3. CHEMICAL COMPOSITION OF ALCOHOLIC BEVERAGES, ADDITIVES AND CONTAMINANTS

3.1 General aspects

Ethanol and water are the main components of most alcoholic beverages, although in some very sweet liqueurs the sugar content can be higher than the ethanol content. Ethanol (CAS Reg. No. 64-17-5) is present in alcoholic beverages as a consequence of the fermentation of carbohydrates with yeast. It can also be manufactured from ethylene obtained from cracked petroleum hydrocarbons. The alcoholic beverage industry has generally agreed not to use synthetic ethanol manufactured from ethylene for the production of alcoholic beverages, due to the presence of impurities. In order to determine whether synthetic ethanol has been used to fortify products, the low ^{14}C content of synthetic ethanol, as compared to fermentation ethanol produced from carbohydrates, can be used as a marker in control analyses (McWeeny & Bates, 1980).

Some physical and chemical characteristics of anhydrous ethanol are as follows (Windholz, 1983):

Description: Clear, colourless liquid
Boiling-point: 78.5°C
Melting-point: −114.1°C
Density: d_4^{20} 0.789

It is widely used in the laboratory and in industry as a solvent for resins, fats and oils. It also finds use in the manufacture of denatured alcohol, in pharmaceuticals and cosmetics (lotions, perfumes), as a chemical intermediate and as a fuel, either alone or in mixtures with gasoline.

Beer, wine and spirits also contain volatile and nonvolatile flavour compounds. Although the term 'volatile compound' is rather diffuse, most of the compounds that occur in alcoholic beverages can be grouped according to whether they are distilled with alcohol and steam, or not. Volatile compounds include aliphatic carbonyl compounds, alcohols, monocarboxylic acids and their esters, nitrogen- and sulphur-containing compounds, hydrocarbons, terpenic compounds, and heterocyclic and aromatic compounds. Non-volatile extracts of alcoholic beverages comprise unfermented sugars, di- and tribasic carboxylic acids, colouring substances, tannic and polyphenolic substances, and inorganic salts. The flavour composition of alcoholic beverages has been described in detail in several reviews (Suomalainen & Nykänen, 1970; Amerine *et al.*, 1972; Nykänen & Suomalainen, 1983), and a recent review on the compounds occurring in distilled alcoholic beverages is available (ter Heide, 1986). The volatile compounds of alcoholic beverages and distillates generally originate from three sources: raw materials, fermentation and the wooden casks in which they are matured (Jouret & Puech, 1975).

During maturation, unpleasant flavours, probably caused by volatile sulphur compounds, disappear. Extensive investigations on the maturation of distillates in oak casks have shown that many compounds are liberated by alcohol from the walls of the casks (Jouret & Puech, 1975; Reazin, 1983; Nykänen, L., 1984; Nykänen *et al.*, 1984). Lignin plays an important role and is responsible for the occurrence of some aromatic aldehydes and phenolic compounds (Jouret & Puech, 1975; Nykänen *et al.*, 1984). These compounds are liberated from oak during the maturation process, together with monosaccharides (pentoses, quercitol), carboxylic acids and 'whisky lactone' (5-butyl-4-methyldihydro-2(3*H*)-furanone) (Nykänen, L., 1984; Nykänen *et al.*, 1984). The occurrence of aromatic compounds has been considered a manifestation of the degradation (oxidation) of oak lignin (Jouret & Puech, 1975).

The distillation procedure influences the occurrence and concentration of volatile flavour compounds in the distillate. Particularly in the manufacture of strong spirits, it is customary to improve the flavour of the distillate by stripping it of low-boiling and high-boiling compounds to a greater or lesser degree.

Certain flavoured alcoholic beverages may contain, in addition to the natural compounds of the beverages, added synthetic substances and ingredients isolated from herbs and spices. For instance, the flavour of vermouths, aperitifs, bitters, liqueurs and some flavoured vodkas is frequently composed of different essential oils or their mixtures; synthetic products and colouring substances, such as caramel (Ministry of Agriculture, Fisheries and Food, 1987), may also be added to improve the perceived flavour.

The exact compositions of many alcoholic beverages are trade secrets; however, there is extensive literature on the aroma components which are usually present at low levels, more than 1300 of which have been identified (Nykänen & Suomalainen, 1983). Information about nonaroma compounds is less extensive. A list of compounds identified in alcoholic beverages is given in Appendix 1 to this volume.

Definitions of the traditional terms for production processes and types of beverage are given by Lord (1979), Jackson (1982) and Johnson (1985). A useful glossary has been drawn up by Keller *et al.* (1982).

3.2 Compounds in beer

(*a*) *Carbonyl compounds*

Carbonyl compounds are among the most volatile substances in alcoholic beverages.

The levels of some aldehydes found in pasteurized and unpasteurized beers are given in Table 25. Acetaldehyde (see also IARC, 1985, 1987a) is the principal carbonyl compound in beer and has been found at similar ranges (0.1-16.4 mg/l) in US, German and Norwegian beers; levels as high as 37.2 mg/l were found in an unspecified beer (Nykänen & Suomalainen, 1983).

Table 25. Content (mg/l) of some aldehydes in pasteurized and unpasteurized beers[a]

Aldehyde	Pasteurized beer	Unpasteurized beer
Acetaldehyde	3.9–9.8	0.9–1.4
Pyruvaldehyde	0.119–0.293	0.040–0.046
Crotonaldehyde	0.002–0.036	$\leq$0.001
Isobutyraldehyde	0.011–0.024	0.007–0.008
Isovaleraldehyde	0.055–0.105	0.047–0.065
Octanal	0.007–0.017	0.005–0.006
Nonanal	0.005–0.014	0.004–0.006
Decanal	0.006–0.015	0.008–0.010
Dodecanal (Lauraldehyde)	0.004–0.016	0.002–0.006

[a] Reported by Nykänen & Suomalainen (1983)

Of the minor carbonyls identified in beer, formaldehyde (see IARC, 1982a, 1987a) has been found at a level of 0.17-0.28 mg/l in a Swiss beer (Steiner *et al.*, 1969); a fresh beer was reported to contain 0.009 mg/l formaldehyde and a stale beer, 0.002 mg/l (Lau & Lindsay, 1972). Some unsaturated aldehydes have also been identified in beer. Particular attention has been paid to the occurrence of *trans*-2-nonenal, which has been shown to be responsible for the oxidized or 'cardboard' flavour of stale beer, and to that of *trans,cis*-2,6-nonadienal, which gives rise to cucumber- or melon-like odours in beer (Visser & Lindsay, 1971; Wohleb *et al.*, 1972; Withycombe & Lindsay, 1973).

Beer also contains some aliphatic ketones. Postel *et al.* (1972a) found 0.3-1.7 mg/l acetone in German beer, and Tressl *et al.* (1978) determined the following ketones: 3-hydroxy-2-butanone (0.42 mg/l), 2-pentanone (0.02 mg/l), 3-hydroxy-2-pentanone (0.05 mg/l), 3-methyl-2-pentanone (0.06 mg/l), 4-methyl-2-pentanone (0.12 mg/l), 2-heptanone

(0.11 mg/l), 6-methyl-5-hepten-2-one (0.05 mg/l), 2-octanone (0.01 mg/l), 2-nonanone (0.03 mg/l) and 2-undecanone (0.001 mg/l).

The occurrence of 2,3-butanedione (diacetyl), 2,3-pentanedione and 3-hydroxy-2-butanone in beer has been investigated. The 2,3-butanedione content of beer generally varies from 0.01 to 0.2 mg/l (Nykänen & Suomalainen, 1983), but concentrations as high as 0.63 mg/l have been determined in British beer. Slightly smaller amounts (0.01-0.16 mg/l) of 2,3-pentanedione were found in British beers (White & Wainwright, 1975).

(*b*) *Alcohols*

The glycerol content of beer varies little — generally from 1100 to 2100 mg/l (Nykänen & Suomalainen, 1983), although Drawert *et al.* (1976) found average glycerol contents ranging up to 3170 mg/l in some German beers.

According to a review (Nykänen & Suomalainen, 1983), beers produced in different countries do not differ greatly in their content of aliphatic fusel alcohol (higher alcohols formed during yeast fermentation of carbohydrates), although the amounts vary to some extent between the different beer types because their formation depends on the yeast used and, in particular, on fermentation conditions. Thus, beers have been found to contain 4-60 mg/l 1-propanol, 6-72 mg/l 2-methyl-1-propanol (isobutanol), 3-41 mg/l 2-methyl-1-butanol and 35-52 mg/l 3-methyl-1-butanol.

Phenethyl alcohol (2-phenylethyl alcohol), an aromatic fusel alcohol, has a relatively strong rose-like odour, and therefore its determination in different beers has been a central subject of many studies; concentrations in beers vary from 4 to 102 mg/l. Benzyl alcohol occurs as a minor component in beer. Tyrosol and tryptophol, which are formed during fermentation, have been found in many beers, the tyrosol content varying from 1 to 29 mg/l and the tryptophol content from 0.2 to 12 mg/l (Nykänen & Suomalainen, 1983).

(*c*) *Volatile acids*

Most of the monocarboxylic acids — from formic acid to C_{18}-acids — are present in beer but, in general, as minor components. The acidity due to volatile compounds has been found to be greater in beer than in wort, indicating that the acids are formed during fermentation. Acetic acid is the most abundant, occurring at 12-155 mg/l in ales, 22-107 mg/l in lagers and 30-35 mg/l in stouts. The levels of other short-chain acids, up to hexanoic acid, vary from 0.3 to 3.4 mg/l. The principal volatile acids in beer have been reported to be hexanoic acid (1-25 mg/l), octanoic acid (2-15.4 mg/l) and decanoic acid (0.1-5.2 mg/l) (Nykänen & Suomalainen, 1983).

