

Release of distillate flavour compounds in Scotch malt whisky

John M Conner,*[†] Alistair Paterson and John R Piggott

Centre for Food Quality, University of Strathclyde, Department of Bioscience and Biotechnology, 204 George Street, Glasgow G1 1XW, Scotland, UK

Abstract: Equilibration of a flavour volatile between a distilled spirit and the headspace is a two-stage process. The first equilibrium occurs between the bulk solution and the headspace spirit interface, and the second between the surface layer and the headspace. The first stage is represented by the activity coefficient of the flavour volatile, which, for hydrophobic compounds, is greatly reduced by the aggregation of ethanol molecules in aqueous solution. The second equilibrium is governed by the vapour pressure of the solute and the ambient temperature and pressure. In mixed saturated solutions the composition of the surface layer and consequently the headspace is determined by the concentration and activity coefficients of the mixture components. Components of wood extract were found to act principally on the first equilibrium. Ethanol lignin acted in the same manner as high molecular weight esters and alcohols of the distillate, displacing volatile components from the surface layer. The suppressant effect of ethanol lignin was lost at 37°C and consequently would only be important in nosing of spirit samples. Wood extract was found to decrease the critical point for the aggregation of ethanol, resulting in reduced activity coefficients for ethyl decanoate from 5 to 30% ethanol at both 25 and 37°C. This effect would reduce the spirit-mouthspace partition coefficient with the resulting decreased release of flavour volatile in the mouth. This mechanism would explain the decreased impact of undesirable, immature aromas when wood matured spirits are consumed.

© 1999 Society of Chemical Industry

Keywords: whisky; flavour; volatility; maturation; wood; oak; distilled spirit; alcohol

INTRODUCTION

Previous studies on the maturation of Scotch whisky have shown that addition of extracted wood components can alter the activity coefficients of distillate aroma compounds.^{1,2} For a Scotch malt distillate, maturation results in quantitative rather than qualitative changes, with the concentration of many components increasing due to the evaporation of ethanol and water.³ Changes in the activity coefficients of aroma compounds, however, suggest a possible mechanism for the reduced perception of less desirable characteristics, described as *oily*, *soapy* and *grassy*, after maturation.⁴

Either aroma or taste can be used to differentiate between different distillates and mature and immature whiskies.⁵ Perceived flavour characteristics are due to a combination of the taste, imparted by non-volatile components, and the smell, imparted by the volatile components.⁶ Part of the perceived quality and intensity of flavour of whisky is therefore related to the concentrations of volatile components released

into the air space of the mouth. Nosing and tasting represent radically different environments for the release of volatiles from whisky. The former generally allows time for equilibration of the headspace in the nosing glass and may be considered a static system. The latter is very definitely a dynamic process with the effects of human physiological processes, such as respiration rate, mouth temperature and saliva composition and volume becoming important.

Changes in the activity coefficients of flavour compounds were demonstrated for static equilibria at 25°C and consequently are most relevant to nosing of spirit. Decreases in the activity coefficients of aroma compounds occurred on addition of either long chain esters or wood extractives.² In combination there was, however, a minimum headspace concentration that could be achieved by the addition of wood extractives and long chain esters.

Harrison and Hills⁷ used the penetration theory of interfacial mass transfer to model dynamic flavour release from liquid emulsions in the mouth. Their

* Correspondence to: John M Conner, The Scotch Whisky Research Institute, The Robertson Trust Building, Research Park North, Riccarton, Edinburgh EH14 4AP, Scotland, UK

[†] Present address: The Scotch Whisky Research Institute, The Robertson Trust Building, Research Park North, Riccarton, Edinburgh EH14 4AP, Scotland, UK

Contract/grant sponsor: Biotechnology and Biological Sciences Research Council, UK

Contract/grant sponsor: Chivas Brothers

(Received 1 July 1997; revised version 30 October 1998; accepted 10 December 1998)

mathematical model was based on the assumption that the rate limiting step was the resistance to mass transfer across the emulsion–gas interface. The model predicted that the maximum ‘flavour’ concentration was dependent on the initial flavour concentration but that the time to reach maximum ‘flavour’ concentration was independent of it. The theory also showed that both measures of ‘flavour’ release were sensitive to other factors such as the mass transfer coefficient and the gas–emulsion partition coefficient.

