The Flavonol Glycosides in the Fruit of *Pyrus communis* L. cultivar Bon Chrétien

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1. Two new flavonol glycosides were isolated from the fruit of *Pyrus communis* L. cultivar Bon Chrétien. These were identified as isorhamnetin 3-rhamnogalactoside and a derivative of isorhamnetin 3-glucoside which was associated (possibly acylated) with an unknown aliphatic organic acid. 2. The melting point of isorhamnetin 3-glucoside isolated from Bon Chrétien pears is different from that of isorhamnetin 3-glucoside previously isolated from *Argemone mexicana* and *Calendula officinalis*. 3. Isorhamnetin 3-rhamnoglucoside was isolated from the fruit of *Pyrus communis* L. cultivar Bon Chrétien. This glycoside appears to be identical with narcissin, previously isolated from *Narcissus tazetta* and *Lilium auratum*. 4. Isoquercitrin, previously reported to be present in pear leaves, was isolated from the fruit of Bon Chrétien pears. 5. The isolated glycosides were present in the peels and flesh of the fruit, but were absent from the cores.

No detailed study appears to have been made of the polyphenolic constituents of the fruit of cultivated pear varieties. The presence of a leuco-anthocyanin in various parts of the fruit of *Pyrus communis* L. cultivar Bon Chrétien was reported by Joslyn & Peterson (1956), cyanidin being the only anthocyanin produced on treatment of an extract of the cores with boiling, dilute aqueous hydrochloric acid. Kieser, Pollard & Williams (1953) also reported the presence of a leuco-anthocyanin in the perry cultivars examined. This substance was found to be of a colloidal nature and quite immobile on paper chromatograms (Williams, 1960). Chlorogenic acid was found by Williams (1960) to be the major phenolic compound present in the fruit of dessert pears, and Cartwright, Roberts, Flood & Williams (1955) tentatively identified *p*-coumarylquinic acids in the fruit. The present study provides details of the isolation and characterization of five flavonol glycosides present in the peels of the fruit of *Pyrus communis* L. cultivar Bon Chrétien.

EXPERIMENTAL AND RESULTS

All melting points are corrected and were determined by the Kofler method. Mixed melting points were determined on the crystalline residue remaining after dissolving equal weights of both compounds in methanol and removing the methanol under reduced pressure (Roux & Maihs, 1960). Analyses of C, H, methoxyl and acetyl were made by K.

Jones, National Chemical Research Laboratory, C.S.I.R., Pretoria. Infrared spectra were recorded by the KBr-disk and Nujol-mull techniques with Perkin-Elmer model 21 and Infracord spectrophotometers. Absorption in the ultraviolet region was recorded with a Carl Zeiss model PMQII spectrophotometer.

Isolation and identification of the flavonol glycosides

Extraction from the peels. Bon Chrétien pears, grown on the experimental farm, Bien Donné, in the Groot-Drakenstein area, were harvested when the average pressure of the fruit was 18 lb. as measured by a Ballauf fruit tester fitted with a $\frac{1}{8}$ -in. plunger. The fruit was stored at 0° for 7 days before ripening at 21° and 85% r.h. The mature pears (90 kg.) were peeled by hand and the peels (about 3 kg.) were immediately dropped into acetone (10 l.) and macerated in a Waring Blendor. The macerate was filtered through Buchner funnels and the residue washed with several portions of aq. 50% (v/v) acetone. The combined extracts (about 18 l.) were concentrated under reduced pressure to about 4 l. and extracted continuously with chloroform (12 hr.) and ethyl acetate (40 hr.). The ethyl acetate in the collecting flask was replaced at intervals of 8–10 hr. The combined ethyl acetate extracts were evaporated under reduced pressure to yield a dark, sticky material (42.5 g.) containing the flavonol glycosides.

Separation by paper chromatography. The crude flavonol extract (41.2 g.) was dissolved in acetone (100 ml.) and filtered to remove the insoluble residue (3.7 g.). The solution was applied in 4 cm. bands along the shorter sides of 50 Whatman no. 3MM paper sheets (58 cm. × 68 cm.),

which were developed by downward migration in aq. 2% (v/v) acetic acid. The flavonol glycosides, visible as dark bands under ultraviolet light, separated roughly into three bands. The development was continued until band I (the most mobile) reached about 5 cm. from the edge of the paper sheets. The chromatograms were dried in a current of air at 35–40° and the individual bands were cut out and eluted with aq. 70% (v/v) ethanol. Each eluate was concentrated under reduced pressure and rechromatographed twice in the same manner, 40 paper sheets being used for each fraction. By this method bands I, II and III could be obtained uncontaminated by one another.

