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Glycosidically Bound Volatile Components in Apricot (*Prunus armeniaca* var. ansu Max.)

Young-Hoi Kim, Kun-Soo Kim, Joon-Young Park and Yong-Tae Kim
Korea Ginseng and Tobacco Research Institute, Taejon

Abstract

Glycosidically bound fraction was separated from apricot by Amberlite XAD-2 adsorption and eluted with methanol. Aglycones were liberated from the bound fraction by enzymatic hydrolysis, acid hydrolysis or by means of simultaneous distillation-extraction at pH 3.0. A total of 40 components were identified in three bound volatile fractions. Besides linalool oxide, linalool, α-terpineol, nerol, geraniol, benzyl alcohol and 2-phenylethyl alcohol, previously reported as glycosidically bound volatiles, the following components were identified for the first time as glycosidically bound volatiles in apricot; 2,6-dimethyl-3,7-octadiene-2,6-diol, 3,7-dimethyl-1,5-octadiene-3,7-diol, (E)- and (Z)-2,6-dimethyl-2,7-octadiene-1,6-diol, 3,4-didehydro-α-ionol, 3-oxo-α-ionol, 3-hydroxy-7,8-dihydro-β-ionol, 3-oxo-7,8-dihydro-α-ionol, 3-hydroxy-β-ionone, eugenol, 4-hydroxyethylphenyl acetate and 2,3-dihydrobenzofuran.

Key words: apricot, glycosidically bound volatiles, enzymatic and acid hydrolysis

Introduction

In the course of research on the volatile components of apricot in this laboratory, we have been found considerable differences in the compositions of volatiles by using different isolation techniques.[1] Under the condition of simultaneous distillation-extraction at atmospheric pressure, apricot at native pH value of 3.1 yielded far greater quantities of monoterpene alcohols, such as linalool, linalool oxides, α-terpineol, nerol and geraniol than those isolated by simultaneous distillation-extraction at neutral pH value of 7.0 or by use of headspace trapping technique.

With regard to the characteristic aroma of apricot, it is well-known that monoterpene alcohols and C_{13} norisoprenoids play an important role in apricot aroma because of their floral, fruity aroma character.[2,3] Recently, it was found that monoterpene alcohols in fruits are present either in a free form or bound to sugar in the form of glycosides.[4,5] In addition, Williams *et al.*[6,7] reported that monoterpene compounds were also produced by chemical rearrangement of nonvolatile diols and triols during heat treatment under weak acidic condition. Furthermore, β-D-glucosides of monoterpene alcohols and aromatic alcohols in apricot cv, Rouge du Roussillon were identified by Salles *et al.*[8]

The present study reports the occurrence of glycosidically bound volatile components which were not previously recognized in apricot.

Corresponding author: Young-Hoi Kim, Division of Chemical Research, Korea Ginseng and Tobacco Research Institute, Yusung-Gu 305-345, Taejon, Korea

Materials and Methods

Materials

Fresh, ripe apricots (*Prunus armeniaca* var. ansu Max.) were purchased from a local market.

Isolation of glycosidically bound fraction

A total of 3.0kg of deseeded apricot was blended with 6 l of methanol for 1 min in a Waring blender. The juice was cleared by vacuum filtration through a bed of celite 545. The filtrate was concentrated under reduced pressure to 500 m*l* at 40 °C and concentrate was then centrifuged at 4000 × g for 30 min. The clarified juice was extracted with diethyl ether (3 × 400 m*l*) to remove free volatiles. The juice was passed through a conditioned Amberlite XAD-2 (20-50 mesh, Fluka, Switzerland) adsorbent (2.5 × 70cm) at a flow rate of 3 m*l*/min according to the method of Gunata et al.[9] After the column was washed with 1.5 *l* of distilled water and 1 *l* of n-pentane to eleminate water soluble components and free volatiles, glycosidically bound fraction was isolated by eluting with 1 *l* of methanol. The methanol eluate was concentrated under reduced pressure to dryness and this dried material was re-

dissolved in 150 m*l* of 0.2 M acetate buffer (pH 5.5) containing phenyl-β-D-glucopyranoside (15.3 mg/150 m*l*, Fluka, Switzerland) as an internal standard. This buffered solution was divided into three equal portions.