(*d*) *Hydroxy and oxo acids*

The occurrence of the L(+) and D(−) forms of lactic acid, which is a major hydroxy acid in beer, indicates bacterial activity during fermentation. Mändl *et al.* (1971a,b, 1973, 1975) found levels as high as 360 mg/l L-lactic acid in a French beer and 430 mg/l D-lactic acid in an Irish porter stout; most of the beers examined contained 2-40 mg/l L-lactic acid and

25-100 mg/l D-lactic acid. Beer also contains small amounts of other short-chain hydroxy acids (see Appendix 1). Some trihydroxy acids have also been detected: German, Austrian and American beers were reported to contain 5-9 mg/l 9,12,13-trihydroxy-10-*trans*-octadecenoic acid, 1-2.4 mg/l 9,10,13-trihydroxy-11-*trans*-octadecenoic acid, and 0.4-0.7 mg/l 9,10,13-trihydroxy-12-*trans*-octadecenoic acid (Nykänen & Suomalainen, 1983).

Of the oxo acids that occur in beer, pyruvic acid is present in fairly high amounts; Mändl *et al.* (1971b, 1973) determined concentrations of 53-89 mg/l in American beers and 17-138 mg/l in European beers.

(*e*) *Nonvolatile (fixed) acids*

The occurrence of certain nonvolatile (fixed) acids in beer is well established. The oxalic acid content is small, but it is of considerable interest because the formation of calcium oxalate may contribute to the appearance of hazes and sediments in beer. Bernstein and Khan (1973) reported 9-15 mg/l oxalic acid in ales and lager beers; Fournet and Montreuil (1975) found 2-11 mg/l in French beers; and German beers were found to contain 14-28 mg/l (Drawert *et al.*, 1975, 1976).

Succinic acid has been reported to occur at concentrations of 12-166 mg/l, malic acid at 0-213 mg/l and citric acid at 5-252 mg/l (Nykänen & Suomalainen, 1983). Numerous acids that occur at concentrations of only a few milligrams per litre are listed in Appendix 1.

(*f*) *Esters*

Beer contains a great number of esters of aliphatic fatty acids. Ethyl acetate is the principal one, occurring at 10-30 mg/l (with values up to 69 mg/l), while isopentyl acetate has been found at 1-7.8 mg/l. Of the high-boiling esters, ethyl hexanoate and ethyl octanoate occur at 0.1-0.5 mg/l and 0.1-1.5 mg/l, respectively, although some beers have been shown to contain 2.4-4 mg/l ethyl octanoate. The amount of phenethyl acetate varies between 0.03 and 1.5 mg/l (Nykänen & Suomalainen, 1983).

(*g*) *Nitrogen compounds*

(i) *Amines and amides*

Amines occur in beer due to the biochemical degradation of amino acids, which may begin during malting and then continues during fermentation. They are listed in Appendix 1. The following acetamides have been found in German dark beer: *N*,*N*-dimethylformamide, 0.015 mg/l; *N*,*N*-dimethylacetamide, 0.01 mg/l; *N*-methylacetamide, traces; *N*-ethylacetamide, 0.02 mg/l; *N*-(2-methylbutyl)acetamide, 0.01 mg/l; *N*-(3-methylbutyl)-acetamide, 0.025 mg/l; *N*-(2-phenylethyl)acetamide, 0.015 mg/l; and *N*-furfurylacetamide, 0.12 mg/l (Tressl *et al.*, 1977). The occurrence of several primary, secondary and tertiary amines in different beers is summarized in Table 26. Aminoacetophenone may be responsible in part for the characteristic odour of beers.

Table 26. Content (mg/l) of some amines in beer[a]

Amine	Amount (range)
Methylamine	0.02–0.32
Ethylamine	0.11–2.12
n-Propylamine	traces–0.17
Isobutylamine	0.004–0.22
n-Butylamine	traces–0.07
Isopentylamine (isoamylamine)	traces–0.14
n-Hexylamine	0.005–0.28
N,*N*-Dimethyl-*n*-butylamine	traces–0.16
2-Aminoacetophenone[b]	0.01
Dimethylamine	0.37–0.78
Trimethylamine	0.03–0.06
para-(2-Aminoethyl) phenol (tyramine)	6.5–11.22

[a] From Palamand *et al.* (1969); Sen (1969; Canadian beer); Palamand *et al.* (1971); Slaughter & Uvgard (1971; English beer); Koike *et al.* (1972; Japanese beer); Tressl *et al.* (1977; German beer)

(ii) N-*Heterocyclic compounds*

N-Heterocyclic compounds present in malt can be detected at low levels in some beers. The amounts of pyrazines in dark Bavarian beer are given in Table 27. Tressl *et al.* (1977) assumed that the pyrroles occurring in some beers are responsible for the smoky odour resembling that of pastry and bread. A number of compounds with such an odour were found, four of which were identified as nicotinic acid esters. The pyrroles and thiazoles found in dark Bavarian beer are listed in Table 28.

(iii) *Histamine and other nonvolatile* N-*heterocyclic compounds*

Granerus *et al.* (1969) showed that the level of histamine was 0.03-0.05 mg/l in Swedish beer and 0.03-0.15 mg/l in Danish beer. Chen and Van Gheluwe (1979) found means of 0.22 mg/l in Canadian ale, 0.20 mg/l in Canadian lager, 0.38 mg/l in Canadian malt liquor, 0.41 mg/l in Canadian porter, 0.13 mg/l in Canadian light beer, 0.13 mg/l in American lager and 0.20 mg/l in European beers. A high histamine content, 1.9 mg/l, was found in a Belgian Gueuze produced by 'spontaneous' fermentation with microorganisms other than brewer's yeast.

The purine and pyrimidine contents of beer have been investigated in several studies (Saha *et al.*, 1971; Buday *et al.*, 1972; Charalambous *et al.*, 1974; Kieninger *et al.*, 1976; Ziegler & Piendl, 1976; Mändl *et al.*, 1979; Boeck & Kieninger, 1979). In most beers the amounts of uracil, cytosine, hypoxanthine, xanthine, adenine, guanine, thymine, thymidine, adenosine and inosine were found to range from 0.1 to 40 mg/l, whereas higher amounts of guanosine (30-160 mg/l), cytidine (18-70 mg/l) and uridine (15-200 mg/l) were detected.

Table 27. Content (mg/l) of pyrazines in dark Bavarian beer[a]

Pyrazine	Amount
Methyl-	0.07
2,5-Dimethyl-	0.11
2,6-Dimethyl-	0.035
2,3-Dimethyl-	0.015
Trimethyl-	0.02
Tetramethyl-	traces
Ethyl-	0.01
2-Ethyl-6-methyl-	0.035
2-Ethyl-5-methyl-	traces
2-Ethyl-3-methyl-	traces
2-Ethyl-3,5-dimethyl-	0.01
2-Ethyl-3,6-dimethyl-	0.02
2-Ethyl-5,6-dimethyl	traces
6,7-Dihydro-5*H*-cyclopenta-	0.01
5-Methyl-6,7-dihydro-5*H*-cyclopenta-	0.015
2-Methyl-6,7-dihydro-5*H*-cyclopenta-	0.01
5-Methylcyclopenta-	traces
2-Furyl-	0.025
2-(2′-Furyl)methyl-	0.01
2-(2′-Furyl)dimethyl-	traces

[a] From Tressl *et al.* (1977)

Table 28. Content (mg/l) of pyrroles, thiazoles and some other cyclic compounds determined in dark Bavarian beer[a]

Compound	Amount
Pyrrole	traces
2-Methylpyrrole	1.8
2-Formylpyrrole	0.03
2-Acetylpyrrole	1.4
2-Acetyl-5-methylpyrrole	0.01
2-Formyl-5-methylpyrrole	0.11
1-Acetylpyrrole	traces
1-Furfurylpyrrole	0.01
2-Pyrrolidone	0.01
1-Methyl-2-pyrrolidone	traces
Indole	traces
2-Acetylthiazole	traces
4-Methyl-5-hydroxyethylthiazole	traces
Methyl nicotinate	traces
Ethyl nicotinate	1.5
3-Methylbutyl nicotinate	traces
2-Phenethyl nicotinate	traces

[a] From Tressl *et al.* (1977)

(*h*) *Aromatic compounds*

(i) *Phenols*

Special attention has been paid to the occurrence of phenols in beer due to their potential influence on the flavour (Nykänen & Suomalainen, 1983). Wackerbauer *et al.* (1977) investigated the sources of the phenolic flavour of beer and found cresol (0.012 mg/l), 4-vinylphenol (0.17 mg/l), 2-methoxy-4-vinylphenol (4-vinylguaiacol; 0.074 mg/l) and 4-hydroxybenzaldehyde (0.018 mg/l) in a flawed beer. Many different phenols have been found in beers (Tressl *et al.*, 1975a, 1976; see also Appendix 1).

(ii) *Aromatic acids*

Beer contains numerous aromatic acids (Appendix 1); their occurrence in beer is summarized in Table 29.

Table 29. Content (mg/l) of aromatic acids in beer[a]

Acid	Amount
Benzoic acid	0.45
Phenyllactic acid	1.2
Salicylic acid (2-Hydroxybenzoic acid)	0.02
4-Hydroxybenzoic acid	0.13
Protocatechuic + vanillic acid	2.4
Phthalic acid	0.02
Phenylacetic acid	0.93
4-Hydroxyphenylacetic acid	0.04
Ferulic acid (4-Hydroxy-3-methoxycinnamic acid)	
,*cis*-	1.1
,*trans*-	4.6
Cinnamic acid	
,*cis*-	<0.01
,*trans*-	0.50
para-Coumaric acid (4-Hydroxycinnamic acid)	
,*cis*-	0.02
,*trans*-	1.9
Phenylpropionic acid	0.01
4-Hydroxyphenylpropionic acid	0.02

[a] From Tressl *et al.* (1975b)

3.3 Compounds in wine

(*a*) *Carbonyl compounds*

Acetaldehyde constitutes more than 90% of the total aldehyde content of wines, occurring at 50-100 mg/l (Nykänen & Suomalainen, 1983). Wucherpfennig and Semmler (1972, 1973) found 74-118 mg/l acetaldehyde in wines produced from different grapes in various vineyards in different countries, and Postel *et al.* (1972b) found 11-160 mg/l in German 'Spätlesen', 'Auslesen' and 'Beerenauslesen' white wines and in red wines; white and red wines had similar aldehyde contents. The aldehyde content is, however, low, and this may be explained by the fact that the sulphur dioxide added to wine reacts with aldehydes to form α-hydroxysulphonic acids, which reduce the free aldehyde content. Furthermore, aldehydes can be chemically bound to ethanol and higher alcohols as acetals.