Malt whisky contains high concentrations of long chain esters and alcohols which produce a super saturated solution when diluted for nosing or consumption.¹ The excess solute forms agglomerates which can incorporate shorter chain esters, alcohols and aldehydes from the solution, decreasing their free solution and consequently headspace concentrations.² The presence of wood components extracted during maturation increases the incorporation of hydrophobic compounds into agglomerates, further reducing their free solution concentration and consequently the headspace concentration. Wood extracts have been shown to affect the size and stability of the ester agglomerates formed on dilution.⁸ Therefore malt whisky, diluted for consumption, may be regarded as an emulsion but with a portion of the flavour molecules forming the disperse phase.

As a first step to understanding the release of distillate flavour compounds in Scotch malt whisky, we investigated the effect of temperature on the partition of ethyl esters into the headspace. Ethyl decanoate was used as a typical hydrophobic aroma compound with similar polarity to those responsible for immature aromas in spirits.⁹ The effect of temperature on the headspace partition coefficients was determined in the presence of long chain aliphatic components of the distillate and non-volatile wood components.

MATERIALS AND METHODS

Materials

Ethanol was HPLC grade (Rathburn Ltd, Walkerburn, UK); water was purified by a MilliU10 system (Millipore UK Ltd, Watford, UK); ethyl esters and 3-methyl-1-butanol (iso-amyl alcohol) were >98% pure (Sigma Chemical, Poole, UK). Model solutions were prepared containing esters at concentrations typical of cask strength and bottled malt whiskies (Table 1).

Headspace analysis

Headspace concentrations and activity coefficients were determined by gas chromatography using previously published methods.¹ The activity of ethyl hexadecanoate was determined using a 7 µm polymethylsiloxane coated micro-extraction fibre (Supelco UK, Poole, UK) exposed in the headspace for 5 min then desorbed at 270°C in the SPI injector of a Finnegan-MAT ITS40 gas chromatograph linked mass spectrometer (GC-MS; Finnegan-MAT, Hemel

Table 1. Composition of models used for headspace analysis. Models were prepared by dilution from 65% ethanol (v/v) containing esters alone (immature model) or with 65% ethanol used to extract cask wood (mature models)

Congener	Cask model	Bottled malt model
3-Methylbutanol	200	200
Ethyl octanoate	9	9
Ethyl decanoate	20	20
Ethyl dodecanoate	15	8
Ethyl tetradecanoate	5	1
Ethyl hexadecanoate	15	3

Concentrations are in mg l⁻¹ at 23% ethanol (v/v).

Table 2. Effect of wood extract fractions on ethyl decanoate headspace concentration over a 25 µg ml⁻¹ solution in 23% ethanol (v/v)

Fraction	Headspace (ng ml ⁻¹)
Blank	88
Fraction 1	76*
Fraction 2	83
Fraction 3	98*
Fraction 4	52**
Fraction 5	56**

* Significant difference from blank ($p < 0.05$).

** Significant difference from blank ($p < 0.01$).

Hempstead, UK). A 0.32 mm × 30 m CP-Sil 52 CB column (Chrompack UK Ltd, London, UK), df=0.5 µm, was employed with helium gas at 1 ml min⁻¹ as carrier and a temperature gradient of 15°C min⁻¹ from 60°C to 240°C. Peak areas were calculated as the sum of responses to m/z 284 and 285.

Wood extracts

Wood extracts were prepared by extracting oak chips (60 g) from a new, charred American oak stave (*Quercus alba*) with 500 ml 65% v/v ethanol at ambient temperature for 1 month. The extract was filtered and stored at -18°C before use.