Enzymatic hydrolysis

One portion of the glycosidically bound compounds dissolved in buffer solution was hydrolyzed by almond β-glucosidase (50 mg, 6 U/mg, Fluka, Switzerland) at 37°C for 36 hr. The liberated aglycones were isolated by liquid-liquid extraction with diethyl ether.

Acid hydrolysis

The second portion was acidified to pH 3.0 by adding 0.5 N HCl solution and refluxed for 2 hr. The liberated aglycones were isolated by liquid-liquid extraction with diethyl ether.

Simultaneous distillation-extraction (SDE)

The third portion was acidified to pH 3.0 by adding 0.5 N HCl solution and the solution was subjected to SDE with n-pentane-diethyl ether (1:1) for 2 hr using a modified Likens-Nickerson apparatus described by Schultz *et al.*[10] Each extract was dried by adding anhydrous sodium sulfate and concentrated to about 0.3 m*l* on a Vigreaux column (30cm)

Gas chromatography (GC) and gas chromatography-mass spectrometry (GC-MS)

GC analysis was carried out on a Hewlett-Packard (HP) 5880A GC, equipped with a FID and a 30m×0.25mm (ID) fused silica capillary column coated with Supelcowax 10. The oven temperature was programmed to increase linearly from 50°C to 230°C at 3°C/min and held at 230°C for 40 min. The injection port and detector temperatures were 250°C. The sample was injected in the split mode with a split ratio of 1:55. The carrier gas was nitrogen at a flow rate of 1.2 m*l*/min. Peak area was calculated by a HP 5880A integrator without consideration of FID response factors (calibration factors F = 1.00 for all components). The linear retention index was calculated by using n-paraffin homologues mixture (C_6-C_{26}) as references.[11]

GC-MS analysis was carried out on a Varian 3700 GC that was coupled by an open-split interface to a Finnigan MAT 212 MS with SS 188 data system and a 30m×0.25mm (ID) fused silica capillary column coated with Supelcowax 10. The column conditions were the same as described above. The MS operation conditions were as follows: carrier gas flow rate, 1.0 m*l*/min helium; ion source temperature, 250°C; ionization voltage, 70 eV; emission current, 1 mA.

Results and Discussion

In our previous study on the volatile components of apricot, we observed that the aroma concentrate of apricot isolated by SDE at native pH value of 3.1 contained high proportions of monoterpene alcohols, aromatic alcohols and naphthalene derivatives which were only minor components or did not exist in aroma concentrates isolated by SDE at neutral pH value of 7.0 or by headspace trapping method at native pH value of 3.1. To investigate glycosidically bound aroma components, a glycosidically bound fraction was isolated by Amberlite XAD-2 adsorption chromatography and this fraction was hydrolyzed by three different conditions.

Fig. 1 shows the gas chromatograms of aglycones liberated under three different hydrolysis conditions. Table 1 lists 40 components identified in the three aroma concentrates. Among these, 23 components were identified by comparing the retention time with authentic standards and mass spectra with those of published mass spectral data and the other 17 components were tentatively identified by comparing the mass spectral data only. Mass spectral data for unknown components and some previously unrecognized apricot components, most of which were liberated by enzyme hydrolysis are listed in Table 2.