Minor amounts of other aliphatic aldehydes and ketones are also present in wine (Appendix 1). Baumes *et al.* (1986) found 3-hydroxy-2-butanone (0.002-0.3 mg/l) and 3-hydroxy-2-pentanone in French white and red wines. The volatile flavour of Chardonnay and Riesling wines has been reported to include minor amounts of 2-methylbutanal, 3-methylbutanal, hexanal and 2-heptanone (Simpson & Miller, 1983, 1984). Benzaldehyde has been found in detectable amounts (0.002-0.504 mg/l) in different French wines (Baumes *et al.*, 1986), in Chardonnay and Riesling wines (Simpson & Miller, 1983, 1984) and in Pinot Noir wine (Brander *et al.*, 1980).

Two vicinal diketones, 2,3-butanedione and 2,3-pentanedione, may be of importance to the flavour nuances, although they occur at low levels. 2,3-Butanedione has been found in white wines at 0.05-3.4 mg/l and in red wines at 0.02-5.4 mg/l, whereas lower values have been reported for 2,3-pentanedione (0.007-0.4 mg/l in white wines and 0.01-0.88 mg/l in red wines; Leppänen *et al.*, 1979; Nykänen & Suomalainen, 1983).

(*b*) *Acetals*

In contrast to beers, wines contain acetal (1,1-diethoxyethane) as a major component of the volatiles. It is generally assumed that the reaction of acetaldehyde with ethanol to yield acetal may 'round' the smell of wines, which is of great importance. In the French wines investigated by Baumes *et al.* (1986), the total amounts of acetal and 2,4-dimethyl-1,3-dioxane were reported to vary from 0.18 to 9.3 mg/l in white wines and from 0.09 to 0.52 mg/l in red wines. Other acetals found at low concentrations were 2,4,5-trimethyl-1,3-dioxolane (previously identified by Brander *et al.* (1980) in Pinot Noir wine), 1,3-dimethyl-4-ethyl-1,3-dioxolane, 1-ethoxy-1-(2′-methylpropoxy)ethane, 1-ethoxy-1-(3′-methylbutoxy)ethane, 1-(3′-methylbutoxy)-1-(2′-methylbutoxy)ethane, 1,1-di-(3′-methylbutoxy)-ethane, 1-ethoxy-1-(2′-phenethoxy)ethane, 1-(3′-methylbutoxy)-1-(2′-phenethoxy)ethane, *cis*-2-methyl-4-hydroxymethyl-1,3-dioxolane and *trans*-2-methyl-4-hydroxymethyl-1,3-dioxolane (Baumes *et al.*, 1986).

An ether possibly related to the acetals, 3-ethoxypropanol, has been identifed in Pinot Noir wine (Brander *et al.*, 1980).

(*c*) *Alcohols*

(i) *Di- and trihydric alcohols*

Apart from ethanol, glycerol and 2,3-butanediol are the principal alcohols in wine. The glycerol content has been reported to range between 2000 and 36 000 mg/l in sound wines (Nykänen & Suomalainen, 1983). The contents of numerous European and American wines have been found to vary from 400 to 1100 mg/l; exceptionally high amounts of 2,3-butanediol were found in a Romanian 'Trockenbeerenauslese' (2700 mg/l) and in an 'Edelauslese' (3300 mg/l; Patschky, 1973).

(ii) *Fusel alcohols and long-chain alcohols*

Numerous investigations on the volatile components of different wines have shown that higher alcohols are ubiquitous. White and red wines produced in various countries contain 1-propanol (11-125 mg/l), 2-methyl-1-propanol (15-174 mg/l), 2-methyl-1-butanol (12-311 mg/l) and 3-methyl-1-butanol (isopentanol; 49-180 mg/l). In addition, wines contain 5-138 mg/l phenethyl alcohol. The occurrence of the aromatic alcohols, tyrosol (4-hydroxyphenethyl alcohol) and tryptophol (3-indolethanol), which are formed by biochemical mechanisms similar to those proposed for the formation of phenethyl alcohol and aliphatic fusel alcohols, has also been established; white and red wines have been reported to contain 5-45 mg/l tyrosol and 0.3-3.1 mg/l tryptophol (Nykänen & Suomalainen, 1983).

A number of long-chain alcohols, such as 1-pentanol, 4-methyl-1-pentanol, 3-methyl-1-pentanol, *Z*-2-penten-1-ol, 1-hexanol, the *E*- and *Z*-isomers of 2-hexen-1-ol and 3-hexen-1-ol, 1-heptanol, 1-octanol, 1-nonanol and 1-decanol, have been identified in wines (Brander *et al.*, 1980; Nykänen & Suomalainen, 1983; Simpson & Miller, 1983, 1984). Of these, 3-methyl-1-pentanol, 1-hexanol and *E*-3-hexen-1-ol seem to be fairly important components (Baumes *et al.*, 1986).

(*d*) *Volatile acids*

Acetic acid is the most abundant of the volatile acidic constituents of wine, although yeast is known to produce only minor amounts of acetic acid in fermentation under anaerobic conditions. Any substantial increase in the volatile acidity in wines thus seems to be due to the activity of spoilage microorganisms. Acetic acid bacteria can oxidize ethanol, first forming acetaldehyde, followed by oxidation of aldehyde to acetic acid, thus restricting volatile acidity to, for example, the permissible values of under 900 mg/l in French wines and in German white wines, under 1200 mg/l in German red wines, under 1100 mg/l in Californian white table wines and under 1200 mg/l in Californian red wines (Nykänen & Suomalainen, 1983). The volatile acidity of German wines, for instance, has been reported to be about 300 mg/l (Schmitt, 1972).

(*e*) *Hydroxy acids*

Wines contain fairly large amounts of L(+)- and D(−)-lactic acid. The total concentration of lactic acid in French wines that have undergone malolactic fermentation has been found to vary from 900 to 2600 mg/l, and total lactic acid contents of 100-5600 mg/l and 200-3100 mg/l have been reported. Other monohydroxy acids occur as minor components: 2,3-dihydroxy-2-methylbutyric acid has been found at 60-523 mg/l in a large number of wines, at 34-205 mg/l in Italian wines and at 70-550 mg/l in Bordeaux wines (Nykänen & Suomalainen, 1983).

(*f*) *'Fixed' acids*

The acidity of wines depends mainly on the presence of nonvolatile acids from the grapes. Tartaric, malic, citric and succinic acids are usually the most abundant and are of great importance, not only because they regulate the acidity of the wine but also because their acidity protects sound wine from spoilage and increases the stability of coloured substances. Their total amount is determined by a titrimetric method (Nykänen & Suomalainen, 1983).

de Smedt *et al.* (1981) found malic acid (3380 mg/l), tartaric acid (2120 mg/l), succinic acid (500 mg/l), citric acid (270 mg/l) and phosphoric acid (240 mg/l). Of the minor components, they determined shikimic acid (70 mg/l) and citramalic acid (20 mg/l) quantitatively.

(*g*) *Esters*

The largest group of flavour compounds in wines consists of esters of the aliphatic monocarboxylic acids. Ethyl acetate and many of the long-chain esters in wine are formed by yeast principally by enzymic reactions during fermentation and not in chemical reactions between ethanol and corresponding acids (Nykänen & Suomalainen, 1983). The acid-catalysed esterification and hydrolysis of the esters, however, may be of importance during prolonged ageing, even though the reactions proceed slowly and equilibrium concentrations are reached only after a long time.

Ethyl acetate is the principal ester component. Postel *et al.* (1972b) found 44-122 mg/l ethyl acetate in white wines and 78-257 mg/l in red wines. Higher levels were found in sound white wines produced in different countries (11-261 mg/l), and similar concentrations were found in red wines (22-232 mg/l; Shinohara & Watanabe, 1976). Late harvest wines, such as 'Spätlese', 'Auslese' and 'Beerenauslese', were found to contain 52-99 mg/l, 92-108 mg/l and 191-285 mg/l ethyl acetate, respectively (Postel *et al.*, 1972b).

Ethyl esters of short-chain acids as well as acetate esters of fusel alcohols are frequently found in white and red wines. The ethyl esters of decanoic and dodecanoic acids are usually the longest chain esters found (Nykänen & Nykänen, 1977; Nykänen *et al.*, 1977).

In addition, a number of esters originating in grapes have been identified in wines, such as methyl anthranilate, of which the odour has been reported to be characteristic of the grape variety *Vitis labrusca* and may be responsible for the 'foxy' ('rosé') character of some

American wines (Nelson *et al.*, 1978; Nykänen, 1986). The cyclic ester, 2,6,6-trimethyl-2-vinyl-4-acetoxytetrahydropyran, together with a tetrahydrofuran derivative, linalool oxide, potentially contributes to flavour in wine (Schreier & Drawert, 1974).

A number of esters of di- and tricarboxylic acids have also been identified in wine, of which diethyl succinate is ubiquitous. Baumes *et al.* (1986) found 2,3-butanediol monoacetate, methylethyl succinate, diethyl malonate, diethyl malate, diethyl 2-ketoglutarate and diethyl 2-hydroxyglutarate in French wines. DiStefano (1983) identified ethyl esters of 2-hydroxyglutaric acid and 2-hydroxyglutaric acid γ-lactone in Italian wine. Diethyl succinate has been found in Australian Chardonnay wines and ethyl 3-methylbutyl succinate in Riesling wines (Simpson & Miller, 1983, 1984). The occurrence of mono- and diethyl esters of tartaric acid in wine has been confirmed in several studies (Shimizu & Watanabe, 1978; Sponholz, 1979; Edwards *et al.*, 1985).

