Development of a technology for reducing polycyclic aromatic hydrocarbons in smoked food and smoked ingredients

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Development of a Zeolite Filter for Removing Polycyclic Aromatic Hydrocarbons from Smoke and Smoked Ingredients whilst Retaining the Smoky Flavor

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ABSTRACT

The popularity of smoked foodstuffs such as sauces, marinades and rubs is on the rise. However, during the traditional smoking process, in addition to the desirable smoky aroma compounds, harmful polycyclic aromatic hydrocarbons (PAHs) are also generated. In this work, a selective filter was developed which reduces PAH concentrations in a smoke by up to 90%, whilst maintaining a desirable smoky flavor. Preliminary studies using a cocktail of 12 PAHs stirred with a zeolite showed the potential for this zeolite to selectively remove PAHs from a simple solution. However pre-treatment of the smoke prior to application removed the PAHs more efficiently and is more widely applicable to a range of food ingredients. Whilst volatile analysis showed that there was a concomitant reduction in the concentration of the smoky compounds such as 2-methoxyphenol (guaiacol), 2-methylphenol (*m*-cresol) and the isoeugenols, sensory profiling showed that the difference in perception of flavor was minimal.

Keywords: polycyclic aromatic hydrocarbons (PAH), smoke flavor, zeolite filter, GC-MS,

INTRODUCTION

Smoking of foods, although historically a means of preservation, is used nowadays to impart a desirable smoky flavor to many popular foods, particularly rubs, sauces, seasonings and marinades. The aroma compounds which contribute to the smoky flavor have been characterized in smoked foods such as salmon,[1] sausages,[2] and smoke cured bacon,[3] as well as in liquid smoke[4] derived from different woods[5] and aromatic plants.[6] The burning of lignin produces phenols, particularly methoxyphenols, which impart potent smoky, burnt, tar, phenolic and spicy notes. GC-Olfactometry has been used to show that 4-methylphenol (*p*-cresol), 4-methoxyphenol (guaiacol) and (*E*)-2-methoxy-4-(1-propenyl)phenol (isoeugenol) contribute to the smoky spicy notes[1] whereas sweeter notes such as vanilla and toffee arise from the formation of vanillin, 2-furancarboxaldehyde, and 2-hydroxy-3-methyl-2-cyclopenten-1-one (cyclotene).

However, the smoking process generates a group of dangerous carcinogens that are responsible for lung cancer in cigarette smokers, and epidemiological evidence has implicated food-derived polycyclic aromatic hydrocarbons (PAHs) in the development of liver and other cancers in humans.[7, 8] Of the hundreds of PAHs generated during the smoking process, the International Agency for Research on Cancer has classified benzo[a]pyrene (BaP) as a Group 1 carcinogen (i.e. known humans carcinogen) and 16 others have been classified as either Group 2A or Group 2B carcinogens (probable and possible carcinogens).[9] The EU Commission Regulation No 1881/2006 recognised the need to achieve levels of as low as reasonably achievable, and set maximum concentrations of BaP between 1 and 20 μg/kg depending on the food ingredient and the intended use. This was updated in 2008 by the European Food Safety Authority[10] who recommended that the sum of the concentrations of benzo[a]pyrene (BaP), chrysene (CHR), benz[a]anthracene (BaA) and benzo[b]fluoranthene

(BbF), which collectively are referred to as PAH4, was more suitable as an indicator of potential toxicity.

The food industry has been preparing smoke flavorings for several decades by condensing aerosol smoke in water, then subjecting the resulting solution to a purification process. Concerns about the occurrence of PAHs in smoke flavorings led the EU to initiate an evaluation of smoke flavorings and determine the risk to consumers. They concluded that there is less of a health concern with these products compared to the natural smoking process,[11] but it is a requirement that these liquid smokes are labelled as "smoke flavoring" and they cannot be labelled as natural. Such labelling is seen less favourably by consumers and retailers in the current drive for natural and 'clean label' products. To address the health concerns relating to smoking processes (particularly traditional smoking processes, but also production of liquid smoke) there is a need for a new technology that is capable of reducing the levels of PAHs in aerosol smoke, thereby reducing the exposure of consumers to PAHs, whilst maintaining the desirable flavor of the smoked food products.

The reduction and potential elimination of PAHs from smoke and liquid smoke is of interest to many industries (tobacco industry, car industry, environmental agencies, and foodstuffs) and strategies have been reviewed.[12] Of those techniques investigated, treatment with a zeolite has produced the most promising results. Radojičić et al.[13] reported the use of a zeolite catalyst CuZSM-5 to reduced PAHs in tobacco smoke, and the PAH content of exhaust gases from a combustion engine was successfully reduced by treatment with the zeolite clinoptilolite.[14] The same zeolite has been used to remove PAHs from paraffin.[15] Alternative strategies have been used in different industries. Microbiological techniques for removal of PAHs from contaminated environments (soil, water) have been reviewed Seo et al.,[16] and Rentz et al. report the specific degradation of BaP by *Sphingomonas yanoikuyae* JAR02.[17]

Our aim was to develop a zeolite filter which could be applied to a range of products throughout the food industry, removing PAHs without compromising the desirable smoke flavor. The zeolite, clinoptilolite, is an inexpensive naturally occurring aluminosilicate mineral which has been shown to remove PAHs from cigarette smoke and paraffin, the latter being a more challenging non-polar matrix. In this work, we demonstrate the effectiveness of this zeolite in removing PAHs from various matrices, including smoke, on a laboratory scale and in an industrial smoking chamber. In addition, our hypothesis, that the retention of PAHs was selective and that aroma molecules would pass through the filter was tested. Both volatile analysis and sensory analysis were used to investigate the flavor in food prepared from both filtered and unfiltered smoke.

MATERIALS AND METHODS

Materials

Chemicals. Perylene and the mixture of PAHs were obtained from LGC Standards (Teddington, U.K.). The mixture contained each of the following at 20.1 ± 0.1 mg/L in dichloromethane: naphthalene (NPTH), acenaphthylene (ACYN), acenaphthene (ACEN), fluorine (FLUO), phenanthrene (PHEN), anthracene (ANTH), fluoranthene (FLA), pyrene (PYR), benz[a]anthracene (BaA), chrysene (CHR), benzo[b]fluoranthene (BbF), benzo[k]fluoranthene (BkF), benzo[a]pyrene (BaP), indeno[1,2,3-cd]pyrene (IcdP), dibenz[a,h]anthracene (DBahA) and benzo[ghi]perylene (BghiP). Cyclohexane (99.5%), methanol and N,N-dimethylformamide (99.8%) were obtained from Sigma-Aldrich Co Ltd. (Poole, U.K.). The aroma compound standards were obtained as follows: 2-furancarboxaldehyde, benzeneacetaldehyde, 2-methylphenol, 3-methylphenol and 4-methylphenol were obtained from Fisher Scientific (Loughborough, U.K.); 2-methoxy-4-prop-2-enylphenol (eugenol)from Givaudan (Milton Keynes, U.K.); (*E*)-2-methoxy-4-(1-propenyl)phenol with (*Z*)-2-methoxy-4-(1-propenyl)phenol present as a minor impurity from

Mane (London, U.K.); 3-methylcyclopentane-1,2-dione from IFF (Haverhill, U.K.); 5-methyl-2-furancarboxaldehyde, 1-(5-methyl-2-furanyl)ethanone, 2-furanmethanol and 1-(5-methyl-2-furanyl)propan-1-one from Oxford Organics (Hartlepool, U.K.); 1-(2-furanyl)ethanone, phenol, benzofuran, 2-methoxyphenol, 4-methyl-2-methoxyphenol, 4-ethyl-2-methoxyphenol, 4-ethenyl-2-methoxyphenol, 5-butyl-4-methyldihyrofuran-2(3*H*)-one (unspecified mix of isomers), 4-hydroxy-3-methoxybenzaldehyde, 4-hydroxy-3-methoxybenzoic acid and 1,3-dimethoxy-2-hydroxybenzene (syringol) from Sigma Aldrich (Poole, U.K.). All other chemicals used were standard laboratory chemicals.