Many of these bound volatile components were previously identified in apricot as free volatiles. However, it is very interesting that some volatile components, such as 2,6-dimethyl-3,7-octadiene-2,6-diol, 3,7-dimethyl-1,5-octadiene-3,7-diol, 4-hydroxyethylphenylacetate, (E)-and (Z)-2,6-dimethyl-2,7-octadiene-1,6-diol, 2,3-dihydrobenzofuran, 3-oxo-α-ionol, 3-hydroxy-7,8-dihydro-β-ionol, 3-oxo-7,8-dihydro-α-ionol and 3-hydroxy-β-ionone were identified only in the glycosidically bound fraction in this study.

As shown in Table 1, the major aglycones liberated by β-glucosidase were 3,7-dimethyl-1,5-

Table 1. Glycosidically bound volatile components identified in apricot

Peak no[a]	Compounds	I_k[b]	Peak area (%)			Evidence
			Enzyme[c]	Acid[d]	SDE[e]	
1	n-Pentanol	1274	–[f]	2.57	0.28	h)
2	n-Hexanol	1381	–	1.06	0.31	h)
3	cis-3-Hexen-1-ol	1394	–	+[g]	0.23	h)
4	Rose oxide	1412	–	+	0.21	i)
5	trans-2-Hexene-1-ol	1427	–	–	0.19	h)
6	Rose oxide (isomer)	1431	–	–	0.21	i)
7	Trimethyltetrahydronaphthalene	1469	–	0.98	0.23	i)
8	Linalool oxide	1510	–	1.27	0.37	h)
9	Linalool oxide (isomer)	1552	–	–	0.73	h)
10	Benzaldehyde	1570	0.66	0.44	1.14	h)
11	Linalool	1574	–	+	12.40	h)
12	Decahydronaphtol	1659	–	–	4.32	i)
13	γ-Butyrolactone	1666	–	+	1.73	h)
14	Butyric acid	1671	–	–	6.39	h)
15	Decahydronaphtol (isomer)	1689	–	2.37	0.93	i)
16	Decahydronaphtol (isomer)	1699	–	1.60	1.60	i)
17	Ocimenol	1715	–	0.73	0.23	h)
18	α-Terpineol	1740	–	0.74	6.66	h)
19	Nerol	1837	–	0.45	0.48	h)
20	β-Damascenone	1873	–	0.42	0.31	h)
21	Geraniol	1885	1.17	0.61	3.91	h)
22	Hexanoic acid	1902	–	1.83	2.09	h)
23	Benzyl alcohol	1929	11.04	13.71	2.02	h)
24	2-Phenylethyl alcohol	1945	0.29	16.94	27.76	h)
25	β-Ionone	1976	+	0.47	0.35	h)
26	2,6-Dimethyl-3,7-octadiene-2,6-diol	2026	0.38	–	0.54	i)
27	Phenol (ISTD, 5.10 mg)	–	6.83	12.49	7.28	–
28	γ-Nonalactone	2072	–	–	0.68	h)
29	3,7-Dimethyl-1,5-octadiene-3,7-diol	2091	39.89	21.75	3.42	i)
30	Eugenol	2150	2.09	7.59	1.60	h)
31	4-Hydroxyethylphenyl acetate	2223	0.75	1.18	0.39	i)
32	(E)-2,6-Dimethyl-2,7-octadiene-1,6-diol	2281	0.31	–	–	i)
33	(Z)-2,6-Dimethyl-2,7-octadiene-1,6-diol	2326	13.33	0.36	–	i)
34	Dihydroactinidiolide	2377	–	0.19	0.63	h)
35	2,3-Dihydrobenzofuran	2386	0.25	0.54	–	i)
36	Unknown	2392	0.48	–	–	–
37	Benzoic acid	2574	0.21	1.31	0.34	h)
38	3,4-Didehydro-β-ionol	2583	4.52	0.46	0.24	i)
39	Unknown	2591	0.21	0.34	+	–
40	Unknown	>2600	0.49	–	–	–
41	3-Oxo-α-ionol	>2600	0.87	1.84	+	i)
42	3-Hydroxy-7,8-dihydro-β-ionol	>2600	8.60	1.40	+	i)
43	3-Oxo-7,8-dihydro-α-ionol	>2600	0.71	0.24	+	i)
44	3-Hydroxy-β-ionone	>2600	2.55	0.34	+	i)
45	Unknown	>2600	0.16	–	–	–
46	Unknown	>2600	0.22	–	–	–
47	Unknown	>2600	0.43	–	–	–
48	Unknown	>2600	0.34	–	–	–
49	Unknown	>2600	0.38	–	–	–

[a] Peak numbers correspond to the numbers in Fig. 1.