(*h*) *Nitrogen compounds*

(i) *Amines and some* N-*heterocyclic compounds*

Amines are probably formed mainly by bacterial decarboxylation of amino acids, but small amounts may also occur as the result of enzymic reactions of yeast. Schreier *et al.* (1975) showed that the yeast *Saccharomyces cerevisiae* can produce the corresponding *N*-acetylamines from 2-methylbutylamine, 3-methylbutylamine and 2-phenethylamine in a fermentation solution. Consequently, some amides detected in wine may be formed by yeast from amines during wine fermentation. Desser and Bandion (1985) showed that certain technological treatments and the storage of bottled wine may decrease the concentrations of biogenic amines such as 1,3-propanediamine, putrescine (1,4-butanediamine), histamine (2-(4-imidazolyl)ethylamine), cadaverine (1,5-pentanediamine), spermidine (*N*-(3-aminopropyl)-1,4-butanediamine) and spermine (*N*,*N*′-bis(3-aminopropyl)-1,4-butanediamine) in wine. Puputti and Suomalainen (1969) determined the concentrations of a number of volatile and nonvolatile compounds in white and red wines (Table 30).

The amounts of amines in different wines vary widely. Spettoli (1971) found ethylamine (traces-0.36 mg/l), isobutylamine (traces-0.7 mg/l), isopentylamine (isoamylamine; 0.04-0.7 mg/l), hexylamine (0.1-0.9 mg/l), ethanolamine (0.05-0.9 mg/l) and *para*-(2-aminoethyl)phenol (tyramine; 0.06-0.7 mg/l) in Italian white and red wines. The diamines, 1,4-butanediamine (putrescine), 1,5-pentanediamine (cadaverine) and tyramine are metabolic products of bacteria; diamines are found in greater amounts in red wines. Concentrations of 1,4-butanediamine have been reported to reach 24 mg/l in Swiss white wines and 45 mg/l in Swiss red wines, whereas the concentrations of 1,5-pentanediamine reached 2 mg/l in white wines and 4 mg/l in red wines (Mayer & Pause, 1973). Woidich *et al.* (1980) found the amines reported in Table 31 in several Austrian wines.

Other amines and *N*-heterocyclic compounds have been identified in wine (Table 32). Bosin *et al.* (1986) determined 1,2,3,4-tetrahydro-β-carboline-3-carboxylic acid and 1-methyl-1,2,3,4-tetrahydro-β-carboline-3-carboxylic acid at 0.8-1.7 and 1.3-9.1 mg/l, respectively. Some of the pyrroles, thiazoles and piperazines that occur in other beverages have also been identified in wine; these are 1-ethyl-2-formylpyrrole, *N*-methylpyrrole,

Table 30. Content (mg/l) of amines in white and red wines[a]

Amine	Burgundy red wine	Bordeaux white wine	Riesling white wine
Volatile amines			
Ethylamine	1.7	0.5	0.7
Isopropylamine	0.05	0.02	0.04
Isobutylamine	0.07	0.02	0.05
n-Butylamine	0.01	traces	traces
Isopentylamine (Isoamylamine)	2.2	1.0	4.0
Pentylamine (n-Amylamine)	0.01	traces	traces
Hexylamine	0.7	0.4	0.4
Nonvolatile amines			
Ethanolamine	0.3	0.08	0.08
1,4-Butanediamine (Putrescine)	0.3	traces	traces
para-(2-Aminoethyl)phenol (Tyramine)	1.0	0.1	0.2
Histamine	0.5	0.04	traces

[a] From Puputti & Suomalainen (1969)

Table 31. Content (mg/l) of some biogenic amines in Austrian wines[a]

Amine	Red wine	White wine
1,4-Butanediamine	0.51-24.2	<0.05-1.7
1,5-Pentanediamine	<0.05-3.0	<0.05-2.8
Histamine	<0.1-13.6	<0.1-3.5
4-Azaheptamethylenediamine	0.05-1.3	<0.05-2.7
Spermine	<0.1-0.4	<0.1-0.8
para-(2-Aminoethyl)phenol	<0.1-8.1	<0.1-2.7
Phenethylamine	<0.1-5.1	<0.1-3.8

[a] From Woidich et al. (1980)

N-ethylpyrrole, *N*-propylpyrrole, benzothiazole, *N*-methylpiperazine, 2-methylpiperazine, *trans*-2,5-dimethylpiperazine and *N*,*N*-dimethylpiperazine (Ough, 1984). The occurrence of 2-methoxypyrazines in wine has been confirmed (Heymann *et al.*, 1986). Serotonin [3-(2-aminoethyl)-1*H*-indol-5-ol] and octopamine [(4-hydroxyphenyl)ethanolamine] have been determined by a high-pressure liquid chromatographic method among the nonvolatile compounds in wine (Lehtonen, 1986).

Table 32. Content (mg/l) of amines and N-heterocyclic compounds in wine[a]

Compound	Content
Dimethylamine	0.1-0.7
Trimethylamine	0.01-0.07
Diethylamine	<0.001
Triethylamine	0.01-0.02
Pyrrolidine	traces-0.06
Piperidine	traces
Morpholine	0.07
2-Pyrrolidone	0.01
1-Methyl-2-pyrrolidone	traces
1-Pyrroline	0.01-0.5
Indole	traces
1-Methyl-1,2,3,4-tetrahydro-β-carboline	0.001-0.084
4,5-Dimethyl-1,3-dioxolan-2-propanamine	traces
2-Aminomethyl benzoate	traces
6-Hydroxy-1-methyl-1,2,3,4-tetrahydro-β-carboline	0-0.0002
Pyridine	traces
Quinoline	traces
N-Methylpiperidine	traces

[a] From Ough (1984)

(ii) *Amides*

Numerous acetamides have been identified in wines (Ough, 1984). These include *N*,*N*-dimethylformamide, *N*-3-methylbutylacetamide, *N*-*n*-pentylacetamide, *N*-ethylacetamide, *N*-*n*-hexylacetamide, *N*-*n*-propylacetamide, *N*-cyclohexylacetamide, *N*-isopropylacetamide, *N*-(3-(methylthio)propylacetamide, *N*-*n*-butylacetamide, *N*-2-phenethylacetamide, *N*-isobutylacetamide, *N*-*tert*-butylacetamide, *N*-piperidylacetamide, *N*-2-methylbutylacetamide and *N*,*N*-diethylacetamide. Only a few quantitative results have been reported; for example, *N*-2-methylbutylacetamide at 0.002-0.02 mg/l and *N*-3-methylbutylacetamide at 0.002 mg/l.

(*i*) *Terpenic compounds*

Wine contains numerous terpene hydrocarbons, terpene aldehydes and ketones, terpene alcohols, esters of terpene alcohols, and their oxidation products (see Appendix 1). The quantities of individual terpenes vary widely according to the wine. Terpene composition depends, in part, on grape varieties; varieties of white *Vitis vinifera* grapes and wines have been classified according to their terpene profiles (Schreier *et al.*, 1976a,b; Rapp *et al.*, 1984), although whether this can be generally used for classifying different grape varieties or wines produced from these grapes is not established. Since free and glycosidic derivatives of monoterpenes are uniformly distributed among the skin, pulp and juice (Wilson *et al.*, 1984; Williams *et al.*, 1985; Wilson, B. *et al.*, 1986), a fairly large proportion of terpenes are present bound to glycosides in young wine; however, when the glycosidic compounds are hydrolysed during ageing, the terpene profile changes (Rapp *et al.*, 1985).

The amounts of terpene compounds in wine have not been reported, but it has been suggested that they contribute markedly to the specific characteristics of wine flavour (Williams, 1982; Rapp *et al.*, 1984; Schreier, 1984; Strauss *et al.*, 1984; Williams *et al.*, 1985; Rapp & Mandery, 1986).

(*j*) *Phenolic compounds*

Besides phenolic alcohols, aldehydes and acids, red wines contain small amounts of phenol, *m*-cresol, guaiacol, 4-ethylphenol, 4-vinylphenol, 4-ethylguaiacol, 4-vinylguaiacol, eugenol and 2,6-dimethoxyphenol. Corbières wine has been reported to contain 0.001–0.1 mg/l phenol, *ortho*-cresol, *meta*-cresol, *para*-cresol, 2-ethylphenol, 4-ethylphenol, 4-vinylphenol, 2-methoxyphenol (guaiacol), 2-methoxy-4-ethylphenol, 2-methoxy-4-vinylphenol, acetovanillone and propiovanillone (Etiévant, 1981). The following volatile phenolic compounds were determined in sherry: 4-ethylphenol (0.35 mg/l), 2-methoxy-4-ethylphenol (4-ethylguaiacol; 0.08 mg/l), 2-ethylphenol (0.05 mg/l), 2-methoxy-4-vinylphenol (4-vinylguaiacol; 0.05 mg/l), 2,6-dimethoxy-4-ethylphenol (ethyl syringol; 0.04 mg/l), 4-vinylphenol (0.02 mg/l), *meta*-cresol (0.01 mg/l), *para*-cresol (0.01 mg/l) and 2-methoxy-4-allylphenol (eugenol; 0.01 mg/l). Phenol, *ortho*-cresol, 2-methoxy-4-methylphenol (4-methylguaiacol) and 2,6-dimethoxy-4-isopropylphenol (isopropyl syringol) were detected at trace levels (Tressl *et al.*, 1976).