Ingredients: Oak wood chips were supplied by Ashwood Smoking Chips Ltd. (Kettering, U.K.), and preheated at 130 °C prior to use to reduce moisture content (12% loss after 3 h). Rapeseed oil (refined and de-odorised) was obtained from BFP Wholesale (Leeds, U.K.) and refined and deodorised MCT coconut oil was obtained from Oleon (Sutton, U.K.). Tomato ketchup was obtained from a local supermarket.

Zeolite. Zeolite was supplied by RS Minerals Ltd. (Guisborough, U.K.). The material is a calcium hydrated aluminosilicate of sedimentary origin, free of fibers and quartz, which contains a minimum of 85% clinoptilolite and a maximum of 15% feldspar, micas and clays. It was supplied both as a coarse grain size (grain size >~5 mm) and as a medium grain size (grain size ~1–4 mm). The medium grain zeolite was used in some experiments as received and 100 g was further fractionated for use in the laboratory experiments. A sieve shaker was used to produce five different fractions: size 1, 75–180 μm (yield: 4.6 g); size 2, 180–355 μm (6.4 g); size 3, 355 μm–1 mm (45.1 g); size 4, 1–1.4 mm (33.3 g); and size 5, >1.4 (10.0 g).

Reduction of PAHs from a simple matrix (experiments 1–3)

For experiment 1, zeolite (0.5 g) was added to a mix (5 ml) of 16 PAHs in dichloromethane (each present at 201 μg/L) and stirred. After 1 min, 500 μL was transferred to an Eppendorf tube and centrifuged in an Eppendorf MiniSpin Microcentrigfuge (Fisher Scientific,

Loughborough, U.K.) at maximum RCF (12,100 g) for 10 min prior to analysis by GC-MS. This was repeated after 5 and 60 min. The grain sizes selected for this experiment were either size 4 (1–1.4 mm) or size 2 (180–355 μm) and the zeolite was used either as received (AR) or after heating at 270 ºC in a GC oven for 12 h (HT).

For experiment 2, coconut oil (10 g) was spiked with 100 μL of the PAH mix (20.1 mg/L) and stirred for 4 h with size 4 zeolite (0.5 g) which had been activated at 270 ºC for 12 h. The coconut oil was filtered and the PAHs extracted from the oil as described below. For experiment 3, an aliquot (10 ml) of coconut oil that had been smoked for 72 h in a traditional smoking chamber, was treated with size 4 activated zeolite (1 g) for either 1 h or 18 h. The PAHs were extracted from the oil as described below prior to analysis by GC-MS. Approximate quantitation was by comparison with 2 and 5 mg/L PAH mixes spiked into unsmoked oil (100 μl in 10 g) and analyzed under the same conditions.

Dual stream laboratory scale smoker (experiments 4–8)

The laboratory scale dual stream smoker is illustrated in Figure S1. Smoke was produced by heating a standard 1 L conical flask containing up to 100 g of chipped oak over a gas burner. The conical flask was placed in a metal box containing 100 g sand (control) to disperse the heat. Air was pumped in through the stopper and out through the side arm to a condenser flask cooled by an ice bath. The uncondensed smoke was split and passed in parallel through two identical glass chromatography columns (50 cm x 2 cm i.d.) with a sintered glass frit at the bottom of each. One column (treated) was filled with zeolite (10 g, medium grain size as received) and the other column (control) was filled with ceramic antibumping granules (70 g) and enough sand (control) to balance the flows of the two columns (3–5 g). The smoke (~200–400 ml/min) was collected in 40 g of water. The treated and the control conditions were alternated between the two columns.

Single stream pilot scale smoker (experiments 9–10)

A single stream stainless steel rig[18] was used for the smoking process. Medium grade zeolite (1.4 mm, sieve size +14) was activated in a Lincat double stone base pizza oven (Lincoln, U.K.) at 265–285 ºC for 3 h prior to use. The wood furnace was loaded with 1.5 kg of dry oak wood shavings and the oil pan was loaded with 700 g of rapeseed oil. The wood was charged with hot ash and set to run for 6 h with the smoke running through one filter column and bubbled through the rapeseed oil. In experiment 9, the rig was employed with a) the filter empty, b) the filter containing 600 g zeolite as received and c) the filter containing 600 g activated zeolite. In a second pilot scale experiment (experiment 10), three different grain sizes of zeolite were used in the filter; size 5 (>1.4 mm), size 3 (355 μm–1 mm) and size 2 (180–355 μm).

Manufacturing scale smoker (experiments 11–12)

Additional trials (experiments 11–12) were carried out in a full scale smoking chamber in which different quantities of activated zeolite were tested. PAH analysis was carried out by Eurofins (Acton, U.K.) using a saponification step, followed by SPE and GC-MS similar to the method described below for extraction from oils.

Extraction of PAHs from aqueous samples (experiments 4–8)

The entire sample (40 ml) was shaken with 22 ml methanol (optimum ratio of water:methanol which had previously been determined) and the internal standard was added (1 ml of perylene (200 μg/L)). The PAHs were extracted with SPE based on a method by Zha et al.[19] The whole sample was passed through a Bond Elut CH SPE cartridge (1 g bed, 6 ml total volume, Crawford Scientific, Strathaven, U.K.) which had previously been conditioned with methanol (10 ml). The column was then washed with HPLC grade water (3 times) and once with 10 ml water methanol (65:35 v/v). The column was dried under vacuum (~70 kPa) for 30 min (previously optimised) and then eluted with cyclohexane (4 ml). Recovery was >80% for all PAHs except DBahA (70%).

Extraction of PAHs from oil (experiments 2–3, 9–10)

Extraction was based on a method reported by Stumpe-Viksna et al.[20] Oil (10 g) was placed into a round bottomed flask, 12 g of potassium hydroxide, 100 ml of ethanol and internal standard (perylene, 100 μL of 2010 ng/μL in dichloromethane) were added. The mixture was heated for 1 h (78 ºC) under reflux, filtered and extracted into cyclohexane. The cyclohexane phase was dried over anhydrous sodium sulfate and concentrated by rotary evaporator under reduced pressure (40 ºC)

The extract was applied to a SPE cartridge (Bond Elute CH, 6ml, Crawford Scientific, Strathaven, U.K.) previously conditioned with cyclohexane (5 ml). The flask was rinsed with cyclohexane (3 ml), and the PAHs were eluted with cyclohexane (6 ml). The collected fraction was evaporated to approximately 1 mL under a gentle stream of nitrogen. The concentrated extracts were transferred to autosampler vials ready for analysis by GC/MS. Two aliquots of unsmoked oil (10 g) were spiked with the mix of 16 PAHs (100 μL of 2 mg/L or 100 μL of 5 mg/L in dichloromethane). A three point calibration curve was used to estimate the concentration of the PAHs present in the smoked oils. For all but the naphthalene, acenaphthylene, acenaphthene and fluorine, there was a good linear relationship passing through the origin.