[b] Kovats indices on Supelcowax 10

[c] Hydrolyzed by almond β-glucosidase at 37 °C for 36 hr

[d] Hydrolyzed by refluxing for 24 hr

[e] Hydrolyzed by means of simultaneous distillation-extraction with n-pentane-diethyl ether (1:1) for 2 hr

[f] Not detected

[g] Peak area less than 0.2%

[h] Comparison of retention time and mass spectrum with that of an authentic standard

[i] Tentatively identification by comparing a published mass spectral data: Williams et al.[6,7], Winterhalter and Schreier[15], Strauss et al.[16], Winterhalter[13], Fujimori et al.[19], Frolich et al.[20]

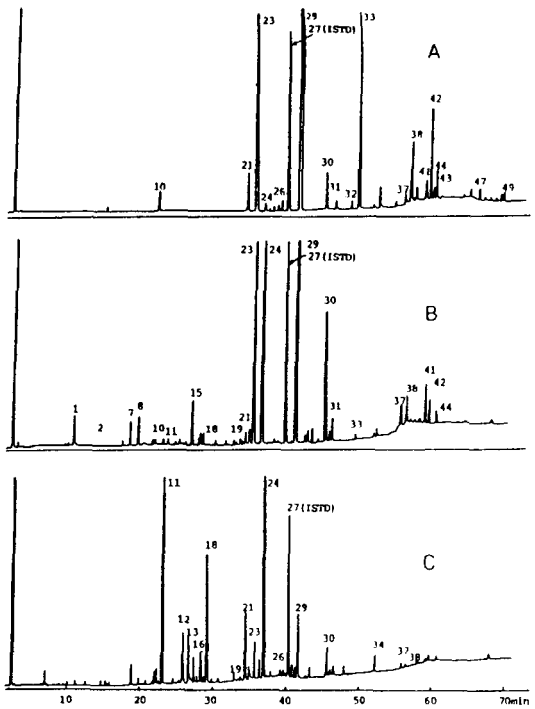

Fig. 1. Gas chromatograms of aglycones liberated from glycosidically bound fraction of apricot under the three different hydrolysis conditions
A: Enzyme hydrolysis, B: Acid hydrolysis at pH 3.0, C: Simultaneous distillation-extraction at pH 3.0
Peak numbers correspond to the numbers in Table 1.

octadiene-3,7-diol (39.89% of total volatiles), (Z)-2,6-dimethyl-2,7-octadiene-1,6-diol-(13.33%), benzyl alcohol (11.04%) and 3-hydroxy-7,8-dihydro-β-ionol (8.60%). The presence of bound monoterpene alcohols and aromatic alcohols, linalool, α-terpineol, nerol, geraniol, benzyl alcohol and 2-phenylethyl alcohol was reported in apricot by Salles *et al.*[8] In contrast to that, monoterpene alcohols, linalool, α-terpineol and nerol were not present in enzyme hydrolyzate in this study.

