Aroma Components of *Vitis vinifera* L. cv. Chenin blanc Grapes and Their Changes During Maturation.

O. P. H. AUGUSTYN[1] AND A. RAPP[2]

[1]Oenological and Viticultural Research Institute, Private Bag X5026, Stellenbosch, 7600
[2]Bundesforschungsanstalt für Rebenzüchtung Geilweilerhof, 6741 Siebeldingen (über Landau/Pfalz), Federal Republic of Germany

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Samples of Chenin blanc grapes from 3 localities and at various stages of maturity were harvested over a period of 11 weeks. The freon extracted aroma components were subjected to gas chromatographic and coupled gas chromatographic-mass spectrometric analyses. No measureable amounts of terpenoid components were detected in any of the samples. Two isomers of 2,4-decadienal, a novel component in *Vitis vinifera* L. cultivars, occurred in relatively large concentrations in all samples. Origin of sample and degree of maturity have an insignificant effect on the concentration of selected aroma components.

Vitis vinifera L. cv. Chenin blanc is economically the most important wine grape cultivar grown in the Republic of South Africa and comprises approximately 26% of all vines.

Chenin blanc grapes have a neutral taste and this neutrality extends to the wine as far as cultivar character is concerned. Most young Chenin blanc wines exhibit a fruity-estery aroma caused by volatiles formed during fermentation. Occasionally some wines of this cultivar exhibit a highly desireable guava-like aroma. This aroma is, however, also found in other cultivars eg. *V. vinifera* L. cv. Colombar (Du Plessis & Augustyn, 1981) and disappears with ageing of the wine.

To date no information relating to the volatile aroma components of Chenin blanc grapes has been published. This study was initiated to determine the volatile aroma components of Chenin blanc grapes and to monitor concentration changes in selected components during berry maturation.

MATERIAL AND METHODS

Material: Chenin blanc grapes (1 kg per sample) were harvested weekly in the Klawer, Stellenbosch and Constantia areas, respectively corresponding to climatic regions V, III - IV and III (Winkler *et al.*, 1974), over a period of eleven weeks.

Samples were harvested from the same vines over the whole period. Each sample contained at least one cluster from each of 5 to 7 selected vines. Samples from Stellenbosch were transported from vineyard to laboratory in plastic bags and those from outside Stellenbosch in coolbags containing ice packs. All samples were processed on the day of harvest.

Sample preparation: Only healthy undamaged berries were used for analysis. Berries were individually cut from the clusters leaving a small portion of the stem attached to the berry. Two 250 gram samples were randomly selected from the mixed berries. The remainder of the sample was used for determination of total dissolved solids (expressed as degrees balling). Each berry sample was homogenised for 30 seconds in a blender with 200 mℓ of a 65% methanol /H_2O mixture previously purified by continuous extraction with freon 11 over 3 days (Rapp, Hastrich & Engel, 1976). Coarse particulate matter was removed from the homogenates by filtration through cheese cloth. The filtrates from each duplicate pair of samples were combined and two representative 250 ml subsamples obtained. Tetradecanol (2,5$\mu\ell$ of a 1% (w/v) solution in ethanol) was added to each sample and the mixture cooled before extraction.

Freon extraction: Double distilled freon 11 (20 mℓ in base of extractor and 40 ml in extracting flask) was used as extracting solvent in a continuous extractor similar to that described by Rapp *et al.* (1976). The formation of troublesome emulsions was prevented by placing a small wad of silanized glass wool in the base of the extractor. Samples were extracted for 20 hours while water at <4°C was circulated through the cooler and the extraction flask held at 30°C in a water bath. On completion of extraction the extraction flasks were glass stoppered and stored at -15°C until needed. All samples were analysed at the BFA für Rebenzüchtung Geilweilerhof, Germany. Before transport to Geilweilerhof extracts were partially concentrated and transferred to 5 ml glass ampules. Special care was taken to avoid any contact between extract and the heated glass surface on sealing the ampules.