A sample of extract (200 ml) was fractionated by the addition of 10 g microgranular cellulose and rotary evaporated to approximately 50 ml under vacuum at 40°C. A further 100 ml of water was added and the mixture stirred for 30 min and then filtered to give fraction 1 (Table 2). The cellulose powder was then successively extracted for 30 min with 200 ml of 23, 40, 65 and 100% ethanol (v/v) to give fractions 2, 3, 4 and 5, respectively. Fractions were stored at -18°C before use. HPLC analysis¹⁰ revealed that fractions 1 and 2 contained the largest concentrations of aromatic aldehydes and acids. Lower concentrations were detected in fraction 3 with compounds virtually absent from fractions 4 and 5.

Wood samples were analysed using a Finnegan-MAT ITS40 GC-MS. A 0.32 mm × 25 m HT-5

column, $df=0.1\ \mu\text{m}$ (SGE Ltd, Milton Keynes, UK), was employed with helium gas at $1\ \text{ml min}^{-1}$ as carrier. Initial oven temperature was 80°C , held for 5 min, thereafter increasing at $10^\circ\text{C min}^{-1}$ to 320°C . Injection temperature was 320°C and the transfer line was held at 340°C throughout the analysis.

For gel permeation chromatography (GPC), a $25 \times 500\ \text{mm}$ column was packed with Sephadex LH-20 gel swollen in 65% ethanol. An aliquot of 10 ml was introduced manually to the top of the column and the flow rate controlled at $1\ \text{ml min}^{-1}$ by an LKB 2132 peristaltic pump. Fractions (10 ml) were collected and spectra from 200 to 600 nm were recorded.

RESULTS AND DISCUSSION

Activity or headspace partition coefficient?

Headspace concentrations may be expressed relative to the solution concentration (partition coefficient) or relative to the headspace concentration over the pure solute (activity). From the latter, the activity coefficient of a dissolved solute can be calculated, which is inversely related to the solubility of the solute. Both measures have advantages. The headspace partition coefficient describes the distribution of the compound between the liquid and gas phases, whereas the activity coefficient describes the interaction of the compound with the solvent. To determine the headspace concentration from the activity coefficient requires knowledge of the vapour pressure of the compound at the required temperature and pressure.

Model solutions (Table 1) were used to measure the effect of temperature on the headspace concentration of ethyl decanoate. Samples were analysed with esters only ('immature' models) and with added wood extract ('mature' models). When expressed as a headspace concentration, ethyl decanoate showed an increase in headspace concentration with temperature (Fig 1). Increases were greater for the immature models. At both ester concentrations, the presence of wood components reduced the effect of temperature on headspace concentration.

When expressed as activities, however, the immature models showed no significant change with

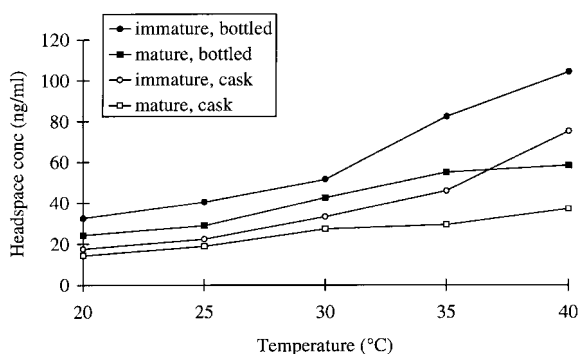


Figure 1. Ethyl decanoate headspace concentrations over model bottled and cask malt whiskies diluted to 23% ethanol (v/v) at different temperatures.