On the other hand, the major aglycones liberated by acid hydrolysis at pH 3.0 were n-pentanol (2.57%), decahydronaphtol (2.37%), benzyl alcohol (13.71%), 2-phenyl ethyl alcohol (16.94%), 3,7-dimethyl-1,5-octadiene-3,7-diol (21.75%) and eugenol (7.59%), while linalool, α-terpineol, nerol and geraniol were present in trace amounts. In contrast to the results of enzymatic and acid hydrolysis, when glycosidically bound fraction was subjected to simultaneous distillation-extraction at pH 3.0 that is typical for the natural pH value of apricot juice, the result showed a very different pattern of volatiles from those obtained by enzymatic or acid hydrolysis. Especially, linalool could be detected only in minute quantities or did not present in acidic or enzymatic hydrolyzate, but its concentration was enhanced considerably by SDE at pH 3.0. Similar behavior was observed for other monoterpene alcohols such as α-terpineol and geraniol.

It well established that, in fruits such as grapes[5,9], passion fruit[4,12,13], papaya[14], and apricot[8], at least some of the nonvolatile precursors of monoterpene alcohols are bound as glycosides. In addition, Williams *et al.*[6,7] and Engel and Tressl[4] have shown that the terpene alcohols were also produced by chemical rearrangement of nonvolatile diols and triols during heat treatment in weak acidic condition. The same authors[4] established that the precursors of volatile monoterpenoids were a series of water soluble hydroxylated linalool derivatives, which include 3,7-dimethyl-1-octen-3,7-diol, 3,7-dimethyl-1,5-octadiene-3 7-diol, 3,7-dimethyl-1,7-octadiene-3,6-diol and 3,7-dimethyl-1-octen-3,6,7-triol.

However, The increase in the concentation of monterpene alcohols observed during SDE at pH 3.0 in this study may be due to chemical rearrangement of hydroxylated linalool derivatives in weak acidic condition in addition to the liberation of bound volatile components.

In the study of the aroma components of apricot, the C_{13} norisoprenoids have aroused more interest than any other class of components. These components, through their intense floral aromas, were important components of the apricot flavor. In this study, a total of 7 norisoprenoids were identified as glycosidically bound volatiles and their structures are outlined in Fig. 2. Among them, 3,4-didehydro-β-ionol, 3-oxo-α-ionol, 3-hydroxy-7,8-dihydro-β-ionol, 3-oxo-7,8-dihydro-α-ional and 3-hydroxy-β-ionone were newly identified as glycosidically bound volatiles in apricot. It is very interesting that predominant norisoprenoid aglycones in this study are 3-oxygenated-α-and β-ionol derivatives.

3-Oxygenated α-or β-ionol derivatives exist predominately in passion fruit,[13] papaya,[14] quince,[15] and grapes.[16]

3-Oxo-α-ionol is known as the synthetic precursor of the isomeric megastigma-4,6,8-trien-3-ones,

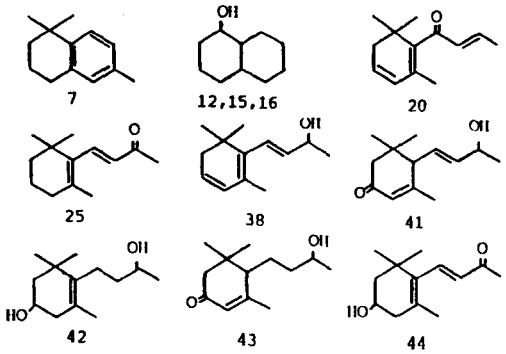

Fig. 2. Structures of norisoprenoid aglycones identified in glycosidically bound fraction of apricot
The numbers correspond to the numbers in Table 1.

key flavor components in burley tobacco.[17] However, in recent model study by Strauss *et al.*[16] this component was only slightly susceptible to weak acid catalyzed reaction and also 3-oxo-α-ionol is related to 3-oxo-7,8-dihydro-α-ionol, with saturated side chain, as well as 3-oxoretro-α-ionol, with rearranged double bond. 3-Oxoretro-α-ionol is known as the most probable precursor of 7-oxo-7,8-dihydroedulanes, which has been described to have an oriental tobacco-type aroma.[18] Unknown component (peak 47) in Table 2 seems to be identical with 3-oxoretro-α-ionol (base peak 149, M^+208) found in purple passion fruit by Winterhalter.[13] Unfortunately, we have neither authentic standard nor valuable information for the interpretation of structure.