Analysis of aroma extracts: Extracts were concentrated in drawn pear shaped flasks under partial reflux on a waterbath held at 28°C until nearly dry. Upon removal from the waterbath the flasks were glass stoppered and placed in a freezer for 10 - 20 minutes. Flasks were then handwarmed to reduce extract volume to 30 - 50$\mu\ell$ and cooled again before 2,5$\mu\ell$ samples were withdrawn for gas chromatographic analysis.

All analyses were performed on a Siemens L 402 gas chromatograph equipped with a *c.* 60 meter Reoplex 400 capillary column. The following parameters were applied; carrier gas and flowrate, H_2 at 1 mℓ per minute; split ratio 30 : 1; injector temperature 200°C; detector temperature 230°C; oven temperature programme 50°C to 150°C at 1,5°C per minute and chart speed 1 cm per minute. The column was held at 150°C until all peaks of interest had appeared. The average length of a run was about 2,5 hours with a further 1,5 - 2,0 hours at 150°C necessary to remove high boiling components from the column.

The relative quantities of different components in the same sample as well as specific components in different samples were compared by peak height measurements and normalization of these values on the height of the internal standard peak.

Coupled gas chromatography-mass spectrometry: All analyses were performed on a Finnigan 3200 instrument equipped with a 150 meter Reoplex 400 capillary column. Helium at 1 mℓ per minute was used as carrier gas and 1,0 - 1,5$\mu\ell$ highly concentrated sample (no split) was used per analysis.

Certain components were identified by comparison of spectra with those of authentic reference compounds. Most components were, however, identified by comparison of spectra obtained during this study with spectra of compounds previously identified at Geilweilerhof.

RESULTS AND DISCUSSION

Aroma components identified: The 29 volatile aroma components identified during this study are listed in Table 1.

TABLE 1
Aroma components identified in Chenin blanc grapes

Identity	Certainty of identification	Identity	Certainty of identification
Diacetyl	**	Ethyl decanoate	**
Hexanal	**	Ethyl benzoate	**
trans-2-Hexenal	***	Ethyl laurate	**
cis-3-Hexenal	**	Diethyl fumarate	*
trans-2, cis-4-Hexadienal	***		
trans-2, trans-4-Hexadienal	***	Pentanol	**
Heptanal	**	Pentenol	**
trans-2-Heptenal	**	Hexanol	***
trans-2, cis-4-Heptadienal	***	trans-2-Hexenol	**
trans-2, trans-4-Heptadienal	***	cis-3-Hexenol	***
Nonanal	**	Octanol	***
trans-2, cis-4-Decadienal	***	1-Octen-3-ol	**
trans-2, trans-4-Decadienal	***	Benzyl alcohol	***
Benzaldehyde	**	2-Phenyl ethanol	**
2-Phenylacetaldehyde	***		
		Styrene	***
Ethyl butyrate	**	Naphthalene	**
Methyl octanoate[1]	**	Phenol	**

[1] Probably formed from methanol and octanoic acid during processing.
*Spectrum compares well with published spectra
**Spectrum compares well with spectra of compounds previously identified at Geilweilerhof
***Spectrum matches that of authentic reference compound analysed under identical conditions. Retention time of reference compound and aroma component identical.

From Table 1 it is clear that no terpenoid components were found in the Chenin blanc samples. Although only a fraction of the volatile components present has been identified, all spectra were perused by persons experienced in the recognition of terpenoid mass spectra. No measurable amounts of any of the terpenoid compounds normally occurring in German wine grapes or wines (Rapp, Knipser & Engel, 1980) were present in these samples. This apparent lack of terpenoid compounds is surprising as almost all of the grape aroma components identified to date are found in all the vine varieties studied (Cordonnier & Bayonove, 1978). Differences in perceived aroma are, therefore, caused by quantitative differences in the concentration of specific components or groups of components (Rapp *et al.*, 1980), and not by the presence or absence of specific components or groups of components. Extraction of larger grape samples and the analysis of extremely concentrated extracts may lead to the detection of traces of terpenoid components in aroma extracts of this grape cultivar.