Gas Chromatography-Mass Spectrometry (GC-MS) of PAH extracts.

The extracts were analyzed using an Agilent 7890A gas chromatograph equipped with a Zebron ZB-AAA column (10 m x 0.25 mm i.d. x 0.25 μm film thickness) coupled to an Agilent 5975C MSD. The carrier gas was helium (1.69 ml/min) and the extract (1 μl) was injected in splitless mode. The GC oven was held at 45 °C for 135 s, the temperature was raised to 280 °C at 8 °C/min and then to 300 °C at 16 °C/min and held for 4 min. Mass spectra were recorded in electron impact mode at an ionization voltage of 70 eV and source temperature of 300 °C. The MS was operated in SIM/SCAN mode using eight time windows,

monitoring the following groups of ions, (dwell time of each 25 ms) to identify and quantitate the PAHs: NPTH *m/z* 128, 127, 102, 63; ACYN *m/z* 154, 153, 152, 76; ACEN *m/z* 154, 153, 152, 76; FLUO *m/*z 166, 1654, 139, 82; PHEN *m/z* 178, 176, 89; ANTH *m/z* 178, 176, 89; FLA *m/z* 202, 200, 101, 100; PYR *m/z* 202, 200, 101, 100; BaA *m/z* 228, 226; CHR *m/z* 228, 226; BbF *m/z* 264, 252, 250, 126; BkF *m/z* 264, 252, 250, 126; BaP *m/z* 264, 252, 250, 126; IcdP *m/z* 278, 276, 139, 138; DBahA *m/z* 278, 276, 139, 138; BghiP *m/z* 278, 276, 139, 138. For each PAH, the identity was confirmed by comparison of the mass spectrum and the retention time with those of the authentic standards. Data were controlled and stored by the ChemStation system. Six point calibration curves in the range 5–500 μg/L were carried out for each PAH in the mix, each point in duplicate. Response factors were obtained for each PAH ($r^2 > 0.99$) and these were used to quantitate the PAHs in the samples against the internal standard. Limits of detection for each method were estimated based on serial dilutions of the standard mix.

Thermogravimetry

A thermogravimetric measurement was carried out on a TA Instruments Q50 thermogravimetric analyzer. Zeolite (53.55 mg) was accurately weighed into a sample pan and placed in the instrument. The sample was first equilibrated at 30 °C then raised to 270 °C at a rate of 5 °C/min, then held at 270 °C to give a total experiment duration of 12 h. The weight of the sample was recorded over the course of the experiment.

Powder X-ray diffraction

Powder X-ray diffraction data were collected from zeolite samples on a Bruker D8 Advance (Cu $K_{\alpha1}$, λ= 1.54056 Å) diffractometer operating in capillary transmission mode. The diffractometer was equipped with a LynxEye detector. Monochromatic Cu $K\alpha_1$ is achieved with the use of a curved Johansson type primary monochromator. Furthermore, an 8 mm detector aperture slit and a metal knife edge collimator were used to minimise air scattering.

Samples were packed into 0.7 mm borosilicate glass capillaries before mounting on the diffractometer and rotated throughout the data collections in order to minimise any preferred orientation effects. An Oxford Cryosystems Cryostream Compact, mounted co-axially with the sample, allowed temperature control of the sample in the range room temperature to 220 °C. The temperature of the Cryostream was ramped from 20 °C to 100 °C directly, and then to 220 °C in 10 °C increments, before finally cooling to 20 °C. The sample was allowed to equilibrate at each temperature for 5 min before a diffraction data collection was started. Diffraction data in the range 4°–45° 2θ were collected with a step size of 0.017° 2θ and a count time per step of 0.6 s. At the end of the experiment, the capillary was stored, open to the atmosphere for four days before data were recollected at room temperature.

Extraction and analysis of volatile compounds

The volatiles were analyzed by SPME/GC-MS. Aliquots of oil (5 g) or tomato ketchup (5 g) were placed in a 20 ml SPME vial and were extracted using a DVB/Carboxen/PDMS Stableflex fiber (SupelCo, Poole, U.K.). The samples were equilibrated at 40 °C for 10 min with intermittent stirring prior to exposure to the fiber for 10 min at 40 °C. The fiber was desorbed in the injection port for 20 min and the volatile compounds analyzed using an Agilent 7890A gas chromatograph equipped with a Zebron ZB-5MSi column (30 m x 0.25 mm i.d. x 1 um film thickness) coupled to an Agilent 5975C MSD. Helium was the carrier gas (1.2 ml/min). After desorption, the oven was maintained at 40 °C for 5 min, then raised to 250 °C at 4°C/min. Mass spectra were recorded in electron impact mode at an ionization voltage of 70 eV and source temperature of 230 °C. A scan range of *m/z* 29–400 with a scan time of 0.69 s was employed and the data were controlled and stored by the ChemStation system. Volatiles were identified by comparing each mass spectrum with those of authentic samples analyzed under similar conditions. To confirm the identification, a homologous series of *n*-alkanes (C_5–C_{30}) were analyzed under the same experimental conditions to obtain LRI values,

which were compared to the LRIs of authentic compounds. The identity was confirmed by running both the sample and the standards on a Stabilwax-DA column (30 m x 0.25 mm i.d. x 0.5 um film thickness) from Thames Restek (Saunderton, U.K.). Analysis was carried out in triplicate for experiment 10, and in duplicate for experiment 12. Each set were run in one randomised block and the peak area of a standard 2-octanol solution run at the beginning and end of the series varied by less than 10%.

Sensory profiling

Tomato ketchup (100 g) was stirred with oil (2 g) which had been prepared (Experiment 10) on the pilot scale rig using either size 2, 3 or 5 activated zeolite or no zeolite (control). All samples were left to equilibrate for 1 h prior to tasting in amber bottles. A panel of nine trained assessors, each with a minimum of six months experience, was used for sensory profiling of the tomato ketchups. The assessors were first asked to describe the sensory characteristics of the smoky tomato ketchups. Following this initial collection of terms, with the help of references, a consensus vocabulary, consisting of 8 odor terms, 4 taste terms, 7 flavor terms, 3 mouthfeel terms and 3 after-effect terms was agreed by the assessors. The quantitative sensory assessment took place in individual sensory booths under red light and at room temperature controlled to 20±0.5 °C. Assessors were provided with a glass of warm water and unsalted crackers for palate cleansing between samples. Samples were presented to the assessors in a balanced randomised order and they were asked to assess the aroma of the ketchup. Then after tasting a small quantity off a teaspoon, they assessed the taste, overall flavor and mouthfeel of the ketchup and, after a 60 s break, the after-effect. The intensity of each attribute for each samples was recorded by the assessors on a 100-point unstructured line scale. All data were collected using Compusense version 5 software (Compusense Inc., Guelph, Ontario, Canada). A duplicate assessment was carried out in a separate session.

Statistical analysis

ANOVA was carried out on the volatile analysis from experiment 10 and multiple pairwise comparisons were done using the Fisher's least significant difference (LSD) test with the significance level set at p = 0.05. SENPAQ version 3.2 (Qi Statistics, Reading, U.K.) was used to carry out two-way ANOVA on sensory profiling data where main effects were tested against the sample by assessor interaction. Multiple pairwise comparisons were done using the Fisher's least significant difference (LSD) test with the significance level set at p = 0.05.