On the other hand, in our previous study,[1] the three groups of naphthalene derivatives, trimethyltetrahydronaphthalene, trimethyldihydronaphthalene and decahydronaphtol, were found in aroma concentrate isolated by SDE at native pH value of 3.1. None of these above-mentioned naphthalene derivatives could be detected from enzyme hydrolyzate in this study. These results demonstrate that naphthalene derivatives originate from some unknown precursors which are not in glycosidic forms by the action of thermal cleavage under weak acidic condition. However, exact relationship between the precursors and these components in apricot is not clear yet.

Table 2. Mass spectral data of unidentified components

Peak no[a]	Mass spectral data (relative intensity)[b]
36	121(100), 95(51), 152(50), 139(49), 71(48), 94(44), 57(40)
39	164(100), 149(71), 152(69), 122(58), 151(33), 105(49), 220(4)
40	68(100), 84(80), 122(51), 67(51), 105(47), 55(46), 77(40)
45	137(100), 224(51), 89(49), 150(44), 73(27), 87(24), 59(20)
46	89(100), 73(93), 57(58), 87(53), 60(38), 55(38), 133(24)
47	149(100), 164(71), 89(56), 87(33), 73(33), 59(29), 208(6)
48	107(100), 89(38), 180(24), 73(24), 87(20), 59(16), 133(22)
49	89(100), 73(73), 87(55), 83(53), 59(37), 55(31), 164(5)

[a] Peak numbers correspond to the numbers in Fig. 1.
[b] The seven intense fragments

References

1. Kim, Y.H., Kwag, J.J., Kwon, Y.J. and Yang, K.K.: Influence of isolation method on the composition of apricot (*Prunus armeniaca* var. ansu Max.) flavor. *Korean J. Food Sci. Technol.*, **22**, 543 (1990)
2. Chairote, G., Rodriguez, F. and Crouzert, J.: Characterization of additional volatile flavor components of apricot. *J. Food Sci.*, **46**, 1898 (1981)
3. Takeoka, G.R., Flath, R.A., Mon, T.R., Teranishi, R. and Guntert, M.: Volatile constituents of apricot (*Prunus armeniaca*). *J. Agric. Food Chem.*, **38**, 471 (1990)
4. Engel, K. H. and Tressl, R.: Formation of aroma components from nonvolatile precursors in passion fruit. *J. Agric. Food Chem.*, **31**, 998 (1983)
5. Wilson, B., Strauss, C.R. and Williams, P.J.: Changes in free and glycosidically bound monoterpenes in developing Muscat grapes. *J. Agric. Food Chem.*, **32**, 919 (1984)
6. Williams, P.J., Strauss, C.R. and Wilson, B.: Hydroxylated linalool derivatives as precursors of volatile monoterpenes of Muscat grapes. *J. Agric. Food Chem.*, **28**, 776 (1980)
7. Williams, P.J., Strauss, C.R., Wilson, B. and Massy-Westropp, R.A.: Studies on the hydrolysis of *Vitis vinifera* monoterpene precursor compounds and model monoterpene β-D-glucosides rationalizing the monoterpene composition of grapes. *J. Agric. Food Chem.*, **30**, 1219 (1982)