Wildenradt *et al.* (1975) isolated Chenin blanc grape leaf volatiles by steam distillation. Amongst the many components identified, a group of terpenes and terpene derivatives featured prominently. Preliminary work by Marais (1982) indicates that terpenoid components are also present in local Chenin blanc grape leaves (J. Marais, 1982 unpublished results). It would, therefore, seem as if some physiological block is present in Chenin blanc preventing the translocation of measurable amounts of terpenes or terpenoid precursors from the leaf to the berry.

Rapp *et al.* (1980) used variations in the concentrations of specific terpenes to differentiate between different grape varieties. Techniques are being developed to determine typical terpene profiles associated with desireable aroma by direct analysis of grape leaves (W. Knipser & A. Rapp, 1981 unpublished results). Such techniques would enable breeders to evaluate the aroma potential of grape seedlings at a very early stage. The present results indicate that caution be exercised in selections of this kind as all varieties with terpenoid components present in the leaves may not necessarily have the desired levels present in the fruit.

Few esters were identified during this study and fewer still were recognised amongst the spectra that have yet to be fully interpreted. This result supports the view of Schreier, Drawert & Junker (1976) that esters are perhaps not genuine grape constituents. This view should, however, be modified to read grapes of *V. vinifera* cultivars as Schreier & Paroschy (1980, 1981) recently identified a large number of esters in *V. riparia* M., *V. labrusca* L. and *V. labrusca* x *V. riparia* grapes.

The enzyme inhibition technique used in this work (Rapp, Hastrich & Engel, 1976) was designed to prevent any possible enzyme action on the terpenoid components of the grape aroma fraction. At the same time it also limits the formation of six carbon aldehydes and alcohols to acceptable concentrations. The amount of methanol added to the grapes before homogenisation is clearly inadequate for immediate and complete inhibition of the lipoxygenase activity on linoleic and linolenic acids (Schreier *et al.*, 1976). As a result of the inadequate enzyme inhibition the six carbon aldehydes and alcohols were prominent amongst the volatiles identified (Table 1).

Schreier *et al.* (1976) reported on the identification of alkadienals in the juice of various German grape varieties. The trans-2, cis-4- and trans-2, trans-4- isomers of hexa- and heptadienal as well as trans-2, trans-4- octadienal were found in trace amounts in enzyme inhibited grape juice. In analysis of grape juice prepared without enzyme inhibition these compounds were present in high concentrations. The concentration of the hexadienal isomers in particular increased dramatically.

No trace of nona- or decadienals was found in these experiments. Trans-2, trans-4-nonadienal and trans-2, trans-4-decadienal were, however, recently identified as minor components in the juice of *V. riparia* (W. Knipser, 1980 unpublished results; Schreier & Paroschy, 1980).

The decadienal isomers identified here (Table 1) are the first to be found in grapes of *V. vinifera*. If the six carbon aldehydes and alcohols are ignored these decadienals are amongst the major aroma compounds in Chenin blanc grape juice (Fig. 1). In direct contrast to the results of Schreier *et al.* (1976) the hexadienal isomers were present in trace amounts only.