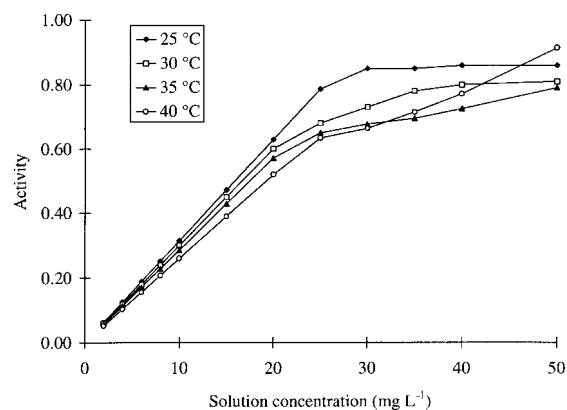


Figure 2. Plot of activity against solution concentration for ethyl decanoate.

temperature (0.28 for bottled malt and 0.18 for cask malt). Mature models therefore showed a decrease in activity with increasing temperature, from 0.23 at 25°C to 0.14 at 40°C for bottled malt and from 0.18 at 25°C to 0.09 at 40°C for cask malt. The increase in headspace concentration is a consequence of the increase in concentration over the pure solute. This showed a log-linear increase with temperature in accordance with the Clapeyron–Clausius equation with a constant latent heat of vaporisation over short temperature ranges.

The model solutions, however, contained a mixture of esters in a saturated solution at 25°C . To confirm that the lack of change in ethyl decanoate activity was the result of unchanged esters solubility rather than an artefact of the solution saturation, the activity coefficient of ethyl decanoate at infinite dilution was calculated in 23% ethanol at 25, 30, 35 and 40°C from the linear portion of the plot of activity against solution concentration (Fig 2). Only a small decrease in the activity coefficient was observed from 3.4×10^5 at 25°C to 2.8×10^5 at 40°C . Results from ethyl hexadecanoate (1.2×10^9 at 25°C to 0.9×10^9 at 40°C) suggest that the effect of temperature does not change with increasing ester chain length.

Effects of distillate components

In previous publications^{1,2} changes in activity coefficients were attributed to changes in the bulk of the solution. The above results, however, suggested that equilibration between the bulk solution and the headspace is a two-stage process. The first equilibrium is between the bulk solution and the air–liquid interface. This equilibrium is described by the activity coefficient. The second equilibrium, between the surface and the headspace, is determined by the vapour pressure of the compound, and the ambient temperature and pressure. Below the limit of solubility the esters behave in a manner analogous to surfactants below their critical micelle concentration.¹¹ The esters in the model solutions, however, and in corresponding whiskies are in saturated solutions at 23% ethanol. As the solution of a single ester becomes saturated, a

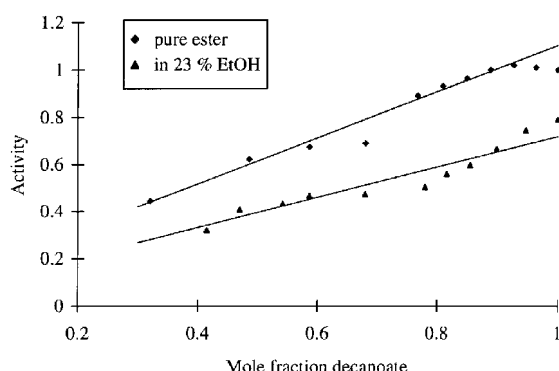


Figure 3. The activity of ethyl decanoate in mixtures with ethyl hexadecanoate either as pure esters or dissolved in 23% ethanol (v/v).

monolayer of ester is formed, resulting in a headspace concentration close to that of the pure solute.

Changes in the activity of one ethyl ester on addition of another could therefore result from the changing composition of the surface layer. Saturated solutions of ethyl decanoate with increasing mole fractions of ethyl hexadecanoate were therefore compared with two-phase systems comprising 23% ethanol in water and ester phases containing different mole fractions of ethyl decanoate and hexadecanoate (Fig 3). The gradient for the two-phase system was 1.0, indicating ideal mixing of the two esters, but for the solution the gradient was 0.65. This difference is most probably due to the activity coefficients of the two esters. The higher activity coefficient of the ethyl hexadecanoate results in a greater proportion in the surface layer, and consequently a reduction in the proportion of ethyl decanoate. Thus it appears that the headspace concentration of an ester may be altered by the addition of another ester, not as a result of an interaction in the bulk solution but as the result of changing composition of the surface layer.