8. Salles, C., Essaied, H., Chalier, P., Jallageas, J.C. and Crouzert, J.: Evidence and characterization of glycosidically bound volatile component in fruit. In *Bioflavor '87*, Schreier, P. (ed), Walter de Gruyter, p. 145 (1988)
9. Gunata, Y.Z.,Bayonove, C.L., Baumes, R.L. and Cordonnier, R.E.: The aroma of grapes I. Extraction and determination of free and glycosidically bound fractions of some grape aroma components. *J. Chromatogr.*, **331**, 83 (1985)
10. Schultz, T.H., Flath, R.A., Mon, T.R., Eggling, S.B. and Teranishi, R.: Isolation of volatile components from a model system *J. Agric. Food Chem.*, **25**, 446 (1977)
11. Kovats, E.: Gas-chromatographische charakterisierung organischer verbindngen teil 1; Retentionindices aliphatischer halogenide, alkohole, aldehyde und ketone. *Helv. Chim. Acta*, **41**, 1915 (1958)
12. Whitfield, F.B. and Last, J.H.: The flavour of the passion fruit a review. In *Progress in Essential Oil Research*, Brunke, E. J. (ed), Walter de Gruyter, Berlin, p.3 (1986)
13. Winterhalter, P.: Bound terpenoids in the juice of the purple passion fruit (*Passiflora edulis* Sims). *J. Agric Food Chem.*, **38**, 452 (1990)
14. Schwab, W., Mahr, C. and Schreier, P.; Studies on the enzyme hydrolysis of bound aroma components from *Carica papaya* fruit. *J. Agric. Food Chem.*, **37**, 1009 (1989)
15. Winterhalter, P. and Schreier, P.: Free and bound C_{13} norisoprenoid in quince (*Cydonia oblonga*, Mill.) fruit. *J. Agric. Food Chem.*, **36**, 1251 (1988)
16. Strauss, C.R., Wilson, B. and Williams, P.J.: 3-Oxo-α-ionol, vomifoliol and roseoside in *Vitis vinifera* fruit *Phytochemistry*, **26**, 1995 (1987)
17. Aasen, A.J., Kimland, B., Almquist, S.O. and Enzell, C.R. New tobacco constituents, Structure of five isomeric megastigmatrienones. *Acta Chem. Scand.* **26B**, 2573 (1972)
18. Winter, M., Schulte-Elte, K. H., Velluz, A., Limacher, J., Pickenhagen, W. and Ohloff, G.: Aromastoffer der roten passionfrucht, Zwei neue edulanderivate. *Helv. Chim. Acta*, **62**, 131 (1979)
19. Fujimori, T., Kasuga, R., Matsushita, H., Kaneko, H. and Noguchi, M.: Neutral aroma constituents in burley tobacco. *Agric. Biol. Chem.*, **40**, 303 (1976)
20. Frohlich, O., Duque, C. and Schreier, P.: Volatile constituents of curuba (*Passiflora mollissima*) fruit. *J. Agric. Food Chem.*, **37**, 421 (1989)

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살구에서 배당체의 형태로 존재하는 휘발성 성분

김영회 · 김근수 · 박준영 · 김용태
한국인삼연초연구소

비이온성 수지인 Amberlite XAD-2 흡착 및 메탄올 용출법에 의해 살구로부터 배당체분획을 얻은 다음 almond β-glucosidase 및 산가수분해(pH 3.0) 또는 pH 3.0에서 simultaneous distillation-extraction(SDE)에 의해 얻어진 가수분해물로부터 GC 및 GC-MS에 의해 40종의 휘발성 성분을 확인하였다. 3종의 가수분해물로부터 이미 배당체로서 살구에 존재하는 것으로 알려져 있는 linalool oxide, linalool, α-terpineol, nerol, geraniol, benzyl alcohol 및 2-phenylethyl alcohol 이외에도 2,6-dimethyl-3,7-octadiene-2,6-diol, 3,7-dimethyl-1,5-octadiene-3,7-diol, (E)- 및 (Z)-2,6-dimethyl-2,7-octadiene-1,6-diol, 3,4-dihydro-β-ionol, 3-oxo-α-ionol, 3-hydroxy-7,8-dihydroβ-ionol, 3-oxo-7,8-dihydro-α-ionol, 3-hydroxy-β-ionone, eugenol, 3-hydroxyethylphenyl acetate 및 2,3-dihydrobenzofuran이 살구에서 처음으로 확인되었다.