 g kg^{-1} skin.

TABLE 2
Amino acids (% total amino acids) from grape skin proteins, cv Palomino

	Extracting solvent					*Mean value*
	A	*B*	*C*	*D*	*E*	
Asp	10·47	11·18	11·73	6·37	7·90	9·53
Glu	9·97	13·65	13·66	12·02	11·39	12·14
Ser	4·96	4·87	5·09	5·72	4·86	5·10
Gly	6·35	6·46	6·04	6·14	6·26	6·25
His	2·63	3·42	2·71	3·05	2·67	2·90
Arg	5·52	5·22	5·13	6·22	5·26	5·47
Thr	5·76	5·46	5·07	5·36	4·85	5·25
Ala	7·64	6·17	6·29	7·36	6·64	6·82
Pro	3·61	5·01	3·89	3·78	3·60	3·98
Tyr	6·33	5·61	5·96	6·63	5·72	6·05
Val	8·35	6·99	6·91	8·03	7·95	7·65
Met	tr	tr	1·16	tr	0·56	—
Cys	tr	tr	tr	tr	tr	tr
Ile	5·87	5·51	5·32	5·80	6·69	5·84
Leu	9·68	9·10	9·13	10·47	10·50	9·78
Phe	5·98	5·59	5·59	5·27	5·79	5·64
Lys	6·54	4·52	5·50	6·87	7·90	6·27
Trp	0·32	0·80	0·44	0·92	0·51	0·60
Orn	tr	0·43	tr	tr	tr	tr

A: Deionised water; B: 0·05 M NaCl; C: Sörensen phosphate buffer pH 7; D: 0·1 M Tris-HCl buffer pH 8·3; E: 0·1 M NaOH.

Table 1 shows that increasing ionic strength reduces the protein extractability, which was considerably lower in NaCl solutions than in $MgCl_2$ solutions. In addition, it seems that concentrations above 0·1 M with any extractant reduce its efficacy. Extractability was not greatly affected by temperatures over 50°C or by more than 30 min of extraction.

On the other hand, large amounts of solvent obviously aid the extraction of proteins from skin, the greatest extractability being obtained with a solid/liquid ratio of 1:2000, but extraction efficacy using a ratio of 1:1000 is less inferior than that observed for seed protein.

As observed with seed protein, pH has a major effect upon extractability of skin proteins. The best yield in salt solutions was obtained at pH 11, but yields observed between this value and pH 7 were nearly as good.

Table 2 shows the amino acid composition of the extracted grape skin proteins which, in general, is similar to that of grapeseed. The major component in every extract was glutamic acid (about 12%) followed by aspartic acid and leucine (about 10%). With respect to the minor components from the grape skin proteins, these show some differences from grape seed proteins (Defrancesco *et al* 1975; Fazio *et al* 1975; Castriotta and Canella 1978; Fantozzi and Bestchart 1979; Fantozzi 1981; Kamel *et al* 1985; Igartuburu *et al* 1991). The limiting amino acids in grape skin

proteins appeared to be tryptophan, methionine and cystine. Lysine was not limiting, in contrast to the seed proteins. Other amino acids found in small amounts were histidine and proline. All extracts showed the presence of small amounts of ornithine.

In contrast to the seed proteins, skin proteins were rich in alanine (about 7%; about 1·5% in seed proteins); their digestibility was lower (64·8% of 76·3% for seed proteins) but protein content was higher (14·7% of 8·4%) (Massanet *et al* 1987).

All these characteristics, together with the higher level of lysine (6·5%), suggest that the skin proteins have better potential for feed use than those of the seeds and some cereals (Wu and Sex 1976; Nuffield Foundation 1984; Husea Lope *et al* 1985), and from this point of view they could be substituted for these materials where they are used as food protein supplements or feed additives.

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