The eluate from band I was rechromatographed on 60 Whatman no. 3MM paper sheets (58 cm. × 68 cm.) with aq. 75% (v/v) phenol as solvent to separate the two components (A and B) present. Development was continued until component A (the more mobile) reached a distance of about 5 cm. from the end of the paper. After drying, the bands containing components A and B were cut out and eluted with aq. 70% (v/v) ethanol.

The eluate from band II contained only one component (C) by chromatography in several solvent systems.

The eluate from band III (the least mobile in aq. 2% acetic acid) was further purified by chromatography in aq. 75% (v/v) phenol as described for band I. A total of 30 Whatman no. 3MM paper sheets (58 cm. × 68 cm.) was used. The material separated into two bands (D and E) when development was continued until component D (the more mobile) reached about 5 cm. from the end of the chromatograms. The components were recovered by elution with aq. 70% (v/v) ethanol. After concentration of eluates A, B, C, D and E under reduced pressure, the solutions were filtered and set aside for crystallization. Crystals separated from the aqueous concentrates in each case.

Isorhamnetin 3-glucoside. The material which separated from eluate D was recrystallized twice from aq. ethanol and dried under reduced pressure at 110° over P₂O₅ for 6 hr. The yellow needles (339 mg.) had m.p. 242–244°; $[\alpha]_D^{20} - 44.1 \pm 1.5^\circ$ (c 1.0 in pyridine); $\lambda_{\max}$. 365, 270 (inflection) and 255 m μ in ethanol (log ϵ 4.26, 4.30 and 4.33 respectively); $\lambda_{\max}$. 407 and 270 m μ in ethanolic 75 mM-aluminium chloride; $\lambda_{\max}$. 423 and 275 m μ in ethanolic 2 mM-sodium ethoxide; $\lambda_{\max}$. 368 and 270 m μ in saturated ethanolic sodium acetate (Jurd & Horowitz, 1957); $\lambda_{\max}$. 364 and 256 m μ in ethanolic 0.15 M-boric acid saturated with sodium acetate (Jurd, 1956) (Found: C, 54.5; H, 4.9; OMe, 6.8. C₂₂H₂₂O₁₂ requires C, 55.2; H, 4.6; OMe, 6.5%).

Hydrolysis of the compound (81.5 mg.) in 2N-HCl (5 ml.) for 90 min. at 100° yielded a yellow precipitate which was filtered off and washed free of acid. The aglycone (51.0 mg., a 93% theoretical yield for a monoglucoside of isorhamnetin) had m.p. 306–308°, unchanged by admixture with authentic isorhamnetin (kindly supplied by Dr R. M. Horowitz). The isolated aglycone was identical with isorhamnetin on chromatography in various solvent systems and by comparison of the i.r. (2.5–15 μ) and u.v. (230–450 m μ) spectra over the ranges indicated. The aglycone (25 mg.) was acetylated with pyridine (0.1 ml.) and acetic anhydride (0.1 ml.) to yield a crystalline acetate (18 mg.), m.p. 207–209° (cf. isorhamnetin tetra-acetate, m.p. 208–210°; Gripenberg, 1962) (Found: C, 59.4; H, 4.3; OMe, 6.6; CO·CH₃, 35.8. Calc. for C₂₄H₂₀O₁₁: C, 59.5; H, 4.2; OMe, 6.4; CO·CH₃, 35.5%). The aqueous acid hydro-

lysate, after removal of the precipitated aglycone, was neutralized, desalted electrolytically, concentrated to about 1 ml. and treated with phenylhydrazine hydrochloride (40 mg.) and sodium acetate (60 mg.) at 100° for 7 min. to obtain characteristic needles of D-glucosazone, m.p. and mixed m.p. 206° (decomp.).

That the glucosyl residue is attached to the flavonol in the 3-position was determined as follows: the purified glycoside from eluate D (80 mg.) was methylated in an excess of diazomethane in ether-methanol at –15° and hydrolysed in 2N-HCl (10 ml.) at 100° for 2 hr. The product crystallized from ethanol as cream-coloured needles (25 mg.), m.p. 195–196°, undepressed by admixture with authentic 3',4',5,7-tetramethoxyflavonol. Acetylation of the methyl ether (23 mg.) with acetic anhydride (0.1 ml.) and pyridine (0.1 ml.) and recrystallization of the product from ethanol yielded cream-coloured needles (15 mg.), m.p. 159–161°, undepressed by admixture with authentic 3-acetoxy-3',4',5,7-tetramethoxyflavone.