RESULTS AND DISCUSSION

Reduction of PAHs from a simple matrix (experiments 1–3)

Preliminary experiments consisted of stirring a standard mixture of 16 PAHs (201 μg/L) in dichloromethane in the presence of the zeolite. Two zeolite grain sizes were investigated, size 4 (1–1.4 mm) or size (2, 180–355 μm), the zeolite was used both as received and after activating at 270 °C, and exposure times were 1, 5 or 60 min. Table 1 shows the concentration of the selected PAHs remaining after exposure to the zeolite, under each set of conditions. The full set of 16 PAHs is given in the supplementary material. There are very clear trends. The greatest difference was observed between the size 4 and size 2 grain size, with far greater reductions achieved when the fine grains were used. The increase in surface area of the particles provides greater exposure of the PAHs to the zeolite structure. A greater reduction in PAHs was also observed in the pre-heated zeolite (HT) compared to zeolite in its natural state (AR), with, for example, the BaP concentration reduced after 60 min to 3% of the original using pre-heated medium zeolite, compared to 60% using the natural zeolite (experiment 1c vs. 1f). Furthermore, where there was a reduction in PAHs observed, there was a strong tendency for a greater reduction as the exposure time increased.

It was also observed that for each of the different PAHs, the concentrations were not all reduced to the same extent. There was a tendency for the higher molecular weight PAHs to decrease more than those of lower molecular weight. There was no significant reduction in

naphthalene with any of the exposure conditions, and only small reductions (<20%) in fluorene and phenanthrene. Benzo[a]pyrene showed the greatest reduction when the heated (HT) zeolite was used (experiment 1e–1g and experiment 1j–1l). The best conditions for reducing PAH levels were achieved in experiment 11 utilizing pre-heated and finely ground zeolite. This produced a reduction in all PAHs except naphthalene, a reduction >94% for each of the four regulated PAHs, and a reduction in BaP of >99.9% (0.2 ng/L remaining). This shows a reduction in the concentration of the Group 1 carcinogen BaP, significantly greater than reported for any other mitigation strategy.

When food grade deodorised coconut oil was used as the matrix (experiment 2), and spiked with the standard mixture of 16 PAHs (201 μg/L), after stirring with size 4 zeolite which had been activated (12 h at 270 °C), the same trends were not observed (Table 1). It may be that in the more lipophilic environment, migration of the PAHs into the zeolite is slower and less energetically favorable. The deodorised oil was also subjected to a real smoking process (rather than addition of a mix of PAHs). The oil was smoked with oak chips in a smoking chamber for 72 h and the resulting smoked oil was stirred with size 4 zeolite for 1 or 18 h and analyzed for PAHs (experiment 3). There was no consistent trend and concentration of PAHs was only reduced on average to 60% of the original (Table 1). Note however, that after 18 h exposure to the heated zeolite, benzo[b]fluoranthene and benz[a]pyrene showed the greatest reduction with only 15 and 20% remaining respectively.

The concentration of PAHs generated during the 72 h smoke varied quite substantially from 20 μg/kg for phenanthrene whereas the more carcinogenic PAHs were present at 1–2 μg/kg in the untreated sample, specifically benzo[a]pyrene was found at 1.4 μg/kg and the PAH4 total was estimated to be 7 μg/L. These levels are within the limits recommended by the EC Commission Regulation No 1881/2006 which set maximum concentrations as low as reasonably achievable (ALARA) at 2 and 20 μg/kg for BaP and PAH4 respectively in coconut

oil intended for direct human consumption or use as an ingredient in food. However, our aim was to reduce these levels yet further to minimise human exposure to these known carcinogens.

Although this technique demonstrated that in principal PAHs can be removed from simple matrices by stirring with zeolite, in practice, the reduction from the coconut oil was not sufficient to make this a useful technique for the food industry. Furthermore, this technique could only be applied to the most simple food matrices and could not be applied more generally to smoked foods such as smoked spices, smoked sauces and other key food ingredients. Subsequent experiments were designed to test the capacity of the zeolite to reduce the concentration of PAHs in smoke used to prepare smoked foods, rather than extracting them from the foods or ingredients post smoking. The working hypothesis was that removing PAHs from an aqueous smoke environment, in which the PAHs are poorly soluble, would be easier than removing them from a lipophilic environment.

Reduction of PAHs from smoke (experiments 4–12)

The generation of smoke from the burning of wood is a highly variable process, particularly when carried out on a laboratory scale. For this reason a dual stream smoker was devised so that one source of smoke could be split into two equal streams which would allow comparison of a treated smoke with a control. The laboratory scale rig is shown in Figure S1. Smoke was collected in 40 ml of water (selected for safety reasons and ease of analysis) and the samples were extracted by SPE prior to analysis by GC-MS. The process was run five times, each with slight modifications (Table 2). Firstly, it is clear that the smoking process was very variable but, in all cases, a decrease in PAHs was observed in the zeolite-treated sample compared to the control. In experiments 4 and 5, the zeolite was only activated in a beaker for 3 h at 270 ^{o}C and the reduction of PAHs was only 40% in the best cases. With 17 h activation in experiment 6, 7 and 8, there was a far greater reduction in all the PAHs. In experiment 6, the

concentration of all PAHs was reduced by >80% and, in the most successful experiment 7, they were all decreased by >90%. Of the group of PAH4, the concentrations of chrysene and benzo[a]anthracene were reduced by 92 and 94% respectively, and benzo[b]fluorene and benzo[a]pyrene were reduced to concentrations both below the limit of detection for this method. Thus, although the generation of smoke is variable, the reduction in the concentration of PAHs is consistent if the zeolite is activated for sufficient time.

Having demonstrated the potential of the zeolite to remove PAHs from smoke, a single stream pilot scale smoker was built, which produced a consistent stream of smoke. The filter could be filled with inert material, or left empty, and the filtered smoke was collected in rapeseed oil. In experiment 9, both the native zeolite and the activated zeolite were tested against a control and, when the activated zeolite was used, there was a consistent decrease in the concentration of PAHs. When this was repeated (experiment 10) using activated zeolite of different grain size (sizes 2, 3 and 5), the coarse zeolite (size 5) was not effective, the medium reduced the concentration of PAHs by only 40–60% but the fine (size 2) produced a smoked oil where the concentrations of all the PAH4 were below 0.5 μg/L. These trends are similar to those where the zeolite came into direct contact with the matrix (Table 1), and indicates that the zeolite must be activated prior to use, and that maximizing the surface area of the zeolite is key to developing an efficient process for removing PAHs from smoke. Further data from smoked oil produced on a manufacturing scale is provided in Table 2 (experiments 11 and 12) to show that this reduction in the concentration of PAHs can be achieved in an industrial smoking chamber.

Zeolite Structure

For successful removal of PAHs, the requirement to activate the zeolite led us to investigate its structure in more detail, in an attempt to understand the structural changes taking place during activation and the rate at which the zeolite structure reverts to its native state. The

thermogravimetric analysis (TGA) (Figure S2) shows a weight loss of ca. 11.5%, due to water loss with the majority of the loss occurring by the time the sample reached 270 °C. Water loss was essentially complete after 2 h.