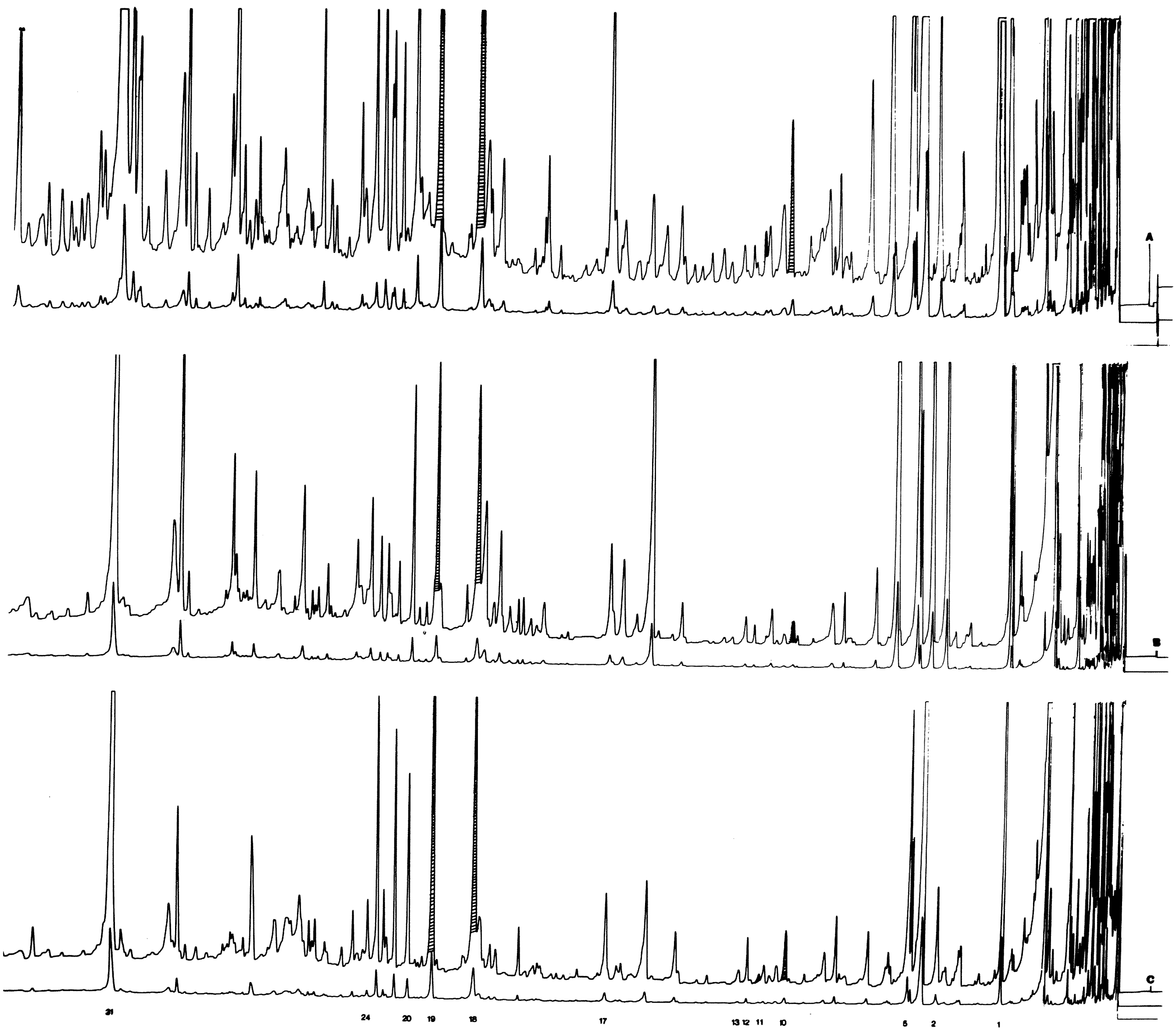

Fig. 1.
Chromatograms of freon extracts of Chenin blanc grapes at 3 different stages of maturity; A=19,5°B; B=13,1°B & C=8,2°B. Origin of samples: Constantia.

Examination of South African Morio Muscat and Weisser Riesling and German Morio Muscat, Weisser Riesling and Sylvaner extracts failed to indicate the presence of measurable amounts of decadienal. The 2,4-decadienal isomers therefore appear to be characteristic of Chenin blanc grape juice. Grosch, Schieberle & Laskawy (1981) recently studied the autoxidation of linolenic acid in model solutions. Their results indicate that the stability of unsaturated aldehydes in general, and 2,4-decadienal in particular, which are formed during this process, is enhanced by the addition of antioxidants to the medium. The possible relevance of these results to the presence of decadienal in Chenin blanc grape juice will be studied.

