Phytochemistry, Vol. 22, No. 2, pp. 477–481, 1983.
Printed in Great Britain.

0031–9422/83/020477–05$03.00/0

QUINIC ACID ESTERS OF HYDROXYCINNAMIC ACIDS IN STONE AND POME FRUIT

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(*Received* 5 *May* 1982)

Key Word Index—Rosaceae; stone fruit; pome fruit; caffeoyl-, *p*-coumaroyl- and feruloylquinic acids.

Abstract—In 21 varieties of stone and pome fruit the quinic acid esters of caffeic, *p*-coumaric and ferulic acids have been determined by capillary GC and HPLC. The total content of hydroxycinnamoylquinic acids was found to be between 80 and almost 900 ppm fr. wt. The results confirm that the main hydroxycinnamic acid compound in pome fruit is 5′-caffeoylquinic acid, whereas in stone fruit it is 3′-caffeoylquinic acid. Our investigations also show the regular occurrence of 3′-*p*-coumaroylquinic acid in stone fruit species. In sweet and sour cherries it can be the main component in concentrations of 100–200 ppm. Significant amounts of feruloylquinic acid are only found in plums and apricots. The 4′-isomers of all acids occur only in very small concentrations with the exception of 4′-*p*-coumaroylquinic acid in apples, the content of which exceeds that of 5′-*p*-coumaroylquinic acid.

INTRODUCTION

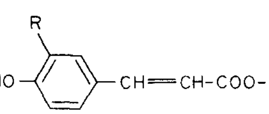
The phenolic constituents of Rosaceae fruits include catechins and proanthocyanidins as well as hydroxycinnamic acid derivatives. Moreover, small concentrations of flavonol glycosides are present in sweet and sour cherries [1], plums [1], apricots [1], peaches [1], apples, pears and berries [2] and anthocyanins in dark coloured fruit. The hydroxycinnamic acids exist partly as glucose esters, but mainly as D-quinic acid esters [3]. In 1972 we summarized the phenolic substances of fruit [4]. Since then more work on hydroxycinnamic acid compounds in fruit has been published (cherry [5], peach [6], apple [7], pear [8, 9], various fruit species [10]).

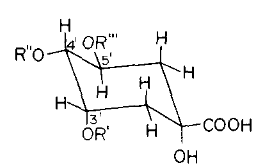
All the previous work on the hydroxycinnamoylquinic acid content of fruit (see Table 4) was based on PC or TLC analysis. Since separation can only be achieved in various successive steps, inaccuracies in quantitative determinations through losses, especially at low concentrations are highly probable. Also, in some work only the sum of the positional isomers was indicated, whereas in other publications it is uncertain whether the method of determination permitted the identification of the positional isomers at all. Hardly any quantitative data exist on *p*-coumaroyl- and feruloylquinic acids. Therefore, we have developed a GC- and a HPLC-method which allow a complete separation of all the possible positional isomers. With this it is possible to determine concentrations as low as 1 ppm and some uncertainties about previous work can be eliminated.

RESULTS AND DISCUSSION

In the present study hydroxycinnamoylquinic acids have been determined in 21 varieties of stone and pome fruit. Table 1 lists the hydroxycinnamoylquinic acids which have been identified. We have chosen the new IUPAC nomenclature instead of the older but still applicable nomenclature. In the IUPAC system the former 3-acyl quinic acids, such as chlorogenic acid, are now marked as the 5′-compounds and the former 5-acyl quinic acids, such as neochlorogenic acid, as the 3′-compounds, respectively. The quinic ester contents of the fruit are determined as the sum of the *cis* and *trans*-isomers. The results are represented in Tables 2 and 3. All the fruits analysed were fully ripe and, therefore, in a comparable state of maturity so that significant differences in the levels of individual esters between varieties are not the result of differences in maturity.