Cinnamic acid derivatives, anthocyanins, flavonols and condensed tannins also occur in wine. Anthocyanins and tannins originating in grapes are the main pigments in red wine, and their presence influences the colour and organoleptic characteristics of the wine. Colour stability increases with the degree of methylation, glycosylation and acylation of the basic anthocyanin moiety (Van Buren *et al.*, 1970). Anthocyanins are water soluble and have a 4′-hydroxyflavylium structure. The individual anthocyanins differ in the number of hydroxyl and methoxyl groups in the molecule, as well as in the nature and number of glycosidically bound sugars. Furthermore, aliphatic and aromatic acids may attach to the skeleton of the aglycone; in the acylated anthocyanins, cinnamic acid and, more generally, *para*-coumaric acid is esterified with the hydroxyl group in the sixth position of a glucose molecule attached glycosidically to the aglycone (Windholz, 1983).

Five anthocyanins — delphinidin, petunidin, malvidin, cyanidin and peonidin — and their 3-monoglucosides and 3,5-diglucosides are found commonly in grapes and wines. Malvidin 3,5-diglucoside is apparently the principal pigment in wines produced from hybrid grapes, whereas malvidin monoglucoside predominates in wines made from *Vitis vinifera* grapes (Van Buren *et al.*, 1970). Quercetin-3-glucoside, quercetin-3-glucuronide and myricetin-3-glucoside (the principal flavonols), kaempferol-3-glucoside, kaempferol-3-galactoside and isorhamnetin-3-glucoside (minor compounds) and caffeoyl tartaric acid and *para*-coumaroyl tartaric acid all contribute to the pigment of grapes and wines.

Kaempferol- and myricetin-3-glucuronides and three diglycosides were also identified tentatively (Cheynier & Rigaud, 1986).

3.4 Compounds in spirits

(*a*) *Carbonyl compounds*

(i) *Aliphatic aldehydes*

Acetaldehyde (see IARC, 1985, 1987a) is frequently the major carbonyl component and generally constitutes more than 90% of the total aldehyde content. It is easily distilled together with water and alcohol and is therefore found in all spirits.

Acetal formation is a reversible reaction, with an equilibrium coefficient of about 0.9 (Misselhorn, 1975); thus, if the alcohol content is between 40% and 50% by volume, as is the case for many strong spirits, only 15-20% of the total amount of acetaldehyde combines with ethanol. Hence, acetal formation does not reduce the free aldehyde content of strong spirits markedly, even after prolonged maturation.

The total aldehyde content in alcoholic beverages has been found to vary widely; some levels are summarized in Table 33. In Scotch whisky and cognac, a number of other aldehydes are present at levels similar to that of acetaldehyde (Table 34).

According to an investigation by Marché *et al.* (1975), wine distillate and brandy contain the saturated aliphatic aldehydes from formaldehyde (C_1; see IARC, 1982a, 1987a) to dodecanal (C_{12}). Liebich *et al.* (1970) investigated the flavour compounds in Jamaican rum and found propionaldehyde (0.01 mg/l), isobutyraldehyde (0.25 mg/l), 2-methylbutyraldehyde (1.5 mg/l) and isovaleraldehyde (1.8 mg/l). Kirsch has been reported to contain the following aldehydes, calculated as mg/l ethanol: formaldehyde, 10-20; propionaldehyde, 10-30; valeraldehyde, $\leqslant 10$; *n*-heptanal, $\leqslant 10$; octanal, $\leqslant 10$; and *n*-nonanal, $\leqslant 10$ (Tuttas & Beye, 1977).

(ii) *Unsaturated aldehydes*

Of the unsaturated aldehydes, only acrolein (see IARC, 1985, 1987a) has been found in new, unaged whisky distillates. Propenal reacts with high concentrations of ethanol to form 1,1,3-triethoxypropane *via* 1,1-diethoxyprop-2-ene and 3-ethoxypropionaldehyde as intermediate products (Kahn *et al.*, 1968).

A number of unsaturated aldehydes have been identified in cognac and brandy. The occurrence of 2-buten-1-al and 2-hexen-1-al was reported in cognac by Marché *et al.* (1975). ter Heide *et al.* (1978) detected 2-methyl-2-propen-1-al in headspace and (*Z*)-2-methyl-2-buten-1-al, 3-methyl-2-buten-1-al, (*E*)-2-penten-1-al, (*E*)-2-methyl-2-penten-1-al, the (*E*)-isomers of 2-C_6, 2-C_7, 2-C_8, 2-C_9-enals and (*E,E*)-hepta-2,4-dien-1-al, nona-2,4-dien-1-al and deca-2,4-dien-1-al in extracts of cognac.

Table 33. Total aldehydes (mg/l ethanol), determined as acetaldehyde, in some brands of distilled beverages[a]

Beverage	Acetaldehyde
American bourbon whiskey	40-120
Canadian whisky	20-70
Irish whiskey	30-140
Scotch blended whisky	40-220
Scotch malt whisky	40-160
Brandy	130-600
Cognac	~210
Rum	20-150

[a] From Nykänen & Suomalainen (1983)

Table 34. Content (mg/l) of some low-boiling aldehydes in Scotch whisky and cognac[a]

Aldehyde	Whisky	Cognac
Acetaldehyde	7	10
Propionaldehyde	1.2	1
Isobutyraldehyde	20	20
2-Methylbutyraldehyde + 3-methylbutyraldehyde (Isovaleraldehyde)	6.3	4

[a] Reported by Nykänen & Suomalainen (1983)

Some unsaturated aldehydes have also been found in rum. Postel and Adam (1982) detected acrolein at 11 mg/l ethanol; ter Heide *et al.* (1981) found (*E*)-2-octen-1-al, (*E*)-2-nonen-1-al, (*E*,*E*)-2,4-decadien-1-al, β-cyclocitral, α-phellandral and geranial.

(iii) *Aliphatic ketones*

A large number of aliphatic ketones, from acetone to tetradecanone, have been identified in spirits; most are monoketones. In general, little attention has been paid to the determination of monoketones in spirits because of their relatively high sensory thresholds. In contrast, the occurrence of 2,3-butanedione (diacetyl) and 2,3-pentanedione in alcoholic beverages has been investigated (ter Heide, 1986).

The acetone content of spirits varies widely, and concentrations of 3-10 mg/l in whiskies, 0.25 mg/l in rums and <3-10 mg/l ethanol in cognacs and brandies have been reported (Nykänen & Suomalainen, 1983). Schreier *et al.* (1979) determined the contents of some monoketones in German and French brandies and French cognacs and found 2-pentanone (0.012-0.274 mg/l), 2-methylcyclopentanone (0.004-0.043 mg/l), 2-hexanone (0.009-0.117 mg/l), 2-heptanone (0.017-0.628 mg/l) and 2-nonanone (<0.001-0.107 mg/l). Liebich *et al.* (1970) found that a Jamaican rum contained 2-butanone (0.03 mg/l), 3-penten-2-one (7 mg/l), 2-pentanone (1.2 mg/l), 4-ethoxy-2-butanone (5 mg/l) and 4-ethoxy-2-pentanone (7.5 mg/l). Tuttas and Beye (1977) found 2-butanone and 2-heptanone at concentrations of 10 mg/l ethanol in kirsch.

(iv) *Unsaturated monoketones*

In an investigation of flavour constituents in Japanese whisky, Nishimura and Masuda (1984) identified three unsaturated ketones — tri-6-decen-2-one, penta-6-decen-2-one and hepta-6-decen-2-one. The unsaturated ketones 3-penten-2-one (Liebich *et al.*, 1970) and (*E*)-6-nonen-2-one (ter Heide *et al.*, 1981) have been identified in rum. Schreier *et al.* (1978) detected 0.05 mg/l 6-methyl-5-hepten-2-one in raw apple brandy. The unsaturated ketones (*E*)-3-penten-2-one, (*E*)-3-nonen-2-one, 6-methyl-5-hepten-2-one, (*E*)- and (*Z*)-6-methylhepta-3,5-diene-2-one, (*E*)-2-nonen-4-one and (*E*)-2-undecen-4-one have been found in cognac (ter Heide *et al.*, 1978).

(v) *Diketones*

The butterscotch odour of 2,3-butanedione can be recognized at very low concentrations. Diketones are marked flavour compounds in alcoholic beverages, many of which contain 2,3-butanedione and 2,3-pentanedione in detectable amounts (Nykänen & Suomalainen, 1983). 2,3-Butanedione (<0.01-4.4 mg/l) and 2,3-pentanedione (<0.003-0.57 mg/l) have been found in whisky, vodka, brandy and rum (Leppänen *et al.*, 1979).

(vi) *Aromatic aldehydes*

The simplest aromatic aldehyde, benzaldehyde, can be found in many distilled beverages. It has been detected in large amounts in brandies produced from stone fruits (Nykänen & Suomalainen, 1983). According to Bandion *et al.* (1976), the benzaldehyde content of cherry brandies is 33-75 mg/l; the highest amount, 129 mg/l, was found in an apricot brandy.

The appearance of aromatic aldehydes in spirits matured in wooden casks is associated with the degradation of wood lignin. During the maturation of whisky distillates, the amounts of aromatic aldehydes liberated depend on the type of cask; alcoholysis of lignin and extraction of the compounds with spirit give different yields when new or old charred or uncharred casks are used. It has been suggested that aromatic aldehydes produced in the free form by charring are extracted directly by the spirits. Moreover, ethanol reacts with lignin to form ethanol lignin, some of which breaks down to yield coniferyl and sinapic alcohols, which can then be oxidized into coniferaldehyde (4-hydroxy-3-methoxycinnamaldehyde) and sinapaldehyde (3,5-dimethoxy-4-hydroxycinnamaldehyde). Aldehydes with a double bond in the side chain, such as coniferaldehyde and sinapaldehyde, are further oxidized to yield vanillin (4-hydroxy-3-methoxybenzaldehyde) and syringaldehyde (3,5-dimethoxy-4-hydroxybenzaldehyde; Baldwin *et al.*, 1967; Baldwin & Andreasen, 1974; Reazin *et al.*, 1976; Nishimura *et al.*, 1983; Reazin, 1983). Concentrations of vanillin and syringaldehyde

Table 35. Content (mg/l) of vanillin and syringaldehyde in some brands of spirits[a]

Beverage	Vanillin	Syringaldehyde
Scotch whisky	0.31–2.06	0.67–4.04
Other whiskies	0.04–5.96	ND–10.2
Cognac and armagnac	0.25–1.85	0.41–3.75
Brandy	0.17–1.16	0.18–2.54
Rum	ND–3.17	ND–8.73

[a] From Lehtonen (1983); ND, not detected

in different commercial brands of distilled beverages are given in Table 35. Coniferaldehyde and sinapaldehyde were found in whisky and cognac, whereas salicylaldehyde was found only in whisky (Lehtonen, 1984).