Surface layers have long been known to affect the evaporation of water.¹¹ Increasing chain length of aliphatic alcohols has been shown to be accompanied by a reduction in the solubility of water in the alcohol.¹² It is therefore possible that increasing concentrations of long chain esters in the surface layer may reduce the concentration of ethanol, with resulting effects on the volatility and pungency.¹³

Activity coefficients of ethanol and ethyl acetate in ethyl decanoate and ethyl hexadecanoate were therefore measured. For each solute-ester combination, 20 points were used that gave a straight line through the origin (ie extrapolating to infinite dilution). Adjusted R^2 were $>97\%$. No significant differences, however were observed between the different esters as solvents. Mean activity coefficients were between 3.8 (standard deviation (SD)=0.4) for ethanol and 0.8 (SD=0.2) for ethyl acetate. The activity coefficient for ethanol in the ethyl esters is very close to its activity coefficient at 23% v/v in water (from Guggenheim).¹⁴ This similarity would explain why the presence of an ester surface layer has little effect on the headspace concentration of ethanol.¹

Effects of wood components

There are thus two possibilities for the interaction of wood components extracted during maturation that would reduce headspace partition coefficients. The first possibility is that non-volatile hydrophobic wood components enter the surface layer and displace volatile components to the solution, or to agglomerates if the solution is saturated. Alternatively wood extract may increase the solubility of esters in 23% ethanol and consequently reduce the excess concentration at the surface.

Fractionation of wood extract showed that fractions active in suppressing volatility were more soluble in ethanol than in water (Table 2). GC-MS analysis identified hexadecanoic and octadecenoic acids together with a mixture of plant sterols, possibly including β -sitosterol and campesterol. Wood matured spirits have previously been shown to contain β -sitosterol, stigmasterol, campesterol and β -sitosterol-D-glucoside.¹⁵ None of these components however had any effect on the headspace concentration of ethyl decanoate or model immature spirits. GPC of combined fractions 4 and 5 (Fig 4) gave one early eluting peak (50 to 200 ml) before both coniferaldehyde (144 to 228 ml) and vanillin (228 to 300 ml). HPLC analysis confirmed the absence of aromatic aldehydes and acids. The combined GPC fractions of this 'ethanol lignin' significantly reduced the ethyl decanoate headspace concentration above a $25\mu\text{gml}^{-1}$ solution in 23% ethanol at 25°C from 88 to 68ngml^{-1} ($p < 0.01$). Increasing the equilibration temperature to 37°C however, resulted in no significant reductions on addition of the 'ethanol lignin'.

The solubility of the 'ethanol lignin' suggests that it is hydrophobic and as such would be expected to develop a surface excess concentration when whisky was diluted for sensory assessment. In nosing, close to static equilibrium at room temperature, 'ethanol lignin' could suppress the release of shorter chain, more volatile compounds by its presence in the surface layer. The suppressant effect, however is lost at higher temperatures, so it would have a limited effect on

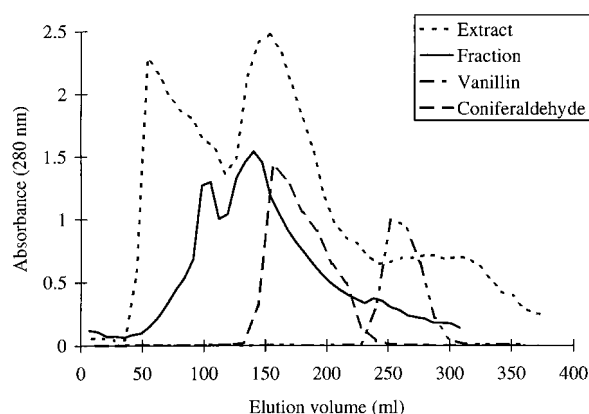


Figure 4. Gel permeation chromatographic trace of oak extract, isolated ethanol lignin fraction and standard solutions of vanillin and coniferaldehyde.