Table 1. Nomenclature of hydroxycinnamoylquinic acids





Quinic acid

p-Coumaroyl	R = H (*p*-CoumQ)	
Caffeoyl-	R = OH (CafQ)	
Feruloyl-	R = OMe (FerQ)	
Chlorogenic acid	= 5′-caffeoylquinic acid	
Cryptochlorogenic acid	= 4′-caffeoylquinic acid	
Neochlorogenic acid	= 3′-caffeoylquinic acid	

The results of the analyses by the two methods employed were in close agreement with each other thus allowing positive identification of positional isomers. Isomerization of the positional isomers during the preparation or the analysis can be excluded since with isomerization, starting with the 3′- or 5′-compound, the 4′-compound is always formed first. In our analyses with many samples, however, the 4′-compound was not found. Also, in agreement with previous work, no free hydroxycinnamic acids were detected. Values between 80 and almost 900 ppm were found for the total content of hydroxycinnamoylquinic acids in fresh fruit.

Table 2. Contents (ppm fr. wt) of hydroxycinnamoylquinic acids in stone and pome fruit by capillary GC

Fruit variety	5′-CafQ	4′-CafQ	3′-CafQ	5′-*p*-CoumQ	4′-*p*-CoumQ	3′-*p*-CoumQ	5′-FerQ	4′-FerQ	3′-FerQ	Total
Sweet cherries										
Souvenir de Charme	23	3	75	1	11	172	+	—	+	285
Maibigarreau	18	7	130	—	12	211	1	—	4	383
Grosse Schwarze Knorpel	25	21	620	—	8	84	+	—	8	766
Sour cherries										
Schattenmorelle Bockelmann	53	1	79	2	11	192	2	—	+	340
Successa	74	4	96	+	9	125	3	—	+	311
Kelleris 16	93	10	400	+	25	101	5	—	3	637
Plums										
First	18	—	98	+	—	4	1	—	9	130
Grosse Grüne Reneclode	66	46	694	—	+	19	6	—	18	849
Hauszwetsche	38	—	438	—	—	28	3	—	25	532
Nancy Mirabelle	136	—	362	2	4	12	7	—	34	557
Apricots										
Ungarische Beste	51	—	26	+	2	2	2	—	3	86
Uhlhorns Wunder	41	+	59	—	—	3	3	—	4	110
Moorpark	70	—	67	+	1	4	3	—	6	151
Aprikose von Nancy	55	—	55	1	+	5	3	—	5	124
Mombacher Frühe	109	—	81	2	—	7	7	—	12	218
Peaches										
South Haven	38	—	29	+	—	2	2	—	1	72
Red Haven	254	—	128	+	—	3	9	—	+	394
Apples										
Thydemanns Early Worcester	435	—	—	10	15	—	2	—	—	462
Jamba	318	—	—	12	46	—	4	—	—	380
Pears										
Conference	145	—	—	2	—	—	—	—	+	147
Gellerts Butterbirne	516	—	—	—	—	—	—	—	2	518

+, Trace; —, not detectable.

Table 3. Contents of (ppm fr. wt) of hydroxycinnamoylquinic acids in stone and pome fruit by HPLC

Fruit variety	5′-CafQ	4′-CafQ	3′-CafQ	5′-*p*-CoumQ	4′-*p*-CoumQ	3′-*p*-CoumQ	5′-FerQ	4′-FerQ	3′-FerQ	Total
Sweet cherries										
Souvenir de Charme	25	—	82	—	12	183	—	—	—	302
Maibigarreau	18	5	141	—	15	218	—	—	5	402
Grosse Schwarze Knorpel	23	18	628	—	11	102	—	—	8	790
Sour cherries										
Schattenmorelle										
Bockelmann	55	—	82	3	7	200	—	—	2	349
Successa	65	—	86	—	7	113	—	—	—	271
Kelleris 16	96	8	405	—	30	106	2	—	4	651
Plums										
First	15	—	88	—	—	4	2	—	12	121
Grosse Grüne Reneclode	70	56	731	—	—	24	—	—	15	896
Hauszwetsche	42	—	448	—	—	36	—	—	18	544
Nancy Mirabelle	129	—	362	—	—	15	—	—	29	335
Apricots										
Ungarische Beste	49	—	26	—	—	2	2	—	4	83
Uhlhorns Wunder	37	—	55	—	—	2	—	—	4	98
Moorpark	66	—	60	—	—	4	—	—	5	134
Aprikose von Nancy	49	—	50	—	—	4	—	—	5	108
Mombacher Frühe	103	—	78	3	—	7	—	—	12	200
Peaches										
South Haven	43	—	33	—	—	3	—	—	2	81
Red Haven	268	—	142	—	—	4	—	—	2	416
Apples										
Thydemanns Early										
Worcester	428	—	—	10	15	—	—	—	—	453
Jamba	339	—	—	11	39	—	—	—	—	389
Pears										
Conference	156	—	—	—	—	—	—	—	—	156
Gellerts Butterbirne	518	—	—	—	—	—	—	—	—	518

—, Not detectable.