Perusal of all the available mass spectra indicated the presence of many aromatic compounds in the various extracts. This group of compounds included the well-known 2-phenyl ethanol, benzaldehyde, 2-phenylacetaldehyde and benzyl alcohol. Styrene was positively identified (Table 1) with the spectra of naphthalene and phenol, comparing well with known information. The latter two compounds are reported in grape juice for the first time. Both these components have, however, been identified in extracts of wine volatiles. Phenol has also been identified in cranberry, bilberry and citrus fruits, whilst naphthalene occurs in cranberry, bilberry, raspberry, bell pepper and potatoes amongst others (Van Straten, De Beauveser & Visscher, 1981). Aromatic compounds tentatively identified include toluene, o-xylene, a cresol and 4-ethyl phenol. The possibility that naphthalene is not a natural plant consituent but an artifact from the environment is mentioned by Kasting, Andersson & Von Sydow (1972). The same reservations could be expressed in connection with some of the other aromatic compounds mentioned above.

Traces of organo-silicone compounds were found in the extracts. As all contact with possible sources of artifacts was rigorously avoided, from the harvest to the final manipulations before analysis, the only obvious source of contamination is the silanized glass wool. Insertion of the glass wool into the extractor has proved invaluable in preventing emulsion formation. Turbid samples such as those analysed in this project, as well as red wines, are almost impossible to extract without this addition of glass wool. Methods to purify the glass wool before use are being tested at present.

Effect of grape maturity on the concentration of selected aroma components: Various monoterpenoid components play an important role in the character of the muscat group of grape cultivars (Williams, Strauss & Wilson, 1981). The development of these terpenoid components during grape maturation has been studied by various groups (Hardy, 1970; Bayonove & Cordonnier, 1971; Rapp & Hastrich, 1976). Distinct trends in terpenoid development were observed, with most of the terpenoid components increasing in concentration after the start of sugar accumulation. No such trend was observed for the aroma components studied here. Fig. 1 illustrates three of the typical chromatograms obtained during this study. Samples illustrated represent extracts of samples from Constantia harvested at A; 19,5°B; B: 13,1°B and C: 8,2°B. The shaded peaks in Fig. 1 represent trans-2, cis-4-heptadienal and trans-2, trans-4-heptadienal (peaks 10 and 11 respectively), as well as the same isomers of decadienal (peaks 18 and 19 respectively). Considering the concentration technique used here it is obviously very difficult to concentrate all samples to precisely the same volume. Differences in aroma compound concentration noticed in Fig. 1 are believed to be attributable to variations in the degree of concentration of the initial sample.

This lack of a specific trend in aroma compound concentration is again found on perusal of tables 2, 3 and 4. In these tables the relative concentrations of 13 selected aroma components, present in all the Chenin blanc samples from all three localities, are listed. From tables 2, 3 and 4 it also appears as if neither stage of maturity nor locality has any obvious effect on Chenin blanc aroma component concentration. This is a surprising observation because the considerable influence of climatic conditions and geographical situation on the aroma of eg. grapes and raspberries is well known (Rapp & Hastrich, 1978; Honkanen, Pyysalo & Hirvi, 1980).

CONCLUSION

The 2,4-alkadienals are well known grape aroma constituents but decadienal has not been identified in other *V. vinifera* cultivars. Consequently the alkadienal profile of Chenin blanc differs dramatically from that reported for other *V. vinifera* cultivars. The factors responsible for the high concentration of decadienal isomers as well as those responsible for the unexpectedly low concentration of hexadienal in Chenin blanc grapes, still have to be investigated.