The powder X-ray diffraction (PXRD) pattern of the zeolite changed markedly upon heating, reflecting the structural changes that arise from the loss of bound water in the structure. Figures S3 and S4 show the changes in the diffraction patterns as the temperature was raised in steps from 20 °C to 220 °C, whilst Figures S5 and S6 compare the pattern of the sample after cooling back to 20 °C either immediately (Figure S5) or after 4 days (Figure S6).

The PXRD and TGA results show structural changes associated with water loss from the zeolite crystal structure. The extent of water loss is in reasonable agreement with previous measurements.[21] That the water loss is associated with structural changes is evidenced by the significant changes in the appearance of the powder X-ray diffraction pattern as a function of temperature. The patterns for the sample at 220 °C and then after cooling back to 20 °C are largely superimposable and even after 4 days of storage, the sample had not fully reverted to its "as received" state. This slow re-uptake of water from the atmosphere was undoubtedly a function of the fact that the sample remained inside the 0.7 mm glass capillary during the storage period. Other experiments (not shown here) indicated that reversion to "as received" state after heating occurred much more quickly when the sample was left fully open to the atmosphere and that grain size played a role in the speed of reversion. This reversion process was important when considering scale-up to industrial smoking chambers.

No further attempt has been made to understand the mechanistic basis of PAH removal by the zeolite, nor to relate size of PAHs removed to zeolite pore size. Such an investigation would require detailed crystallographic and computational work that lies outside the scope of the current investigation.

Volatile Analysis

The smoked rapeseed oils produced in experiments 10 and 12 were retained for volatile analysis. Figure 1 shows the volatile profile of the control smoke vs. the smoke filtered through the fine grain zeolite. Inspection of Figure 1 suggests that the loss of volatile components is minimal when the fine filter is employed, corroborated by only a 7% decrease in the total area of those peaks analysed (Table 3). This is extremely encouraging and shows that most of the volatile compounds were not retained by the fine particulate zeolite and, more importantly, it demonstrates that the observed reduction in the concentration of PAHs is not simply a result of less smoke being passed through the rig. There was, however, a tendency for the peak areas of the later eluting compounds to be diminished, which warranted a more detailed analysis of the volatile data.

In excess of 200 compounds were identified, of which 24 were selected for comparison. The selection was based on previous GC-O work by Varlet et al,[1] with some additional compounds of interest. The details of the volatile analysis are shown in Table 3, where the changes in volatiles are expressed as the mean peak area normalized to the control where no filter was employed. The full data including the coefficients of variation for experiments 10 and 12 are shown in Table S1. The overall trends in Table 3 are clear. Whereas the overall volatile profiles are similar, for all compounds analyzed except 5-methyl-2-furancarboxaldehyde, there were significant differences between the samples. For all compounds except 2-furancarboxaldehyde, 2-furanmethanol, 5-methyl-2-furancarboxaldehyde and 1-(2-furanyl)ethanone, the unfiltered smoke contained the greatest amount whereas use of the fine filter produced the least, suggesting that the filter did indeed retain some of the key smoky compounds. Whilst 50% of the 4-methoxyphenol was retained, the isoeugenol isomers were not detected when the fine filter was used. The furans were relatively unchanged across the four oils, but the key smoky compounds were more affected, particularly the methoxyphenols (guaiacols), many of which were reduced to 30% or less of

the original amount. These changes are likely to affect the overall perception of the smoky aroma, so further work is in progress to estimate the relative contribution of the different compounds to the aroma of smoke.

It is also noticeable in Table 3 that there were some apparent anomalies in the volatile profiles of the two intermediate samples (size 5 vs. size 3 zeolite filter), particularly in phenol and 2-methoxyphenol where the peak areas were higher in the sample prepared with the smaller grain size 3. This trend can be seen in the total peak area and in other volatiles, particularly the lower molecular weight ones (Table 3) and can only be explained as a result of the inconsistency of the smoking process, which for some reason has produced more of the highly volatile smoke compounds. This difference may be widespread across the whole chromatogram, but just less evident when the reduction in volatiles is greater (higher molecular weight).

Informal sensory assessment of the smoked oils from experiment 10 revealed some minor changes in the aroma of the sample after treatment with the filtered smoke, compared to the control, but these differences tended to be a reduction in the harsh acrid notes and the overall flavor was even anecdotally improved. Formal sensory profiling was carried out on these four samples dosed into tomato ketchup at 2% (see below). The changes in the volatile profiles of the tomato ketchups are shown in Table 3, and the trends are similar to those found in the oils.

The oils generated in experiment 11 were also analyzed for volatile compounds. In this experiment, the weight of zeolite used increased across the series, and there was concomitant decrease in the PAHs. In this series, there was a very clear trend in the volatile profile: as the weight of zeolite used increased, all the phenols, guaiacols, and syringol showed a significant decrease in peak area. This was less so for the group of furans which remained relatively stable across the series. Thus in both series, there was a consistent decrease in key smoke compounds as the filter “strength” was increased, and in the most extreme cases, syringol was

reduced to 6% of the control and the isoeugenols were removed completely. It is important to establish what the impact on flavor perception is, given this decrease in key smoke compounds.

Sensory Analysis

Sensory analysis of the tomato ketchups revealed few significant differences between the samples (Table 4). Of the 25 attributes scored, only two (sweet aroma and throat burn) showed significant differences between samples of ketchup. However, Fisher's LSD also showed some emerging trends which are consistent with the volatile data. There was a tendency for the smoky bonfire and smoky mackerel notes to decrease across the series and this could be explained by the concomitant decrease in smoke-related volatiles such as the higher molecular weight guaiacols, isoeugenols and 1,3-dimethoxy-2-hydroxybenzene. However, the differences in these attributes were not observed in the flavor attributes, possibly masked by the intensity of the neat ketchup. The ketchup produced from the oil prepared with the fine zeolite filter was found to have a significantly sweeter aroma than the other three, and this may be a result of a decrease in the some of the smoky notes which otherwise mask the sweetness of the ketchup. Juicy fruity and tomato aroma followed the same trend but the difference was not significant at $p < 0.05$ ($p = 0.13$ and 0.2 respectively). Interestingly, when size 3 zeolite was used, the corresponding ketchup was found to have significantly more throatburn than the others. This may be related to the anomaly in the volatile profile discussed above.

The volatile data suggest that many of the key smoke compounds were present at lower concentrations in the PAH-reduced oils. However, the preliminary sensory data suggests that the impact on the flavor perception is minimal and the filtered smoke may even produce a slightly sweeter but less smoky oil. This relationships between the flavor perception,

consumer preference and the volatile profile are currently being investigated in much more detail in a range of food ingredients.

Comparison with other mitigation strategies

Mitigation strategies for PAHs in smoked foods have been proposed at all stages of the smoking process. Selection of the wood has already been widely investigated with respect to minimising PAH formation. Smoking of hardwoods such as oak, apple and alder produce fewer PAHs compared to softwoods like spruce and pine[20] due to the lower lignin content (oak 24% c.f. pine 35%).[22] Hitzel et al,[23] in the only paper where the aroma compounds were investigated in any detail, showed that during the smoking of frankfurters and mini salamis, the PAHs could be reduced by 35–55% by replacing beech with poplar or hickory, with very little change in the content of the key aroma compounds (guaiacol, 4-methylguaiacol, syringol, eugenol and (*E*)-isoeugenol). In liquid smoke, a reduction in total carcinogenic PAHs from 0.78 μg/kg (poplar) to 0.2–0.4 μg/kg for oak, cherry and beech was achieved on a laboratory scale,[24] but such low concentrations may not be representative of large scale smokers.