All fruit with the exception of pears contained quinic acid esters of caffeic, *p*-coumaric and ferulic acids. In contrast, no sinapic acid esters were found in any sample, again in agreement with previous results [13, 14]. Also, in our analyses of fruit species for hydroxycinnamic acid glucose esters [15], we were unable to find sinapic acid compounds in stone or pome fruit.

In general the caffeic acid esters were predominant, whereas *p*-coumaric and ferulic acid esters accounted for 10% or less of the total hydroxycinnamic acid esters present. In the case of cherries, however, the *p*-coumaroylquinic acids may constitute more than half of the total hydroxycinnamoylquinic acids present.

The hydroxycinnamic acid patterns of stone and pome fruit differ considerably. Thus, in *Prunus* species the 3'-isomers are major constituents, whereas the 5'-isomers are found almost exclusively in apples and pears. This is in agreement with literature data (Table 4). The 4'-isomers play a subordinate role in all samples. 4'-Feruloylquinic acid was not detected in any sample. However, in apples 4'-*p*-coumaroylquinic acid was present in larger amounts than 5'-*p*-coumaroylquinic acid. Literature data on *p*-coumaroylquinic acids in apples are contradictory. For the variety 'Yarlington Mill', Williams [16] tentatively identified 5'-*p*-coumaroylquinic acid but later Whiting and Coggins [17] corrected this finding to 4'-*p*-coumaroylquinic acid. However, Macheix [7] found both compounds in the variety 'Calville Blanc' as we did in the varieties which we analysed. The amounts of the different hydroxycinnamoylquinic acids in various samples are very variable; the variety differences often being greater than the species differences. But with consideration of fluctuations in variety, ripeness and cultivation conditions, the determined values are largely in agreement with those given in the literature. Further, they correspond very well to our previous investigations [13, 14] on hydroxycinnamic acids in fruit after hydrolysis and TLC/spectrophotometric determination. In conclusion, hydroxycinnamic acids in ripe stone and pome fruit occur largely as quinic acid esters.

The main component in pome fruit is 5'-caffeoylquinic acid. In the case of stone fruit 3'-caffeoylquinic acid predominates in sweet and sour cherries and in plums, whereas in apricots and peaches the differences in concentration between the two caffeoylquinic acids are less marked and 5'-caffeoylquinic acid is often the major component. 4'-Caffeoylquinic acid, which was recorded in 1958 by Sondheimer [12] but seldom mentioned in later work, has been found in the present investigations only in very small concentrations.

We have also shown, for the first time, the regular occurrence of 3'-*p*-coumaroylquinic acid in stone fruit species; in sweet and sour cherries nearly 100–200 ppm. In contrast 5'-*p*-coumaroylquinic acid has been found only in trace amounts. 4'-*p*-Coumaroylquinic acid occurred in samples with a high content of 3'-*p*-coumaroylquinic acid with values of *ca* 10 ppm, otherwise it was only found in trace amounts. In apples, however, 4'-*p*-coumaroylquinic acid was the major *p*-coumaroylquinic ester. Feruloylquinic acids only occur in significant amounts in plums and apricots.

EXPERIMENTAL

Extraction and purification. 100 g depitted fruit was homogenized with 80% MeOH, made up to 1 l. with more MeOH, and extracted for 30 min with stirring and refluxing in N_2 at room temp. The extraction was repeated once more using the same conditions, the extracts combined and evaporated under red. pres. to an aq. residue (250 ml), which was purified on a polyamide column (250 × 35 mm i.d.; MN-Polyamid-SC-6, 0.05–0.16 mm, Macherey-Nagel, Düren, W. Germany). After sample application, the column was eluted with 800 ml H_2O (to remove sugars and fruit acids) and 800 ml MeOH (to remove hydroxycinnamoyl glucoses, flavonoids and anthocyanins) and 800 ml MeOH–HOAc (199:1).