Preliminary studies have indicated that terpenoid components are present in local Chenin blanc leaves. It is unknown at this stage whether the number of terpenoid

TABLE 2
Change in aroma component concentration during maturation of Chenin blanc grapes from Stellenbosch

Peak No	Component	Stage of maturity (°B)							
		13,2	14,5	17,2	19,6	19,7	20,4	22,6	23,3
1	trans-2-Hexenal	6484	19754	3295	8355	13401	7179	4033	635
2	Hexanol	242	553	115	193	274	114	73	365
5	cis-3-Hexenol	429	970	173	247	216	179	48	104
10	trans-2, cis-4-Heptadienal	46	20	10	20	30	19	10	8
11	trans-2, trans-4-Heptadienal	23	8	5	13	17	8	5	12
12	Benzaldehyde	23	18	3	8	6	–	–	2
13	Octanol	23	10	6	6	9	9	4	22
17	2-Phenylacetaldehyde	103	46	13	44	34	24	12	19
18	trans-2, cis-4-Decadienal	238	77	84	169	225	100	106	184
19	trans-2, trans-4-Decadienal	361	17	32	142	95	61	33	35
20	Benzyl alcohol	189	253	84	268	138	123	60	859
24	2-Phenyl ethanol	128	102	52	123	174	137	77	730

(millimeter peak height calculated on peak height of tetradecanol internal standard)

TABLE 3
Change in aroma component concentration during maturation of Chenin blanc grapes from Constantia

Peak	Compound	Stage of maturity (°B)										
		8,2	10,6	13,1	13,3	15,0	16,0	16,3	17,4	18,6	19,5	19,7
1	trans-2-Hexenal	405	1919	1998	5237	10644	17408	8583	8869	7922	12257	1601
2	Hexanol	59	158	355	212	817	473	92	60	69	173	151
5	cis-3-Hexenol	161	495	315	509	2254	1425	212	236	49	312	123
10	trans-2, cis-4-Heptadienal	31	72	12	24	59	112	21	20	26	72	67
11	trans-2, trans-4-Heptadienal	6	17	3	6	21	18	4	5	4	15	14
12	Benzaldehyde	28	19	10	13	27	35	3	3	3	15	6
13	Octanol	8	19	14	14	29	24	5	7	7	13	16
17	2-Phenylacetaldehyde	53	96	49	43	80	59	58	16	65	144	84
18	trans-2, cis-4-Decadienal	170	252	116	125	246	270	209	127	200	318	274
19	trans-2, trans-4-Decadienal	283	356	143	104	179	94	212	62	248	399	284
20	Benzyl alcohol	116	252	128	595	1010	873	86	133	88	255	741
24	2-Phenyl ethanol	37	105	69	230	414	338	44	62	46	104	394

(millimeter peak height calculated on peak height of tetradecanol internal standard)

TABLE 4
Change in aroma component concentration during maturation of Chenin blanc grapes from Klawer

Peak No.	Component	Stage of Maturity (°B)		
		16,2	17,7	19,5
1	trans-2-Hexenal	21682	5128	5429
2	Hexanol	950	197	405
5	cis-3-Hexenol	645	151	271
10	trans-2, cis-4-Heptadienal	73	20	31
11	trans-2, trans-4-Heptadienal	28	28	43
12	Benzaldehyde	35	3	6
13	Octanol	24	13	21
17	2-Phenylacetaldehyde	59	30	90
18	trans-2, cis-4-Decadienal	242	121	174
19	trans-2, trans-4-Decadienal	145	58	58
20	Benzyl alcohol	149	64	138
24	2-Phenyl ethanol	129	72	131

(millimeter peak height calculated on peak height of tetradecanol internal standard)

components, as well as their levels of concentration, are similar to those found elsewhere. It is possible that local climatic conditions result in a low terpenoid presence in the Chenin blanc grape leaf. Consequently the concentrations of the terpenoid components translocated to the berries could be below the detection limits used in this study. On the other hand the lack of measurable amounts of terpenoid components may be the result of some form of physiological block that inhibits the translocation of terpenoid components from leaf to berry.

LITERATURE CITED

BAYONOVE, C. & CORDONNIER, R., 1971. Recherches sur l'arôme du Muscat. 111. Étude de la fraction terpénique. *Ann. Technol. Agric.* **20,** 347 - 355.

CORDONNIER, R. & BAYONOVE, C., 1978. Les composantes variétales et préfermentaires de l'arôme des vins. *Parfums, Cosmetiques, Arômes* **24,** 67 - 77.