(*b*) *Alcohols*

Some spirits, such as vodka, contain few flavour compounds and consist essentially of ethanol and water. In contrast, whiskies, cognacs, brandies and rums frequently contain large numbers of different volatile compounds.

(i) *Methanol*

Methanol is not a by-product of yeast fermentation but originates from pectins in the must and juice when grapes and fruits are macerated. In general, the methanol content of commercial alcoholic beverages is fairly small, except in those produced from grapes in prolonged contact with pectinesterase and in some brandies produced from stone fruits, such as cherries and plums. Apricot brandies have been found to contain up to 10 810 mg, plum brandies, up to 8850 mg, and cherry brandies, up to 5290 mg methanol/l pure alcohol. Cognac and grape brandies contain 103-835 mg/l and Scotch whisky 80-260 mg/l methanol (Nykänen & Suomalainen, 1983).

(ii) *Higher alcohols*

Higher alcohols and fusel alcohols (1-propanol, 2-methylpropanol, 2-methylbutanol, 3-methylbutanol and phenylethyl alcohol) are formed in biochemical reactions by yeast on amino acids and carbohydrates. The amounts in different beverages vary considerably. Scotch whisky has been reported to contain 1-propanol (70-255 mg/l), 2-methyl-1-propanol (170-410 mg/l), 2-methyl-1-butanol (74-124 mg/l) and 3-methyl-1-butanol (215-352 mg/l). Irish whiskey, Canadian whisky and Japanese whisky do not differ considerably from Scotch whiskies in concentrations of fusel alcohols, whereas American bourbon whiskeys can contain up to 1390 mg/l 2-methyl-1-butanol and up to 1465 mg/l 3-methyl-1-butanol.

Brandies and cognacs contain slightly more fusel alcohols than Scotch whiskies: 1-propanol (53-895 mg/l), 2-methyl-1-propanol (7-688 mg/l), 2-methyl-1-butanol (21-396 mg/l) and 3-methyl-1-butanol (98-2108 mg/l). The total amounts of fusel alcohols in rums correlate with the total congener content. Exceptionally high values have been reported for 1-propanol in some heavily flavoured Jamaican rums, in which the concentration of congeners was more than 9000 mg/l pure alcohol; the 1-propanol content ranged from 23 840 to 31 300 mg/l pure alcohol (Horak *et al.*, 1974a,b; Mesley *et al.*, 1975; Nykänen & Suomalainen, 1983).

The rose-like odour of phenethyl alcohol can be recognized in some whiskies, which usually contain 1-30 mg/l (ter Heide, 1986). In brandies and rums, the concentration is much lower. A very high content, 131 mg/l, was found in an American bourbon whiskey (Kahn & Conner, 1972).

A number of long-chain alcohols, up to C_{18}, have been found in distilled alcoholic beverages, but the concentrations are very small (Nykänen & Suomalainen, 1983).

(*c*) *Acids*

(i) *Aliphatic acids*

All the straight-chain monocarboxylic acids from C_2 to C_{18} and a large number of branched-chain acids have been identified in distilled alcoholic beverages (ter Heide, 1986); most are produced by yeast during fermentation. Recently, the presence of formic acid in cognac and rum has been confirmed, and it is one of the major acids in whisky (ter Heide, 1984, 1986). Saturated C_3-C_{18} straight-chain acids predominate, and acetic acid is generally the main component (ter Heide, 1986). Its relative proportion in Scotch whiskies is approximately 50% of total volatile acids, and that in other whiskies, 60-95%; cognacs and rums contain quantities of acetic acid amounting to 50-75% and 75-90% of the total volatile acids, respectively (Nykänen *et al.*, 1968).

The second largest acid component in distilled beverages is decanoic acid, followed by octanoic acid and dodecanoic acid or lauric acid. The concentration of palmitic acid and (*Z*)-hexadec-9-enoic acid (palmitoleic acid) is relatively high in Scotch whisky in particular. Of the short-chain acids, propionic, 2-methylpropionic, butyric, 3-methylbutyric and pentanoic acids are present in abundance. The concentrations of short-chain acids in rums were: acetic acid (4.5-11.7 mg/l), propionic acid (0.5-4.2 mg/l), butyric acid (0.4-2.6 mg/l), 2-ethyl-3-methylbutyric acid (0.1-2.2 mg/l) and hexanoic acid (0.3-1.3 mg/l; Nykänen & Nykänen, 1983); West Indian and Martinique rums contained 2-propenoic acid (0.1-0.2 mg/l) and dark rums, *trans*-2-butenoic acid, among others (ter Heide, 1986).

(ii) *Aromatic acids*

Small amounts of aromatic acids can be found in distilled alcoholic beverages (Nykänen & Suomalainen, 1983). Most are phenolic acids and probably originate in wooden casks used for maturation. In investigations of the nonvolatile compounds liberated by alcohol from oak chips, Nykänen, L. (1984) and Nykänen *et al.* (1984) found benzoic acid, phenylacetic acid, cinnamic acid, 2-hydroxybenzoic acid and benzenetricarboxylic acid in extracts. 3-Phenylpropanoic acid, salicylic acid and homovanillic acid ((4-hydroxy-3-methoxyphenyl)acetic acid) were identified in dark rum, and 4-hydroxybenzoic acid was identified in cognac matured for 50 years. In addition, coumaric acid (4-hydroxycinnamic acid) has been found in whisky, and gallic acid (3,4,5-trihydroxybenzoic acid), vanillic acid (4-hydroxy-3-methoxybenzoic acid), syringic acid (3,5-dimethoxy-4-hydroxybenzoic acid) and ferulic acid (4-hydroxy-3-methoxycinnamic acid) in rum (ter Heide, 1986). A prolonged maturation may increase the content of some aromatic acids, although, in armagnac, concentrations of cinnamic acid, benzoic acid, syringic acid, vanillic acid, ferulic acid, 4-hydroxybenzoic acid and 4-hydroxycinnamic acid reached their highest values after 15 years; in 30-year-old armagnac, the total amounts of these acids had decreased to approximately 30% of the maximal value (Puech, 1978).

(*d*) *Esters*

(i) *Esters of aliphatic acids*

Numerically, the largest group of flavour compounds in whisky, cognac and rum consists of esters (Nykänen & Suomalainen, 1983; ter Heide, 1986), most of which are ethyl esters of monocarboxylic acids. The straight-chain ethyl esters from C_2 up to C_{18} acids, and some ethyl esters of branched-chain acids, are present in whisky, cognac and heavily flavoured rum. The number of esters increases further by esterification of acids with fusel alcohols and with long-chain fatty alcohols as well as by the appearance of aromatic esters formed during maturation.

The total ester content varies widely in strong spirits. Esters have been found in aged Scotch malt whiskies (360 mg/l), in Scotch whiskies (550 mg/l), Irish whiskeys (1010 mg/l), Canadian whiskies (645 mg/l) and American whiskeys (269-785 mg/l). The ester contents of rums (44-643 mg/l) and brandies (300-6000 mg/l) are similar (Schoeneman *et al.*, 1971; Schoeneman & Dyer, 1973; Reazin *et al.*, 1976; Reinhard, 1977; Nykänen & Suomalainen, 1983).

Ethyl formate is a common component of spirits. Its concentration varies between 4 and 27 mg/l in whiskies and 13 and 33 mg/l in cognacs (Carroll, 1970; Nykänen & Suomalainen, 1983). Postel *et al.* (1975) reported 5-35 mg ethyl formate/l in rums. Ethyl acetate is quantitatively the most important component of the ester fraction, usually accounting for over 50%. Many short-chain esters, such as isobutyl acetate, ethyl isobutyrate, ethyl *n*-butyrate, ethyl isovalerate, 2-methylbutyl acetate and 3-methylbutyl acetate, have fairly strong odours; therefore, their occurrence in whisky, cognac and rum has been investigated extensively (Nykänen & Suomalainen, 1983).

In whisky, the concentrations of long-chain carboxylic acid esters increase from ethyl hexanoate up to ethyl decanoate and then decrease, so that C_{18} ethyl esters are usually the last components to be detected. In Scotch whisky, the ethyl esters of hexadec-9-enoic acid and hexadecanoic acid frequently occur in nearly equal amounts. In cognac, brandy and rum, the concentrations of the ethyl esters of C_{14}-C_{18} acids, and particularly of ethyl hexadec-9-enoate, are smaller than those in whisky (Suomalainen & Nykänen, 1970; ter Heide, 1986).

(ii) *Esters of aromatic acids*

The aromatic acids that occur in whisky, cognac and rum are also present as ethyl esters, although in very small amounts (Nykänen & Suomalainen, 1983). Higher amounts of ethyl benzoate have been found in plum brandies (ter Heide, 1986). Postel *et al.* (1975) found 8 mg ethyl benzoate/l alcohol in plum brandy, 10 mg/l in mirabelle brandy and 6 mg/l in kirsch. Beaud and Ramuz (1978) found 15-18 and 12-13 mg ethyl benzoate/l alcohol in kirsch and Morello cherry brandy, respectively. Schreier *et al.* (1978) reported that apple brandy contains 0.32 mg/l ethyl benzoate. Minor amounts of ethyl phenylacetate have been detected in cognac, German and French brandies and apple brandy (Schreier *et al.*, 1978, 1979).

(*e*) *Phenolic compounds*

Minor amounts of phenols, probably originating from raw materials, have been found in spirits. The phenolic compounds determined in whisky by Nishimura and Masuda (1971) and by Lehtonen and Suomalainen (1979) are listed in Table 36.