The conditions of the smoking process have also been thoroughly investigated. In a comparison reported by Duedahl-Olesen et al,[25] cold smoking of fish with BaP concentration ranging from 0–0.8 μg/kg was preferable to hot smoking (0.1–2 μg/kg), traditional smoking (mean, 5.3 μg/kg, n=213) or home smoking (up to 11 μg/kg). Several authors have determined optimum temperatures for the burning zone in terms of reducing the concentration of PAHs,[25] but information on the flavor is often lacking. Temperatures below 450 °C may limit the formation of PAHs, but also the formation of flavor.[12]

However, the treatment of the smoke prior to the smoking process has achieved greater reductions in the concentration of PAHs. More than 50 years ago, electrostatic precipitation of the solids in smoke was shown to reduce BaP by 66%[26] and treatment of smoke with an

494 aqueous scrubber prior to smoking fish was shown to reduce BaP by at least 70%.[22] More
495 recently, a filter comprising ice, cloth and activated carbon[27] has been patented specifically to
496 produce “tasteless super-purified smoke”.
497 Downstream from the smoking process, UV light and oxygen have been used to effect an
498 80% reduction of BaP in smoked herring[28] but very little information is available on the
499 relative toxicity of the products of UV oxidation and it has been speculated that they may be
500 more toxic than the PAHs.[12] Storage in polyethylene packaging has been shown to reduce the
501 concentration of PAHs in food,[23] and low density polyethylene (LDPE) achieved 97%
502 reduction in PAHs over a 7 day storage period in an aqueous model. A significantly smaller
503 reduction was achieved in oil or water-oil emulsion or in roasted duck skin (73% reduction).[29]
504 In these cases the mechanism is simply presumed to be migration of the PAHs into the
505 polymer.
506 The use of our clinoptilolite filter has several advantages over these methods. The filters
507 already reported simply act as a barrier to tar, PAHs and flavor. We have shown that although
508 inevitably some tar is removed by the zeolite filter, the PAH content of the smoke which
509 passes through is further reduced. BaP was reduced by 90% and PAH4 by 85% in a
510 commercial smoking chamber. We have demonstrated that the smoke filtered through the
511 zeolite is far from tasteless and has flavor properties very similar to the unfiltered smoke.
512 The use of a zeolite to treat the smoke is far more readily applicable than any biotechnological
513 method and more consistent than relying on packaging material to reduce the concentration of
514 PAHs, although the latter may be used to further reduce low levels of PAHs. It has been
515 successfully applied to oak-smoke oil and water which have subsequently been applied to
516 food products, and it has been used for direct smoking of paprika, jalapeno and tomato flakes.

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Associated Content

The dual stream processor is shown in Figure S1, the thermogravimetric analysis in Figure S2 and the PDXRA data in Figures S3–6. Full details of the volatile analysis are given in Tables S1, S2 and S3. This material is available free of charge via the Internet at http://pubs.acs.org.

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FIGURE CAPTIONS

Figure 1 Comparison of the total ion chromatogram of rapeseed oils smoked with and without a fine zeolite filter

Table 1 Concentration of PAHs in Dichloromethane or Coconut Oil after Exposure to Zeolite, (Control=201 μg/L)

expt no	experimental conditions zeolite[b]	size[c]	time[d]	BaA[a]	CHR[a]	BbF[a]	BkF	BaP[a]	IcdP	DBahA	BghiP
matrix = dichloromethane											
1a	AR	4	1	179[e]	199	126	163	154	123	117	138
1b	AR	4	5	167	195	108	149	135	117	103	108
1c	AR	4	60	146	183	98	130	120	115	103	128
1d	HT	4	1	90	132	71	69	45	97	83	69
1e	HT	4	5	58	92	56	45	16	59	56	32
1f	HT	4	60	39	65	46	23	5	33	50	10
1g	AR	2	1	31	42	31	17	27	40	29	39
1h	AR	2	5	42	66	38	27	31	43	40	34
1i	AR	2	60	40	59	37	26	29	46	42	33
1j	HT	2	1	8.6	11.8	11.5	3.9	0.3	nd	3.2	nd
1k	HT	2	5	8.1	13.0	12.7	3.8	0.3	1.4	5.7	0.9
1l	HT	2	60	7.1	12.2	12.1	3.2	0.1	nd	6.6	nd
matrix = coconut oil											
2a	HT	M	240	209	208	184	185	174	169	163	149
3a	None			1.4[f]	2.3	1.5	0.3	1.4	1.3	0.5	0.8
3b	HT	M	60	1.2[g]	1.8	0.8	0.2	1.1	1.4	0.3	0.4
3c	HT	M	1080	0.8[g]	1.6	0.2	0.2	0.3	1.5	0.2	0.3

[a]EU regulated PAH4

[b]AR=zeolite as received, HT=zeolite heated at 270 °C for 12 h

[c]Zeolite grain size 4 (1–1.4 mm) or size 2 (180–355 μm)

[d]Exposure time (min)

[e]Concentration (μg/L) compared to control (control = 201 μg/L)

[f]Concentration (μg/L) of naturally smoked oil before treatment

[g]Concentration (μg/L) of naturally smoked oil after treatment

nd = not detected

Table 2 Concentration of PAHs in Smoke Treated with Zeolite Filter compared to Untreated Control

expt no		experimental conditions			BaA[a]	CHR[a]	BbF[a]	BkF	BaP[a]	IcdP	DBahA	BghiP
		filter[b]	time[c]	flow rate[d]	dual stream laboratory scale smoker collected in CH_2Cl_2 (ng/L)							
4a	C	sand	68	200	295	273	146	115	123	67	48	21
4b	T	zeolite HT (3 h)	68	200	140	121	65	54	59	45	23	29
5a	C	sand	20	400	21	42	nd	nd	nd	nd	nd	nd
5b	T	zeolite HT (3 h)	20	400	16	24	nd	nd	nd	nd	nd	nd
6a	C	sand	20	400	208	192	31	19	53	27	16	19
6b	T	zeolite HT (17 h)	20	400	25	26	nd	nd	nd	nd	nd	nd
7a	C	sand	20	400	283	278	31	16	57	17	12	11
7b	T	zeolite HT (17 h)	20	400	17	21	nd	nd	nd	nd	nd	nd
8a	C	sand	20	400	36	38	5	5	11	nd	nd	nd
8b	T	zeolite HT (17 h)	20	400	19	22	nd	nd	nd	nd	nd	nd
		filter[b]	zeolite size[e]		single stream pilot scale smoker collected in rapeseed oil (μg/L)							
9a	C	empty			2	5	1.6	0.3	2.1	1.3	0.7	0.8
9b	T	zeolite AR	4		1	3	1.5	0.6	<0.5	3.4	1.7	1.7
9c	T	zeolite HT (3 h)	4		1	1	0.4	0.2	<0.2	0.4	<0.2	0.4
10a	C	empty			8.5	14	4.3	1.4	3.7	0.81	<0.5	1.7
10b	T	zeolite HT (3 h)	5		8.2	14	2.9	1.3	2.8	0.75	<0.5	1.0
10c	T	zeolite HT (3 h)	3		3.3	5.7	2.1	0.92	1.4	0.51	<0.5	0.89
10d	T	zeolite HT (3 h)	2		<0.5	<0.5	<0.5	<0.5	<0.5	<0.5	<0.5	<0.5
		filter[b]	filter comp[f]		full scale smoking chamber collected in rapeseed oil (μg/L)							
11a	C	empty			51	80	29	6.5	20	4.8	<1	8.9
11b	T	zeolite HT (3 h)	10%		32	57	16	4.2	11	3.4	<1	4.1
11c	T	zeolite HT (3 h)	20%		20	33	12	3	7.2	2.2	<1	3.7
11d	T	zeolite HT (3 h)	30%		8.5	15	3.9	1.3	2.4	0.78	<1	0.96
11e	T	zeolite HT (3 h)	40%		7.8	11	4.4	2.4	3.7	0.83	<1	0.92
12a	C	empty			63	92	31	5.9	23	8.9	<1	10
12b	T	zeolite HT (3 h)	30%		16	24	5.4	1.6	3.3	0.98	<1	1.5
12c	T	zeolite HT (3 h)	40%		18	30	11	1.3	5.5	2	<1	3.1
12d	T	zeolite HT (3 h)	50%		9.5	16	5	0.9	2.4	0.91	<1	1.4