Compound C. The crystalline material which separated from eluate C was recrystallized twice from aq. ethanol and dried under reduced pressure at 110° over P₂O₅ for 6 hr. The yellow needles (407 mg.) had m.p. 167–168°; $[\alpha]_D^{20} - 30.6 \pm 1.0^\circ$ (c 1.1 in pyridine); $\lambda_{\max}$. 364, 270 (inflection) and 256 m μ in ethanol; $\lambda_{\max}$. 406 and 270 m μ in ethanolic 75 mM-aluminium chloride; $\lambda_{\max}$. 423 and 274 m μ in ethanolic 2 mM-sodium ethoxide; $\lambda_{\max}$. 370 and 270 m μ in saturated ethanolic sodium acetate; $\lambda_{\max}$. 364 and 257 m μ in ethanolic 0.15 M-boric acid saturated with sodium acetate (Found: C, 50.5; H, 4.9%). The i.r. spectrum of the new compound showed, in addition to the expected heterocyclic carbonyl absorption of a 5-hydroxyflavonol at 1652 cm.⁻¹ (Lebreton & Chopin, 1963), a high-intensity absorption band at 1725 cm.⁻¹.

Hydrolysis of the compound (100.6 mg.) in 2N-HCl (10 ml.) for 90 min. at 100° yielded a yellow precipitate (56.6 mg.), m.p. 306–308°, undepressed on admixture with authentic isorhamnetin. The material was identical with isorhamnetin by chromatography. The aqueous hydrolysate, after removal of the precipitated aglycone, was neutralized, desalted electrolytically and concentrated to about 1 ml. Treatment with phenylhydrazine hydrochloride (40 mg.) and sodium acetate (60 mg.) at 100° for 7 min. yielded characteristic needles of D-glucosazone, m.p. and mixed m.p. 206° (decomp.).

The crystalline substance from eluate C was methylated with diazomethane and hydrolysed in 2N-HCl as described for isorhamnetin 3-glucoside. The product had m.p. 195–196°, unchanged by admixture with authentic 3',4',5,7-tetramethoxyflavonol. Acetylation of the methyl ether yielded 3-acetoxy-3',4',5,7-tetramethoxyflavone, m.p. 158–160°, unchanged by admixture with an authentic specimen.

The purified material from eluate C (100 mg.) was treated with aq. 5% (w/v) NaOH (20 ml.) for 3 hr. at 30° before acidification with 10N-H₂SO₄ to pH 1.5. The yellow precipitate which formed recrystallized from aq. ethanol as yellow needles (53 mg.), m.p. 242–244°, undepressed on admixture with isorhamnetin 3-glucoside obtained from eluate D. The filtrate, after removal of the precipitate, was extracted continuously with ether for 48 hr. The ethereal extract was dried over anhydrous Na₂SO₄ and purified by chromatography on a single Whatman no. 3MM paper sheet (46 cm. × 57 cm.), with pentan-1-ol-5M-formic acid (1:1, v/v) as developing solvent. The major component was

detected by spraying a strip along the side of the paper sheet with ethanolic 0.04% (w/v) bromophenol blue (Buch, Montgomery & Porter, 1952). The band which showed a positive reaction towards this reagent was cut out and eluted with aq. 70% (v/v) ethanol. The eluate was concentrated under reduced pressure to a small volume and extracted with diethyl ether. On evaporation of the ether, colourless needles (5 mg.), m.p. 129–134° (decomp.), were obtained. This product was not obtained on ether extraction of an alkaline solution (pH 8.0), indicating further the acidic nature of the compound. The structure of this acid is being investigated (see the Discussion section).