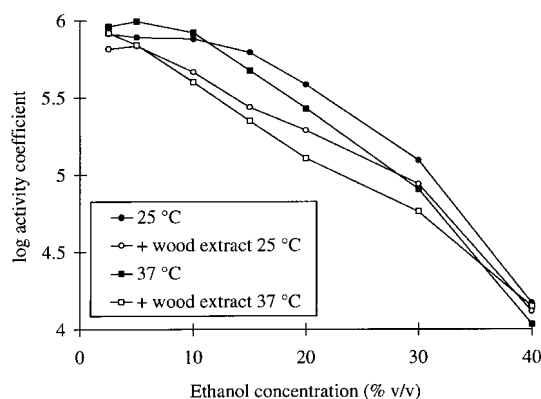


Figure 5. The effect of changing ethanol concentration on the activity coefficient of ethyl decanoate in different model systems.

flavour release in the mouth during tasting of whisky. Further, the elution of the 'ethanol lignin' from cellulose required a concentration of 65% ethanol or higher. This suggests that a large proportion would be removed when whisky is chill filtered prior to bottling.¹⁶

Changes in solubility

The effect of changing ethanol concentration on the activity coefficient of ethyl decanoate was determined in the presence and absence of wood extract at both 25 and 37 °C (Fig 5). At 25 °C ethanol water mixtures without wood extract showed no significant differences in activity coefficient up to 15% ethanol. Thereafter activity coefficients decreased with increasing ethanol concentration. Addition of wood extract resulted in a significant decrease in the maximum activity coefficient ($p < 0.05$). Decreases in activity coefficients with increasing ethanol concentration started from 5%. At 37 °C in ethanol water without added wood extract no significant difference in the maximum activity coefficient was observed, but a significant decrease ($p < 0.05$) in activity coefficient occurred between 10 and 15% ethanol. Addition of wood extract at 37 °C produced no significant difference in the maximum activity coefficient, but the activity coefficient decreased from 2.5% ethanol.

The effect of changing ethanol concentration on the activity coefficients of ethyl esters has previously been determined.¹⁷ Below a critical ethanol concentration, activity coefficients remained constant, but above this concentration, values decreased rapidly with increasing ethanol concentration. This behaviour could be understood from physico-chemical studies of the behaviour of ethanol in aqueous solution.¹⁸ Infrared and ultrasonic compressibility measurements showed that below 15% ethanol such solutions are monodisperse. Above 20% ethanol the water structure is unable to retain all ethanol molecules and the excess aggregates to form micelle-like structures. Activity coefficient data suggest that the monodisperse solution behaves as an aqueous single phase. In contrast the solution of higher ethanol content behaves as a

microemulsion and the rapid decrease in activity coefficients is the result of incorporation of esters into the ethanol aggregates.

Fig 5 identifies two parameters that have an effect on the activity coefficient of ethyl decanoate, both apparently acting through the aggregation of ethanol. The first is temperature. Increasing temperature from 25 °C to 37 °C results in a decrease in the critical point for aggregation. Similar shifts with temperature have been reported for *tert*-butanol.¹⁹ The enthalpy of solution of ethanol is negative ($-52.4 \text{ kJ mol}^{-1}$)²⁰ resulting in a lower solubility of ethanol at higher temperatures and consequently a lower critical point for aggregation. Conversely the small decreases in activity coefficient of ethyl decanoate and hexadecanoate indicate a small positive enthalpy of solution, and similar data have been reported for alcohols and other amphiphiles in water.²¹