[a]EU regulated PAH4

[b]Filter T = smoke treated with zeolite, C = control with either sand dispersed through antibumping granules (dual stream processor) or nothing (single stream processor and smoking chamber). The zeolite was used as received (AR) or after activation (HT) by heating at 270 °C for the specified time (h)

[c]Total run time (h)

[d]Flow rate through rig (ml/min)

[e]Zeolite grain size as defined in methods section

[f]Composition of filter (% zeolite)

nd = not detected, limit of detection estimated at 5–10 ng/L, % coefficient of variation of the method determined at ~10%

Table 3 Changes in the Relative Peak Areas of Selected Aroma Compounds in Smoked Oil when Different Filter Treatments were Used to Produce the Smoke, and in Tomato Ketchup Dosed with 2% of the Corresponding Oils

compound	identification		smoked oils					tomato ketchup with smoked oils (2%)				
	LRI DB5[c]	LRI Wax[d]	none[a]	coarse[a] size 5	medium[a] size 3	fine[a] size 2	sig[b]	none[a]	coarse[a] size 5	medium[a] size 3	fine[a] size 2	sig[b]
2-furancarboxaldehyde	835	1382	100 d[e]	103 c	114 b	118 a	***	100 b	101 b	115 a	117 a	***
2-furanmethanol	854	1575	100 b	119 a	92 c	74 d	***	100 a	105 a	88 a	78 a	ns
1-(2-furanyl)ethanone	913	1424	100 b	91 c	111 a	98 b	***	100 b	89 c	117 a	96 b	***
5-methyl-2-furancarboxaldehyde	967	1498	100 a	87 b	104 ab	86 a	ns	100 b	87 c	103 a	101 c	***
phenol	977	1927	100 a	71 b	94 a	31 c	***	100 a	70 c	94 b	32 d	***
benzofuran	1006	1426	100 b	60 c	112 a	62 c	***	100 b	56 c	116 a	63 c	***
3-methylcyclopentane-1,2-dione	1031	1749	100 a	81 b	56 c	14 d	***	100 a	81 a	51 b	12 c	***
1-(5-methyl-2-furanyl)ethanone	1041	1424	100 a	69 b	98 a	51 c	**	100 a	70 b	104 a	52 c	***
benzeneacetaldehyde	1050	1557	100 a	80 b	72 b	49 c	***	100 a	81 b	88 ab	74 b	*
2-methylphenol	1053	1923	100 a	68 b	88 ab	31 c	***	100 a	68 c	89 b	31 d	***
3/4-methylphenol	1073	2013	100 a	65 b	81 ab	18 c	***	100 a	65 c	82 b	19 d	***
2-methoxyphenol	1094	1783	100 a	73 b	86 ab	49 c	**	100 a	71 c	86 b	47 d	***
1-(5-methyl-2-furanyl)propanone	1134	1605	100 a	63 bc	87 ab	42 c	**	100 a	64 b	89 a	41 c	***
4-methyl-2-methoxyphenol	1199	1880	100 a	68 b	71 b	27 c	***	100 a	69 b	73 b	29 c	***
4-ethyl-2-methoxyphenol	1287	1953	100 a	63 b	60 b	20 c	***	100 a	64 b	64 b	21 c	***
5-butyl-4-methyldihydrofuran-2(3*H*)-one[f]	1298	1818	100 a	65 b	56 b	25 c	***	nd	nd	nd	nd	
4-ethenyl-2-methoxyphenol	1322	2139	100 a	62 b	27 c	9 c	***	100 a	65 b	29 c	9 d	***
5-butyl-4-methyldihydrofuran-2(3*H*)-one[f]	1332	1889	100 a	64 b	46 c	13 d	***	100 a	63 b	46 c	13 d	***
1,3-dimethoxy-2-hydroxybenzene	1358	2203	100 a	74 b	32 c	6 d	***	100 a	73 b	38 c	5 d	***
2-methoxy-4-prop-2-enylphenol	1367	2111	100 a	67 b	40 c	13 d	***	na	na	na	na	
4-hydroxy-3-methoxybenzaldehyde	1406	2556	100 a	71 a	30 b	4 b	**	100 a	72 a	30 b	nd b	**
(*Z*)-2-methoxy-4-(prop-1-enyl)phenol	1409	2195	100 a	58 ab	39 b	nd c	***	na	na	na	na	
4-hydroxy-3-methoxybenzoic acid	1428		100 a	76 b	15 c	2 c	***	100 a	80 b	22 c	nd c	
(*E*)-2-methoxy-4-(prop-1-enyl)phenol	1431	2299	100 a	50 b	25 bc	nd c	***	na	na	na	na	***

total peak area for these compounds	100	93	104	93	100	86	102	83

[a]Zeolite filter and grain size used in the smoking

[b]Significance of difference between samples, obtained from ANOVA; ns, no significant difference between means ($p > 0.05$); * significant at the 5% level; ** significant at the 1% level; *** significant at the 0.1% level

[c]Linear retention index (LRI), calculated from a linear equation between each pair of straight chain alkanes C5–C30 on a DB5 column

[d]LRI on Stabilwax DA column

[e]Relative amount (mean n=3) compared to control (100), within each row values with the same letter are not significantly different using Fisher's least square difference at $p=0.05$

[f]Unspecified mix of isomers

nd = not detected

na = not analysed because compound present in large amounts in the control tomato ketchupas part of spice blend.