Isorhamnetin 3-rhamnogalactoside. The crystals which separated from eluate A were recrystallized twice from aq. ethanol and dried under reduced pressure at 110° over P₂O₅ for 6 hr. to yield yellow needles (80 mg.), m.p. 183–185°; $[\alpha]_D^{20} - 32.6 \pm 1.1^\circ$ (c 1.0 in pyridine); $\lambda_{\max}$. 363, 270 (inflection) and 255 m μ in ethanol (log ϵ 4.23, 4.26 and 4.30 respectively); $\lambda_{\max}$. 408 and 270 m μ in ethanolic 75 mm-aluminium chloride; $\lambda_{\max}$. 423 and 275 m μ in ethanolic 2 mm-sodium ethoxide; $\lambda_{\max}$. 370 and 270 m μ in saturated ethanolic sodium acetate; $\lambda_{\max}$. 362 and 255 m μ in ethanolic 0.15 M-boric acid saturated with sodium acetate (Found: C, 53.1; H, 5.5; OMe, 4.7. C₂₈H₃₂O₁₆ requires C, 53.9; H, 5.2; OMe, 5.0%).

Hydrolysis of the compound (15.0 mg.) in 2N-HCl (5 ml.) for 90 min. at 100° produced a yellow precipitate (6.8 mg., a 95% theoretical yield for a rhamnogalactoside of isorhamnetin), m.p. 306–308°, unchanged by admixture with authentic isorhamnetin. The precipitated material was inseparable from authentic isorhamnetin on paper chromatograms and these substances had identical i.r. (2.5–15 μ) and u.v. (230–450 m μ) spectra. The aq. acid hydrolysate, after removal of the precipitated aglycone, was neutralized and desalted electrolytically. The two sugars present in the hydrolysate were identified as rhamnose and galactose respectively by chromatography in several solvent systems and spraying with aniline hydrogen oxalate (Horrocks & Manning, 1949). The solution contained 3.1 mg. of rhamnose (78% theoretical yield for an isorhamnetin diglycoside) and 3.8 mg. of galactose (88% theoretical yield for an isorhamnetin diglycoside), determined by the spectrophotometric method of Pridham (1956), modified by Harborne (1960).

The purified material from eluate A (10 mg.) was refluxed for 5 hr. with cyclohexanol-formic acid (3:1, v/v) (5 ml.), as described by Fox, Savage & Wender (1953). Examination by paper chromatography revealed the presence of a new compound in the reaction mixture. This substance was separated by chromatography on a single Whatman no. 3MM paper sheet (46 cm. $\times$ 57 cm.) with aq. 2% (v/v) acetic acid as solvent, eluted from the chromatogram with aq. 70% (v/v) ethanol and hydrolysed in 2N-HCl to yield isorhamnetin and galactose, identified by paper chromatography.

That the rhamnogalactosyl residue is attached to the flavonol in the 3-position was determined as follows. The purified substance obtained from eluate A (80 mg.) was methylated with diazomethane and hydrolysed in 2N-HCl to yield a crystalline product (35 mg.), m.p. 195–196°, identical with authentic 3',4',5',7-tetramethoxyflavonol by mixed m.p., paper chromatography, u.v. and i.r. spectroscopy. Acetylation of the methyl ether (30 mg.) yielded a crystalline acetate (18 mg.), m.p. 159–161°, undepressed

by admixture with authentic 3-acetoxy-3',4',5',7-tetramethoxyflavone.

Isorhamnetin 3-rhamnoglucoside. The solid which separated from eluate B was recrystallized twice from aq. ethanol and dried under reduced pressure at 110° over P₂O₅ for 6 hr. to yield yellow needles (168 mg.), m.p. 180–182°; $[\alpha]_D^{20} - 30.2 \pm 1.0^\circ$ (c 1.1 in pyridine); $\lambda_{\max}$. 363, 270 (inflection) and 256 m μ in ethanol (log ϵ 4.22, 4.24 and 4.29 respectively); $\lambda_{\max}$. 408 and 270 m μ in ethanolic 75 mm-aluminium chloride; $\lambda_{\max}$. 420 and 274 m μ in ethanolic 2 mm-sodium ethoxide; $\lambda_{\max}$. 370 and 270 m μ in saturated ethanolic sodium acetate; $\lambda_{\max}$. 363 and 255 m μ in ethanolic 0.15 M-boric acid saturated with sodium acetate (Found: C, 53.3; H, 5.3; OMe, 5.5. Calc. for C₂₈H₃₂O₁₆: C, 53.9; H, 5.2; OMe, 5.0%).