A shift in the first critical point for aggregation is also observed in the presence of wood extract. This shift is observed at both 25 °C and 37 °C and results in reduced activity coefficients of ethyl decanoate for ethanol concentrations from 5 to 30%. Changes in the physico-chemical nature of whiskies and brandies during maturation have been studied by nuclear magnetic resonance,^{22,23} differential scanning calorimetry,²⁴ small angle X-ray scattering²³ and mass spectrometric analysis of liquid clusters,²⁵ and suggest some changes in the structural properties of ethanol and water. Results from small angle X-ray scattering suggested the existence of a greater degree of non-uniform structure in aged brandies. The mass spectrometric analysis of liquid clusters found that maturation of whisky in a cask increased the amount of large ethanol polymer hydrates. Therefore the reduction in the activity coefficient of ethyl decanoate in the presence of wood components could be the result of either more or larger ethanol aggregates with a greater capacity for solubilising ester.

The effective components extracted from wood during maturation have yet to be identified. Changes observed in O^{17} nuclear magnetic resonance and differential scanning calorimetry were attributed to fractions containing different wood and whisky components. The driving force for both the formation of the surface layer and the aggregation of ethanol and longer chain esters is the hydrophobic effect. Further research is required to ascertain whether the effects of 'structure making' components in the wood extract are the same as ionic co-solutes on the CMC of non-ionic surfactants.²⁶

CONCLUSIONS

Two factors affected the headspace-spirit partition coefficient of a typical hydrophobic aroma compound. High molecular weight esters displaced volatile compounds from the surface layer, resulting in lower headspace concentrations. The activity coefficients of ethyl esters were only slightly affected by temperature,

and headspace concentrations increased as ester vapour pressure increased with temperature. Wood extract decreased the critical point for the aggregation of ethanol, resulting in reduced activity coefficients of ethyl decanoate from 5 to 30% ethanol. Both factors could reduce the spirit-headspace partition coefficient of volatile distillate compounds at 37°C. The gas-liquid partition coefficient influences both the time to reach maximum flavour volatile concentration and the maximum flavour volatile concentration released from emulsions in the mouth. This suggests a link between previously reported physico-chemical changes during maturation and the sensory quality of distilled spirits. The reduction in activity coefficients by wood components extracted during maturation would reduce the concentrations of hydrophobic aroma compounds released in the mouth and decrease the impact of undesirable, immature aromas when matured spirits are consumed.

ACKNOWLEDGEMENTS

This research is funded by the Biotechnology and Biological Sciences Research Council of the UK and Chivas Brothers, Paisley, Scotland.