DU PLESSIS, C. S. & AUGUSTYN, O. P. H., 1981. Initial study of the guava aroma of Chenin blanc and Colombar wines. *S. Afr. J. Enol. Vitic.* **2,** 101 - 103.

GROSCH, W., SCHIEBERLE, P. & LASKAWY, G., 1981. Model experiments about the formation of volatile carbonyl compounds from fatty acid hydroperoxides. Flavour 81. 3rd Weurman Symposium, Proceedings of the International Conference, Munich 1981. Berlin: Walter de Gruyter. 433 - 448.

HARDY, P. J., 1970. Changes in volatiles of muscat grapes during ripening. *Phytochem.* **9,** 709 - 715.

HONKANEN, E., PYYSALO, T. & HIRVI, T., 1980. The aroma of Finnish wild raspberries, *Rubus idaeus* L. *Z. Lebens. Unters. Forsch.* **171,** 180 - 182.

KASTING, R., ANDERSSON, J & VON SYDOW, E., 1972. Volatile constituents in leaves of parsley. *Phytochem.* **11,** 2277 - 2282.

RAPP, A. & HASTRICH, H., 1976. Gaschromatographische Untersuchungen über die Aromastoffe von Weinbeeren 11. Möglichkeiten der Sortencharakterisierung. *Vitis* **15,** 183 - 192.

RAPP, A. & HASTRICH, H., 1978. Gaschromatographische Untersuchungen über die Aromastoffe von Weinbeeren 111. Die Bedeutung des Standortes für die Aromastoffzusammensetzung der Rebsorte Riesling. *Vitis* **17,** 288 - 298.

RAPP, A., HASTRICH, H. & ENGEL, L., 1976. Gaschromatographische Untersuchungen über die Aromastoffe von Weinbeeren 1. Anreicherung und kapillarchromatographische Auftrennung. *Vitis* **15,** 29 - 36.

RAPP, A., HASTRICH, H., ENGEL, L. & KNIPSER, W., 1980. Possibilities of characterizing wine quality and vine varieties by means of capillary chromatography. Flavor of foods and beverages. New York: Academic Press. 391 - 417.

RAPP, A., KNIPSER, W. & ENGEL, L., 1980. Identifizierung von 3,7-dimethyl-okta-1, 7-dien-3,6-diol im Trauben- und Weinaroma von Muskatsorten. *Vitis* **19,** 226 - 229.

SCHREIER, P., DRAWERT, F. & JUNKER, A., 1976. Identification of volatile constituents from grapes. *J. Agric. Food Chem.* **24,** 331 - 336.

SCHREIER, P. & PAROSCHY, J. H., 1980. Volatile components of wild grapes, *Vitis riparia* M. *Can. Inst. Food Sci. Technol. J.* **13,** 118 - 121.

SCHREIER, P. & PAROSCHY, J. H., 1981. Volatile constituents from Concord, Niagara (*Vitis labrusca* L.) and Elvira (*V. labrusca* L. x *V. riparia* M.) grapes. *Can. Inst. Food Sci. Technol. J.* **14,** 112 - 118.

VAN STRATEN, S., DE BEAUVESER, J. C. & VISSCHER, C. A., 1981. Volatile compounds in food, supplement 6, Zeist - the Netherlands: Central Institute for Nutrition and Food Research. 5, 6, 15, 25, 26.

WILDENRADT, H. L., CHRISTENSEN, E. N., STACKLER, B., CAPUTI, A., SLINKARD, K. & SCUTT, K., 1975. Volatile constituents of grape leaves. 1. *Vitis vinifera* variety Chenin blanc. *Am. J. Enol. Vitic.* **26,** 148 - 153.

WILLIAMS, P. J., STRAUSS, C. R. & WILSON, B., 1981. Classification of the monoterpenoid composition of muscat grapes. *Am. J. Enol. Vitic.* **32,** 230 - 235.

WINKLER, A. J., COOK, J. A., KLIWER, W. M. & LIDER, L. A., 1974. General Viticulture. Univ. Calif. Press, Berkeley.