Hydrolysis of the compound (15.0 mg.) in 2N-HCl (5 ml.) for 90 min. at 100° produced a yellow precipitate (6.5 mg., a 95% theoretical yield for a rhamnoglucoside of isorhamnetin), m.p. 306–308°, unchanged by admixture with authentic isorhamnetin. The precipitated material was inseparable from isorhamnetin on paper chromatograms and had identical u.v. (230–450 m μ) and i.r. (2.5–15 μ) spectra. The aq. acid hydrolysate, after removal of the precipitated aglycone, was neutralized and desalted electrolytically. The sugars present in this solution were identified as rhamnose and glucose respectively by chromatography in several solvent systems and spraying with aniline hydrogen oxalate (Horrocks & Manning, 1949). The solution contained 3.2 mg. of rhamnose (83% theoretical yield for an isorhamnetin diglycoside) and 3.8 mg. of glucose (88% theoretical yield for an isorhamnetin diglycoside), determined spectrophotometrically. Controlled acid hydrolysis with cyclohexanol-formic acid as described for isorhamnetin 3-rhamnogalactoside yielded a monoglucoside which was inseparable from isorhamnetin 3-glucoside (obtained from eluate D) by chromatography in aq. 2% (v/v) acetic acid, aq. 15% (v/v) acetic acid, aq. 60% (v/v) acetic acid, aq. 75% (v/v) phenol and butan-1-ol-acetic acid-water (20:5:11, by vol.).

The attachment of the rhamnoglucosyl residue in the 3-position of the flavonol was confirmed by methylation of the purified substance obtained from eluate B (80 mg.) and hydrolysis in the usual way to yield 3',4',5',7-tetramethoxyflavonol (35 mg.), m.p. and mixed m.p. 195–196°. Acetylation of this product yielded 3-acetoxy-3',4',5',7-tetramethoxyflavone (18 mg.), m.p. and mixed m.p. 159–161°.

Isoquercitrin. The crystals which separated from eluate E were recrystallized twice from aq. ethanol and dried under reduced pressure at 110° over P₂O₅ for 6 hr. to yield yellow needles (209 mg.), m.p. 223–224°, m.p. 221–223° admixed with isoquercitrin, m.p. 221–223°, isolated from *Aspalathus acuminatus* (Koeppen, Smit & Roux, 1962); $[\alpha]_D^{20} - 40.4 \pm 1.1^\circ$ (c 1.1 in pyridine); $\lambda_{\max}$. 366, 270 (inflection) and 260 m μ in ethanol (log ϵ 4.28, 4.35 and 4.38 respectively); $\lambda_{\max}$. 419 and 275 m μ in ethanolic 75 mm-aluminium chloride; $\lambda_{\max}$. 416 and 275 m μ in ethanolic 2 mm-sodium ethoxide; $\lambda_{\max}$. 372 and 270 m μ in saturated ethanolic sodium acetate; $\lambda_{\max}$. 380 and 256 m μ in ethanolic 0.15 M-boric acid saturated with sodium acetate (Found: C, 53.5; H, 4.6. Calc. for C₂₁H₂₀O₁₂: C, 54.3; H, 4.3%).

Hydrolysis of the compound in 2N-HCl yielded quercetin, identified by m.p., mixed m.p., paper chromatography, u.v. (230–450 m μ) and i.r. (2.5–15 μ) spectra. The sugar

released on hydrolysis was identified, by paper chromatography, as glucose.

The purified substance obtained from eluate E (80 mg.) was methylated and hydrolysed to yield 3',4',5,7-tetramethoxyflavonol (35 mg.), m.p. 195–196°, identified by mixed m.p., paper chromatography, u.v. (230–450 m μ) and i.r. (2.5–15 μ) spectra. Acetylation of the methyl ether (30 mg.) yielded 3-acetoxy-3',4',5,7-tetramethoxyflavone (18 mg.), m.p. and mixed m.p. 159–161°. The identity of component E as isoquercitrin is therefore unequivocally established.

DISCUSSION

The presence of flavonol glycosides in *Pyrus* species was reported by Wagenbreth (1959) but no positive identifications were made. Kaempferol and quercetin glycosides have been found in the leaves of *Pyrus communis* (Williams, 1960) and recently it was reported that apigenin and luteolin glycosides are present in the leaves of certain *Pyrus* species (Williams, 1964). Of the various flavonoid compounds found in pear leaves, only isoquercitrin (quercetin 3-glucoside) could be detected in the fruit of the Bon Chrétien cultivar during the present investigation.