REFERENCES

- Conner JM, Paterson A and Piggott JR, Agglomeration of ethyl esters in model spirit solutions and malt whiskies. *J Sci Food Agric* 66:45–53 (1994).
- Conner JM, Paterson A and Piggott JR, Interactions between ethyl esters and aroma compounds in model spirit solutions. *J Agric Food Chem* 42:2231–2234 (1994).
- Philp JM, Cask quality and warehouse conditions, in *The Science and Technology of Whiskies*, Ed by Piggott JR, Sharp R and Duncan REB, Longman, Harlow, UK, pp 264–294 (1989).
- Piggott JR, Conner JM, Paterson A and Clyne J, Effects on Scotch whisky composition and flavour of maturation in oak casks with varying histories. *Int J Food Sci Technol* 28:303–318 (1993).
- Piggott JR and Jardine SP, Descriptive sensory analysis of whisky flavour. *J Inst Brewing* 85:82–85 (1979).
- Delahunty CM, Piggott JR, Conner JM and Paterson A, Comparative volatile release from traditional and reduced-fat cheddar cheese upon mastication in the mouth. *Ital J Food Sci* 8:89–98 (1996).
- Harrison M and Hills BP, Effects of air-flow rate on flavour release from liquid emulsions in the mouth. *Int J Food Sci Technol* 32:1–9 (1997).
- Paterson A, Piggott JR, Horne DS and Conner JM, Solute structures in aged malt distillates, in *Proceedings of the Fourth Aviemore Conference on Malting, Brewing and Distilling*, Ed by Campbell I and Priest FG, Institute of Brewing, London, UK, pp 222–225 (1994).
- Piggott JR, Conner JM, Clyne J and Paterson A, The influence of non-volatile constituents on the extraction of ethyl esters from brandies. *J Sci Food Agric* 59:477–482 (1992).
- Conner JM, Paterson A and Piggott JR, Analysis of ligin from oak casks used for the maturation of Scotch whisky. *J Sci Food Agric* 60:349–353 (1992).
- Adamson AW, *Physical Chemistry of Surfaces*, Wiley, New York, USA, pp 46–98 (1976).
- Davis SS, Higuchi T and Rytting JH, Determination of thermodynamics of functional groups in solutions of drug molecules, in *Advances in Pharmaceutical Sciences*, Vol 4, Ed by Bean HS, Bechett AH and Carless JE, Academic Press, New York, USA, pp 73–261 (1974).
- Withers SJ, Piggott JR, Leroy G, Conner JM and Paterson A, Factors affecting pungency of malt distillates and ethanol-water mixtures. *J Sensory Studies* 10:273–283 (1995).
- Guggenheim EA, *Thermodynamics*, North-Holland Physics Publishing, Amsterdam, The Netherlands, pp 170–219 (1968).
- Black RA and Andreasen AA, Steroids in aged whiskey. *JAOAC* 56:1357–1361 (1973).
- Booth M, Shaw W and Morhalo L, Blending and bottling, in *The Science and Technology of Whiskies*, Ed by Piggott JR, Sharp R and Duncan REB, Longman, Harlow, UK, pp 295–326 (1989).
- Conner JM, Birkmyre L, Paterson A and Piggott JR, Headspace concentrations of ethyl esters at different alcoholic strengths. *J Sci Food Agric* 77:121–126 (1998).
- D'Angelo M, Onori G and Santucci A, Self-association of monohydric alcohols in water: compressibility and infrared absorption measurements. *J Chem Phys* 100:3107–3113 (1994).
- Iwasaki K and Fujiyama T, Light-scattering study of clathrate hydrate formation in binary mixtures of *tert*-butyl alcohol and water. 2. Temperature effect. *J Phys Chem* 83:463–468 (1979).
- Franks F and Desnoyers JE, Alcohol-water mixtures revisited, in *Water Science Reviews*, Vol. 1, Ed by Franks F, Cambridge University Press, Cambridge, UK, pp 171–232 (1985).
- Tanford C, *The Hydrophobic Effect*, Wiley, New York, USA, pp 14–20 (1980).
- Aishima T and Matsushita K, Measurements of food ageing by multinuclear NMR, in *Frontiers of Flavour*, Ed by Charalambous G, Elsevier Science Publishers, Amsterdam, The Netherlands, pp 321–337 (1988).
- Aishima T, Matsushita K and Nishikawa K, Measurements of brandy ageing using O^{17} NMR and small angle X-ray scattering, in *Elaboration et Connaissance des Spiritueux*, Ed by Cantagrel R, Lavoisier-Tec & Doc, Paris, France, pp 473–478 (1992).
- Nishimura K, Ohnishi M, Masuda M, Kunimasa K and Matsuyama R, Reactions of wood components during maturation, in *Flavours of Distilled Beverages*, Ed by Piggott JR, Ellis Horwood, Chichester, UK, pp 241–255 (1983).
- Furusawa T, Saita M and Nishi N, Analysis of ethanol water clusters in whisky, in *Proceedings of the Third Aviemore Conference on Malting, Brewing and Distilling*, Ed by Campbell I, Institute of Brewing, London, UK, pp 431–438 (1990).
- Elworthy PH, Florence AT and MacFarlane CB, *Solubilisation by Surface-Active Agents*, Chapman & Hall, London, UK, pp 13–60 (1968).