Table 4 Changes in the Relative Peak Areas of Selected Aroma Compounds in Smoked Oil with Different % of Zeolite Incorporated in The Filter Used to Produce the Smoke

compound	identification LRI DB5[c]	identification LRI Wax[d]	ID[b]	% zeolite in filter (experiment 12)[a] none	30%	40%	50%
2-furancarboxaldehyde	835	1382	A	100[e]	117	117	119
2-furanmethanol	854	1575	A	100	39	26	20
1-(2-furanyl)ethanone	913	1424	A	100	90	75	72
5-methyl-2-furancarboxaldehyde	967	1498	A	100	116	94	91
phenol	977	1927	A	100	76	49	46
benzofuran	1006	1426	A	100	49	73	87
3-methylcyclopentane-1,2-dione	1031	1749	A	100	83	41	28
1-(5-methyl-2-furanyl)ethanone	1041	1424	A	100	79	52	45
benzeneacetaldehyde	1050	1557	A	100	110	89	89
2-methylphenol	1053	1923	A	100	61	38	34
3/4-methylphenol	1073	2013	A	100	59	37	31
2-methoxyphenol	1094	1783	A	100	94	64	59
1-(5-methyl-2-furanyl)propanone	1134	1605	A	100	48	38	33
4-methyl-2-methoxyphenol	1199	1880	A	100	104	67	58
4-ethyl-2-methoxyphenol	1287	1953	A	100	79	46	35
5-butyl-4-methyldihydrofuran-2(3*H*)-one[f]	1298	1818	A	100	75	47	39
4-ethenyl-2-methoxyphenol	1322	2139	A	100	79	48	40
5-butyl-4-methyldihydrofuran-2(3*H*)-one[f]	1332	1889	A	100	67	43	38
1,3-dimethoxy-2-hydroxybenzene	1358	2203	A	100	57	29	20
2-methoxy-4-prop-2-enylphenol	1367	2111	A	100	84	54	45
4-hydroxy-3-methoxybenzaldehyde	1406	2556	A	100	124	72	52
(*Z*)-2-methoxy-4-(prop-1-enyl)phenol	1409	2195	C	100	80	49	38
4-hydroxy-3-methoxybenzoic acid	1428		B	100	62	29	18
(*E*)-2-methoxy-4-(prop-1-enyl)phenol	1431	2299	A	100	86	57	44
total peak area of these compounds				100	94	97	77

[a] Zeolite (%) incorporated into the filter during smoking

[b] A indicates MS and LRI agree with those of the authentic compound run under the same conditions on both columns, B indicates agreement with authentic compound on DB5 column, C indicates agreement on both columns with minor isomer present in authentic compound

[c] Linear retention index (LRI), calculated from a linear equation between each pair of straight chain alkanes C_5–C_{30} on a DB5 column

[d] LRI on Stabilwax DA column

[e] Relative amount compared to control, control = 100

Table 5 Mean Scores for Sensory Attributes of Tomato Ketchup Dosed with Smoked Oil

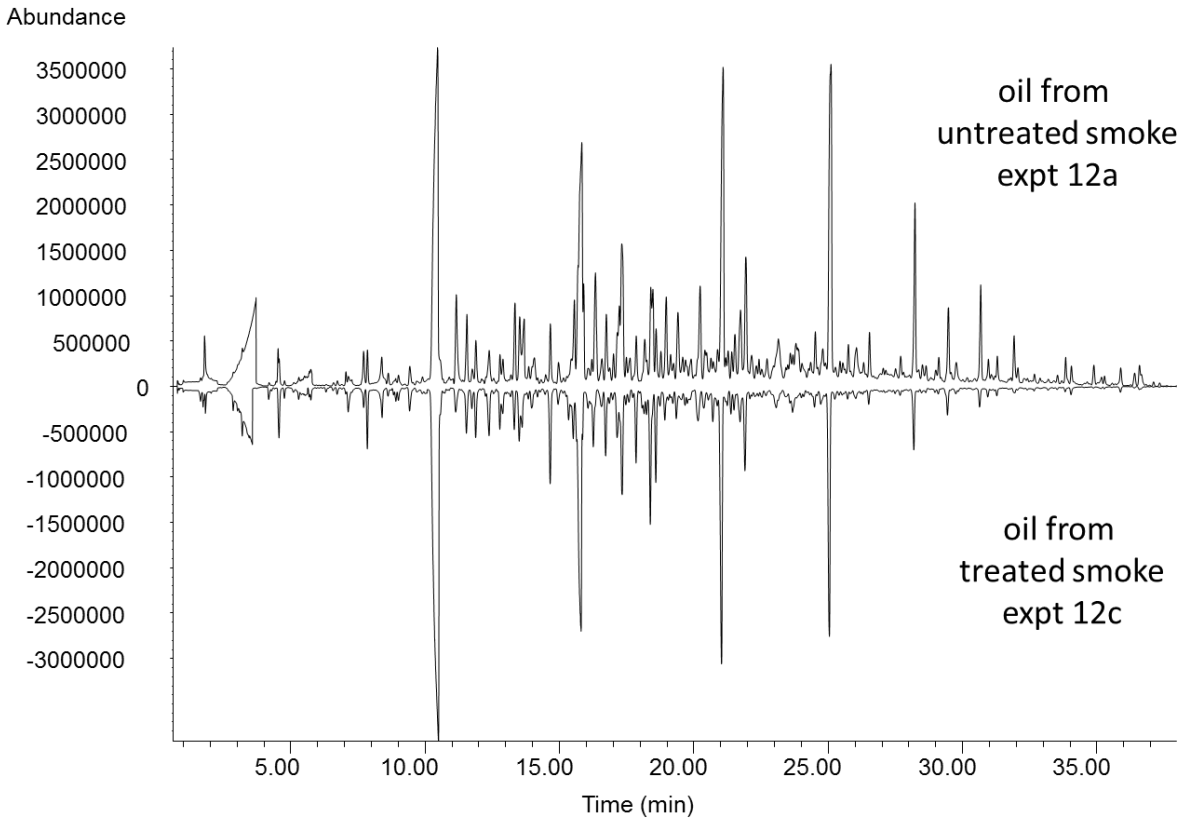
attribute	zeolite grain size					
	control exp 10a	size 5 expt 10b	size 3 expt 10c	size 2 expt 10d	LSD[a]	prob[b]
aroma						
smoky bonfire (burnt paper)[c]	32 ab [d]	34 ab	36 a	26 b	9	0.14
smoked mackerel (smoked mackerel)	24 a	22 ab	15 b	18 ab	9	0.16
coal tar/diesel (coal tar soap)	8	7	8	6	3	0.36
rubber (rubber tube)	8	13	8	14	8	0.32
melted plastic	6	9	6	7	5	0.44
tomato	20	20	21	24	5	0.20
juicy/fruity	5 b	4 b	7 ab	9 a	4	0.13
sweet	21 b	23 b	22 b	27 a	4	0.03
taste						
sweet taste	23	26	25	26	7	0.74
acidic	23	24	21	22	5	0.75
salty taste	10 b	13 a	12 ab	11 ab	3	0.19
umami	21	20	22	23	6	0.70
flavor						
smoky bonfire	28	28	29	27	7	0.95
smoked mackerel	20	15	14	15	8	0.38
coal tar/diesel	9	10	13	10	4	0.23
rubber	15	21	15	22	11	0.50
melted plastic	19	20	23	22	7	0.70
tomato	22	18	18	21	5	0.22
juicy/fruity	7	3	4	5	4	0.22
mouthfeel						
drying mouthfeel	21	21	21	21	5	0.99
tingle	4	4	3	4	3	0.91
throat burn	4 b	3 b	7 a	3 b	3	0.05
after-effects						
sweet	18	16	18	20	5	0.49
bitter	9	10	9	9	4	0.86
rubber	13	18	12	15	8	0.56

[a]Fisher's least significance difference at $p = 0.05$

[b]Probability, obtained from ANOVA, that there is a difference between means

[c]Aroma references

[d]Mean of two replicate assessment for each assessor (18 replicates in total), means labelled with the same letters (or not labelled) are not significantly different $p < 0.05$.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-1500000
-2000000
-2500000
-3000000
oil from
untreated smoke
expt 12a
oil from
treated smoke
expt 12c
5.00
10.00
15.00
20.00
25.00
30.00
35.00
Time (min)

TOC GRAPHIC