Known glycosides of isorhamnetin include isorhamnetin 3- α -L-arabinoside (distichin), isolated from the leaves of *Taxodium distichum* (Kariyone, Takahashi, Ito & Masutani, 1960), isorhamnetin 3,4'-diglucoside (dactylin), present in the pollen of timothy and orchard grass (Moore & Moore, 1931), and isorhamnetin 3-rutinoside (narcissin), isolated from the corolla, sepals and paracorolla of *Narcissus tazetta* (Kubota & Hase, 1956) and the pollen of *Lilium auratum* (Arakawa, 1956). Narcissin appears to be identical with the isorhamnetin 3-rhamnoglucoside isolated from Bon Chrétien pears, melting at 180–182° and consisting of the same components in the same sequence. Isorhamnetin 3-glucoside and isorhamnetin 3-rhamnoglactoside, isolated from the peels of Bon Chrétien pears in the present investigation, appear to be new glycosides of isorhamnetin. Although Rahman & Ilyas (1962) have reported the isolation of isorhamnetin 3-glucoside (m.p. 165–167°) from the flowers of *Argemone mexicana*, and Friedrich (1962) has reported the isolation of this compound in amorphous form (m.p. 168–172°) from *Calendula officinalis*, it would appear that neither of these compounds is identical with the isorhamnetin 3-glucoside (m.p. 242–244°) of Bon Chrétien pears. The two diglycosides, isorhamnetin 3-rhamnoglactoside and isorhamnetin 3-rhamnoglucoside, are not easily separated by chromatography and the only method found suitable for separation was by paper chromatography using aq. 75% (v/v) phenol as developing solvent. Tappi & Menziani (1955) isolated a flavonoid substance from *Lilium candidum* which yielded isorhamnetin, glucose, rham-

nose and galactose on hydrolysis and was considered to be isorhamnetin glucosido-rhamnido-galactoside. Since this material does not appear to have been obtained in the crystalline state it seems possible that it might have been a mixture of isorhamnetin 3-rhamnoglucoside and isorhamnetin 3-rhamnoglactoside.

The isolation of what appears to be an acylated isorhamnetin 3-glucoside from Bon Chrétien pears is of special interest since all the known acylated flavonols are acylated with aromatic acids (Birkhofer & Kaiser, 1962; Harborne, 1963, 1964). The organic acid derived from this compound could be readily distinguished from *p*-coumaric acid, ferulic acid and caffeic acid, which are commonly associated with acylated flavonoids (Harborne, 1964). Further evidence to this effect is derived from the R_F values of isorhamnetin 3-glucoside (R_F 0.71) and acylated isorhamnetin 3-glucoside (R_F 0.70) in butan-1-ol-acetic acid-water (20:5:11, by vol.). Acylation of anthocyanins with aromatic acids results in higher R_F values in this solvent system due to decreased polarity of the molecule (Harborne, 1958). The above R_F values indicate that acylation does not change the polarity of the glucoside significantly. The non-aromatic nature of the acid was indicated by the absence of characteristic absorption bands in the infrared spectrum and the absence of a light-absorption maximum in the ultraviolet (210–325 m μ) region.

Comparison of the elementary analyses for isorhamnetin 3-glucoside and compound C, which appears to be an acyl derivative of this glucoside, reveals that the percentage of carbon atoms in the latter is significantly lower than in the former. This further argues against the acyl residue being aromatic as all the known benzoyl or cinnamoyl residues associated with flavonoid compounds would raise, and not lower, the percentage of carbon atoms in the acylated flavonol. The finding further suggests that the residue is highly oxygenated and the acid liberated by alkaline hydrolysis might therefore be dicarboxylic. Examination by paper chromatography and mixed melting points revealed, however, that this acid is different from malic acid, maleic acid, malonic acid, succinic acid, fumaric acid, tartaric acid, α -oxoglutaric acid and aconitic acid. When the method described for the isolation of the unknown acid from component C was applied to component D (isorhamnetin 3-glucoside), no trace of the formation of this product could be detected. The isolated acid therefore arises from some structural feature of component C not present in component D even though the former has also been shown to be an isorhamnetin 3-glucoside. As the infrared spectrum of component C and its alkaline hydrolysis to isorhamnetin 3-glucoside strongly suggest the presence of an ester

linkage in the parent compound, it is tentatively suggested that the compound contains the acyl residue of the unknown acid which has been isolated. The spectral properties of component C and the preparation of the tetramethyl derivative of the flavonol suggest that the acyl residue is attached to one of the alcoholic hydroxyl groups of the glucosyl residue.

Examination of the different parts of the Bon Chrétien pear fruit by means of paper chromatography revealed that the isolated glycosides are present in small amounts in the flesh, but their presence could not be established in extracts from the cores.

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