GC conditions. Derivatization with BSA–TMCS (20:1). Carlo Erba GC 2150, FID, SE-30-glass capillary (WCOT, 37 m × 0.27 mm i.d.), 220–270°, 4°/min, injector and detector 300°.

HPLC conditions. Philips-Pye Unicam, LiChrosorb RP-18 (250 × 4 mm i.d., 5 μm, Merck, Darmstadt, West Germany), flow rate 0.8 ml/min, 320 nm. Gradient elution solvent: (A), 2%

Table 4. Concentrations (ppm fr. wt) of hydroxycinnamoylquinic acids of stone and pome fruit according to literature data

Fruit	5'-CafQ	4'-CafQ	3'-CafQ	*p*-CoumQ	Ref.
Cherries	1–5	—	20–240	—	[10]
	—	—	450	60	[5]
	10	50(?)	135	—	[12]
Plums	8	—	440	—	[10]
	90	5	1200	—	[12]
	35	20	50	—	[12]
Peaches	20–150			—	[6]
	40	—	50	—	[12]
Apples	620	—	—	—	[11]
	210	—	—	—	[10]
	500	—	—	70	[7]
	150	20	—	—	[12]
Pears	260			—	[9]
	220	30	10	—	[12]
	345	—	—	—	[10]

HOAc; (B), MeOH, from 7% to 15% in 10 min, then from 15% to 35% in 50 min.

The GC and HPLC methods, as well as the isolation of reference substances out of plant material, have been described in detail elsewhere [18].

Acknowledgements—We wish to thank Mr. A. M. Boudet (Centre de Physiologie Végétale, Université Paul Sabatier, Toulouse) and Mr. J. J. Macheix and Mrs. A. Fleuriet (Laboratoire de Physiologie Végétale, Université des Sciences et Techniques, Montpellier), who enabled the research stay of Mrs. B. Möller in their laboratories. By this cooperation the isolation of reference substances from plant material was achieved. We also thank Miss I. H. Slaney, University of Pretoria, for translation of this manuscript. The work was supported by a grant from Deutsche Forschungsgemeinschaft.

REFERENCES

1. Henning, W. and Herrmann, K. (1980) *Z. Lebensm.-Unters. Forsch.* **170**, 433; **171**, 111, 183.
2. Henning, W. (1981) *Z. Lebensm.-Unters. Forsch.* **173**, 180.
3. Herrmann, K. (1978) *Prog. Chem. Org. Nat. Prod.* **35**, 73.
4. Herrmann, K. (1973) *Z. Lebensm.-Unters. Forsch.* **151**, 41.
5. Melin, C., Billot, J. and Dupin, J. F. (1979) *Physiol. Veg.* **17**, 557.
6. Bureau, D., Macheix, J. J. and Rouet-Mayer, M. A. (1977) *Lebensm-Wiss. Technol.* **10**, 211.
7. Macheix, J. J. (1974) *Physiol. Veg.* **12**, 25.
8. Fuertes-Lasala, E., Fernández, M., Carcía-Mina, C., Vega, F. A. and Valls, L. M. (1974) *An. Bromatol.* **26**, 59.
9. Billot, J., Hartmann, C., Macheix, J. J. and Rateau, J. (1978) *Physiol. Veg.* **16**, 693.
10. Hanefeld, M. and Herrmann, K. (1976) *Deutsche Lebensm.-Rundsch.* **72**, 383.
11. Fleuriet, A. and Macheix, J. J. (1974) *J. Chromatogr.* **74**, 339.
12. Sondheimer, E. (1958) *Arch. Biochem. Biophys.* **74**, 131.
13. Mosel, H.-D. and Herrmann, K. (1974) *Z. Lebensm.-Unters. Forsch.* **154**, 6.
14. Stöhr, H., Mosel, H.-D. and Herrmann, K. (1975) *Z. Lebensm-Unters. Forsch.* **159**, 85.
15. Reschke, A. and Herrmann, K. (1981) *Z. Lebensm.-Unters. Forsch.* **173**, 458.
16. Williams, A. H. (1958) *Chem. Ind.* 1200.
17. Whiting, G. C. and Coggins, R. A. (1975) *Phytochemistry* **14**, 593.
18. Möller, B. and Herrmann, K. (1982) *J. Chromatogr.* **241**, 371.