Table 36. Content (mg/l) of phenols in commercial whiskies

Compound	Malt whisky[a]	Blended Scotch whisky[a]	Scotch whisky[b]	Japanese whisky[b]
Phenol	0.1	0.19	0.003	0.012
ortho-Cresol	0.075	0.075	0.015	0.013
meta-Cresol	0.030	0.035	ND	ND
para-Cresol	0.050	0.050	0.009	0.010
2-Methoxyphenol (Guaiacol)	0.090	0.120	0.012	0.006
2,6-Xylenol	ND	ND	0.001	ND
2-Ethylphenol	ND	ND	0.002	0.002
4-Ethylphenol	0.070	0.040	0.009	0.002
2-Methoxy-4-methylphenol	ND	ND	0.005	traces
2-Methoxy-6-methylphenol	ND	ND	ND	traces
2-Methoxy-4-ethylphenol	0.100	0.030	0.035	0.002
2-Methoxy-4-allylphenol (Eugenol)	0.050	0.100	0.032	0.011

[a] From Lehtonen and Suomalainen (1979)
[b] From Nishimura and Masuda (1971)
ND, not detected

The phenolic compounds found in cognac are 2-methoxy-4-ethylphenol (0.29 mg/l), 2-methoxy-4-allylphenol (0.14 mg/l), phenol (0.03 mg/l), 4-ethylphenol (0.03 mg/l) and 2-methoxyphenol (0.03 mg/l). In addition, *ortho*-cresol, *meta*-cresol and *para*-cresol were detected as trace components. In a commercial dark Martinique rum, 4-ethylphenol (1.8 mg/l), 2-methoxy-4-ethylphenol (1.1 mg/l), 2-methoxy-4-allylphenol (0.8 mg/l), 2-methoxyphenol (0.7 mg/l), phenol (0.2 mg/l), *para*-cresol (0.08 mg/l), *ortho*-cresol (0.06 mg/l) and *meta*-cresol (0.04 mg/l) were found (Jounela-Eriksson & Lehtonen, 1981). Schreier *et al.* (1979) reported the occurrence of 4-ethylphenol and 2-methoxy-4-ethylphenol in grape brandy.

Tannins are present in spirits matured in wooden caskes. Brandies were reported to contain epicatechin, gallocatechin, catechin, flavonones and a number of other phenolic compounds (Marché *et al.*, 1975). The concentration of tannins in brandy aged for four to ten years in oak barrels ranged between 240 and 1120 mg/l (Guymon & Crowell, 1972). Bourbon whiskeys matured in charred barrels for up to 12 years contained 230-670 mg/l tannins (Baldwin & Andreasen, 1974).

3.5 Additives and contaminants

(*a*) *Flavouring additives*

Hops and hop extracts are used by breweries to improve the flavour of beers. The presence of nonvolatile, bitter and other substances — hop acids and volatile terpenes — in hops has been reviewed (Verzele, 1986) and the chemical composition of hops is summarized in Table 37.

Table 37. Chemical composition of hops[a]

Compound	Amount (%)
α-Acids	2-12
β-Acids	1-10
Essential oil	0.5-1.5
Polyphenols	2-5
Oil and fatty acids	traces-25
Protein	15
Cellulose	40-50
Water	8-12
Pectins	2
Salts (ash)	10

[a] From Verzele (1986)

Various plant extracts and essential oils are used in the manufacture of alcoholic beverages, in addition to synthetic products, to flavour liqueurs, aperitif beverages like vermouth and some vodkas. For instance, the strongly flavoured Russian vodka *subrowka* contains a blade of sweet, or holy, grass (*Hierochloe odorata*), beloved of the European bison, from which colouring matter and flavouring compounds are extracted by alcohol during storage. Many terpenic compounds, a number of ketones, alcohols, aldehydes, esters, lactones, phenols and phenol ethers, and acids have been identified as the flavour components of *H. odorata*. The principal component of the grass is coumarin, which represents about 60% of the total content of the volatile compounds (Nykänen, I., 1984).

Anethole, which has a strong aniseed-like odour, is another natural substance encountered in many beverages, and particularly in liqueurs. Natural anethole is obtained from plant materials, but it is also produced synthetically. A large number of other natural and synthetic flavourings with the same odour are used (Liddle & Bossard, 1984, 1985).

(*b*) *Other additives*

Preservatives are often added to beers and wines to prevent the activity of bacteria and moulds. In the UK, breweries are permitted to use sulphur dioxide as an antibacterial and antioxidant agent at a statutory limit of 70 mg/l. Many yeasts can form small quantities of sulphur dioxide during fermentation. However, the sulphur dioxide formed in beers is bound to naturally occurring compounds, and only small amounts can be detected. The sulphur dioxide content of German, Belgian and Dutch beers varies from none to 2.3 mg/l as free sulphur dioxide and from 0.8 to 2.4 mg/l as total sulphur dioxide (Nykänen & Suomalainen, 1983).

Sulphur dioxide is also one of the most important additives in wine making. It is added in aqueous solution or as potassium metabisulphite water solution; most is bound to aldehydes (Ough, 1987), pigments and polyphenols. In many countries, the permitted amount of free sulphur dioxide in commercial wines is 35-100 mg/l; the concentrations reported depend on the analytical method used. Most wines contain similar amounts of total sulphur dioxide ($>$100 mg/l; Nykänen & Suomalainen, 1983). Sulphur dioxide reacts slowly with free oxygen in wine and is therefore a poor antioxidant, unless it is added to wine at much higher levels than those generally accepted for inhibiting bacterial activity. Addition of ascorbic acid to wine just before bottling maintains a moderate level of sulphur dioxide. In the presence of oxygen, ascorbic acid reacts rapidly to yield hydrogen peroxide and dehydroascorbic acid. Sulphur dioxide then reacts with hydrogen peroxide to form sulphate ions (Ough, 1987).

The use of sorbic acid (hexa-2,4-dienoic acid) is permitted to protect wine against the activity of bacteria and moulds. At concentrations of 180-200 mg/l, it inhibits yeast growth but does not affect bacteria. Certain genera of malolactic bacteria convert sorbic acid to 2-ethoxy-3,5-hexadiene (Ough, 1987).

A number of spirits contain added colouring agents on which little data have been published. Sunset yellow FCF (FD & C yellow 6; see IARC, 1975, 1987a) has been reported to be present in cocktails and liqueurs (Anon., 1987).

(*c*) *Trace elements*

Most of the published literature on trace elements in alcoholic beverages concerns wine.

Concentrations of trace elements found in wines and some other alcoholic beverages are presented in Table 38.

Table 38. Content (mg/l) of trace elements in some alcoholic beverages[a]

Trace element	Light beer	White wine	Red wine	Whisky
Calcium	20–50	80–110	34–140	11–17
Chloride	–	20–80	18–390	–
Cobalt	–	0–0.012	0–0.012	–
Copper	0.2–0.5	0.4–1.0	–	–
Fluoride	–	0.2–0.5	0.06–0.40	–
Iodide	–	0.1–0.6	0.1–0.6	–
Iron	–	4–10	–	–
Magnesium	–	60–150	65–110	1.9–2.8
Manganese	–	0–3.0	0–2	–
Phosphorus	250–300	100–200	150–400	–
Potassium	320–440	660–920	750–1160	26–30
Sodium	20–60	5–40	10–140	1–3
Zinc	0.1–0.2	1.0–3.4	1–3	–

[a] From Hoofdproduktschap voor Akkerbouwprodukten (1984)

Trace elements from grapes are transferred during crushing into the must and eventually into wine (Eschnauer, 1982). The total concentration of mineral constituents in wine may be as high as 1000 mg/l and more (Eschnauer, 1967). The main trace elements are potassium, magnesium, calcium and sodium (see Table 38), but iron, copper, manganese and zinc are also present. In most wines, the iron content varies from 1 to 5 mg/l and copper from 0.1 to 1 mg/l.

A concentration of 0.0002-0.003 mg/l cadmium has been reported in European wines, the majority of levels being in the range 0.0002-0.0015 mg/l (Golimowski *et al.*, 1979a,b). The natural lead content of German wines has been reported to be 0.01-0.03 mg/l, and the average chromium content is 0.065 mg/l. It has been suggested that in younger wines the chromium content may be slightly higher (0.18 mg/l) than in old wines because they are more frequently in contact with stainless steel (Eschnauer, 1982). Interesse *et al.* (1984) determined 14 trace elements in 51 southern Italian wines; the chromium content was found to range from 0.01 to 0.81 mg/l and that of nickel from <0.01 to 0.09 mg/l.

In the 1960s, an epidemic of cardiomyopathy in Québec was seen after the introduction of cobalt to enhance the 'head' of foam on commercially produced beer (Morin & Daniel, 1967; Milon *et al.*, 1968; Dölle, 1969).

(*d*) *Contaminants*

For the purposes of this section of the monograph, the term 'contaminants' refers to those minor constituents sometimes present in alcoholic beverages which are not essential to the flavour and properties of the product. Some of these contaminants have known toxicological and, in some cases, carcinogenic effects.

(i) N-*Nitrosamines*

The occurrence of nitrosamines (see IARC, 1978) in alcoholic beverages has been well established in many investigations despite analytical difficulties (McGlashan *et al.*, 1968, 1970; Collis *et al.*, 1971; Bassir & Maduagwu, 1978). Reviews on the chemistry of formation of nitrosamines, with special reference to malting, are available, which report that the most important source of *N*-nitrosodimethylamine (NDMA) in beer is malt kilning by reactions involving nitrogen oxides (Wainwright, 1986a,b); a number of other kilning practices have been tested to reduce the quantities of *N*-nitrosamines in malt. The mechanism of formation suggests that small amounts of NDMA may occur in whisky (Klein, 1981). The concentrations found in various alcoholic beverages are given in Table 39. Leppänen and Ronkainen (1982) reported NDMA levels in Scotch whisky of 0.6-1.1 μg/l, an average of 0.3 μg/l in Irish whisky, of <0.1 μg/l in Bourbon whiskey and <0.05-1.7 μg/l in beer. Of 158 samples of beer, 70% were found to contain NDMA; the mean concentration in all samples was 2.7 μg/l; the highest value, 68 μg/l, was found in a so-called 'Rauchbier' which is made from smoked malt to give a smokey taste (Spiegelhalder *et al.*, 1979).

Other nitrosamines that have been identified in beer include *N*-nitrosopyrrolidine (Klein, 1981) and *N*-nitrosoproline (Massey *et al.*, 1982).

Table 39. Average amounts of N-nitrosamines in alcoholic beverages[a]

Beverage	No. of samples	NDMA (μg/l)	NDEA (μg/l)	NDPA (μg/l)
Cider distillate	120	<0.05–10	<0.05–2	<0.05–2.6
Cider	21	<0.05–1.8	<0.05–2.2	<0.05–1.1
White alcohol[b]	9	<0.05–2.2	<0.05–4.8	ND
Whisky	8	<0.05–0.7	<0.05–0.4	<0.05–0.4
Rum	8	<0.05–0.3	<0.05–0.2	<0.05–0.2
Cognac and armagnac	12	<0.05–1.6	<0.05–0.15	<0.05–0.3
Wine	33	<0.05–0.6	0.3 (one sample)	ND
Beer	40	<0.05–8.6	<0.05–0.8	ND

[a] From Klein (1981); NDMA, N-nitrosodimethylamine; NDEA, N-nitrosodiethylamine; NDPA, N-nitrosodipropylamine; ND, not detected

[b] From fruit or grains

(ii) *Mycotoxins*

A wide variety of moulds is found on grapes; *Aspergillus flavus* may be among them, and hence aflatoxins may occur exceptionally in wines. In an investigation by Schuller *et al.* (1967), aflatoxin B_1 was found to be present in two (Ruländer 1964 and Gewürztraminer 1964) of 33 German wines analysed at amounts of <1 μg/l. Lehtonen (1973) investigated the occurrence of aflatoxins in 22 wines from different countries by a thin-layer chromatographic method and reported <1 μg/l in 11 samples and 1.2-2.6 μg/l in five samples; six samples were aflatoxin-free. Takahashi (1974) increased the sensitivity of the method and was able to determine aflatoxins B_1, B_2, G_1 and G_2 at concentrations of 0.25 μg/l wine; aflatoxins were not determined in 11 samples of French red wine, Spanish sherry, madeira and port wine. French wines and German wines investigated using improved methods have also been found to be free of aflatoxins (Drawert & Barton, 1973, 1974; Lemperle *et al.*, 1975).

Aflatoxins were found by Peers and Linsell (1973) in 16 of 304 Kenyan beer samples at concentrations of 1-2.5 μg/l. The probable source was rejected maize, which is often used in the production of local beers. In a study in the Philippines, 47% of 55 samples of [unspecified] alcoholic beverages contained aflatoxins at an average concentration of 1.9 μg/l (Bulatao-Jayme *et al.*, 1982).

Two other mycotoxins, ochratoxin A (see IARC, 1983a, 1987a) and zearalenone (see IARC, 1983a), have been found in beer made from contaminated barley. Krogh *et al.* (1974) reported that the malting process degraded ochratoxin A in moderately contaminated (830 μg/kg and 420 μg/kg) barley lots; however, small amounts of ochratoxin A (11 μg/l and 20 μg/l) were left in beer produced from heavily contaminated barley (2060 μg/kg and 27 500 μg/kg). Commercial and home-made Zambian beer brewed from maize contained zearalenone at concentrations ranging from no detectable amount to 2470 μg/l. The concentration of zearalenone in 12% of 140 beer samples in Lesotho was 300-2000 μg/l (Food and Agricultural Organization, 1979).

(iii) *Ethyl carbamate (urethane)*

Urethane (see IARC, 1974, 1987a) is formed by the reaction of carbamyl phosphate with ethanol (Ough, 1976a, 1984) and is, therefore, present in most fermented beverages. Ough (1976b, 1984) found urethane in commercial ales (0.5-4 25g/l), in saké (0.1-0.6 mg/l) and in some experimental (0.6-4.3 μg/l) and commercial wines (0.3-5.4 μg/l).

Numerous samples of distilled alcoholic beverages have been analysed for their urethane content, probably because of the high amounts found in stone fruit brandies. Christoph *et al.* (1986) reported urethane in Yugoslavian plum brandy (1.2-7 mg/l), in Hungarian apricot brandy (0.3-1.5 mg/l), in various plum brandies (0.4-10 mg/l), in kirsch (2-7 mg/l), in fruit brandy (0.1-5 mg/l), in Scotch, American bourbon, Canadian and Irish whiskies (0.03-0.3 mg/l) and in cognac and armagnac (0.2-0.6 mg/l). Urethane contents of alcoholic beverages reported by Mildau *et al.* (1987) are given in Table 40. Adam and Postel (1987) reported the following average urethane contents in some fruit brandies: kirsch (1.8 mg/l), plum brandy (1.7 mg/l), mirabelle plum brandy (4.3 mg/l), Williams pear brandy (0.18 mg/l), apple brandy (0.5 mg/l), Jerusalem artichoke brandy (0.7 mg/l) and tequila (0.1 mg/l).

Table 40. Content (mg/l) of urethane in some alcoholic beverages[a]

Beverage	Number of samples	Content
Kirsch	67	0.2-5.5
Plum brandy	27	0.1-7.0
Mirabelle plum brandy	8	0.2-2.3
Stone fruit brandy	2	0.2-0.3
Grain spirit	1	ND
Wine distillate	2	ND-0.05
Cognac	1	0.04
Rum	4	ND-0.06
Whisky	2	0.04-0.08
Liqueur	9	ND-0.16
Sherry	5	0.02-0.07
White wine	10	ND-0.02
Red wine	7	ND-0.05

[a] From Mildau et al. (1987); ND, not detected (<0.01 mg/l)

(iv) *Asbestos*

Asbestos (see IARC, 1977a, 1987a) fibres have been identified in some alcoholic beverages, possibly arising from the filters used for clarifying beverages, from water used during the production processes and from asbestos-cement water pipes. Biles and Emerson (1968) detected fibres of chrysotile asbestos in British beers by electron microscopy followed by electron diffraction examination. Cunningham and Pontefract (1971) found asbestos fibres in Canadian and US beers (1.1-6.6 million fibres/l), in South African, Spanish and Canadian sherries (2.0-4.1 million fibres/l), in Canadian port (2.1 million fibres/l), in French and Italian vermouth (1.8 and 11.7 million fibres/l), and in European and Canadian wine [concentrations not reported]. The asbestos fibres in Canadian beer and sherry were identified as chrysotile, while some of the European samples contained amphibole asbestos. Fibres of chrysotile asbestos were also found by electron microscopy in three of nine samples of US gin (estimated maximal concentrations, 13-24 million fibres/l; Wehman & Plantholt, 1974).

In some European countries, the use of asbestos to filter alimentary fluids has been prohibited (e.g., Ministère de l'Agriculture, 1980).

(v) *Arsenic compounds, pesticides and adulterants*

The use of arsenic-containing fungicides in vineyards may lead to elevated levels of arsenic in grapes and wines (see IARC, 1980, 1987a). Crecelius (1977) analysed 19 samples of red and white wines and found arsenite (As^{+3}) at <0.001-0.42 mg/l and arsenate (As^{+5}) at 0.001-0.11 mg/l. Four samples were further analysed for total arsenic content by X-ray fluorescence (diffractometry), which confirmed these results. A review of older studies by Noble *et al.* (1976) reported concentrations ranging from 0.02 to 0.11 mg/l in nine US wines.

Aguilar *et al.* (1987) have investigated the occurrence of arsenic in Spanish wines. They found that crushing, pressing, cloud removal and yeast removal after fermentation and finally clarification and ageing markedly reduced the arsenic content of wine. Arsenic contamination of German wines has been reported to have decreased since 1940, levelling out at 0.009 mg/l in wines after 1970. The natural arsenic content has been assumed to be 0.003 mg/l, with earlier arsenic contents reaching the level of 1.0 mg/l due to use of arsenic-containing pesticides and insecticides (Eschnauer, 1982).

The fungicides used in vineyards include zineb (see IARC, 1976a, 1987a), maneb (see IARC, 1976a, 1987a), mancozeb, nabam, metalaxyl, furalaxyl, benalaxyl, cymoxanil, triadimefon, dichlofluanid, captan (see IARC, 1983b, 1987a), captafol, folpet, benomyl, carbendazim, thiophanate, methyl thiophanate, iprodione, procymidone, vinclozolin, chlozolinate (Cabras *et al.*, 1987) and simazine (Anon., 1980). Residues of metalaxyl, carbendazim, vinclozolin, iprodione and procymidone may be found in wine. In addition, ethylenethiourea (see IARC, 1974, 1987a), an impurity and a degradation product of ethylene bisdithiocarbamates (zineb, maneb, mancozeb, nabam), has been reported to be present at trace levels in wine (Cabras *et al.*, 1987). Hiramatsu and Furutani (1978) reported that the concentrations of trichlorfon and its metabolite, dichlorvos (see IARC, 1979b, 1987a), are higher in wine than in berries, indicating that they may be accumulated in wine.

Fungicides that are prohibited in most European countries, Australia and the USA but may be used in other countries include diethyl dicarbonate, dimethyl dicarbonate, pimaricin, 5-nitrofurylacrylic acid and *n*-alkyl esters of 4-hydroxybenzoate (Ough, 1987).

Occasionally, illegal additives, which may be very toxic and which are not permitted for use in commercial production in most countries, have been identified in alcoholic beverages. These include methanol, diethylene glycol (used as a sweetener), chloroacetic acid or its bromine analogue, sodium azide and salicylic acid, used as fungicides or bactericides (Ough, 1987).