4.2.1 *Intake data*

The total annual volume of production of the 47 substances in this group destined for use as flavouring agents is approximately 200 tonnes in Europe and 1700 tonnes in the USA. On the basis of the reported annual volumes of production, the estimated total daily per capita intakes of these substances resulting from their use as flavouring agents is 28mg in Europe and 300mg in the USA.

Fumaric acid (no. 618) and (–)-malic acid ((2*S*)hydroxybutanedioic acid; no. 619) account for approximately 59% of the total daily per capita intake of these 47 substances in Europe and 88% in the USA. The estimated daily per capita intake of fumaric acid resulting from its use as a flavouring agent is approximately 0.9mg in Europe and 219mg in the USA. The estimated daily per capita intake of (–)-malic acid resulting from its use as a flavouring agent is about 16mg in Europe and 58mg in the USA. The intake of each substance in the group in μg per day in Europe and the USA is reported in Table 3.

Of the 47 substances evaluated, 25 have been detected as natural components of traditional foods.

4.2.2 *Absorption, metabolism and elimination*

Studies on the absorption, metabolism and elimination of aliphatic primary alcohols, aldehydes, carboxylic acids, acetals and esters with additional oxygenated functional groups show that these substances are readily hydrolysed and absorbed and are completely metabolized. Many of these substances or their metabolites are endogenous in humans.

The majority of the substances in this group are esters or diesters and are expected to undergo hydrolysis to their corresponding saturated linear or branched-chain aliphatic primary alcohols or branched-chain hydroxy or keto alcohols. The presence of a second oxygenated functional group has little, if any, effect on the hydrolysis of these esters. β-Keto acids and derivatives such as acetoacetic acid readily undergo decarboxylation and, together with α-keto and α-hydroxyacids, yield breakdown products which are incorporated into normal biochemical pathways. The γ-keto acids and related substances may undergo complete or partial β-oxidation to yield metabolites, which are eliminated in the urine. The ω-substituted derivatives are readily oxidized and/or excreted in the urine. The simple aliphatic di- and tricarboxylic acids either occur endogenously in humans or are structurally related to endogenous substances. They are metabolized through the fatty acid β-oxidation pathway or the tricarboxylic acid cycle.

4.2.3 *Application of the Procedure for the Safety Evaluation of Flavouring Agents*

Step 1

In applying the Procedure for the Safety Evaluation of Flavouring Agents (Fig. 1) to the above-mentioned aliphatic primary alcohols, aldehydes, carboxylic acids, acetals and esters containing additional oxygenated functional groups, the Committee assigned all 47 substances to structural class I.

Step 2

Metabolic data on individual members of the group are limited, but the common structural features and common pathways of metabolism allow some general conclusions to be drawn about the likely metabolic fate of these agents. Of the 47 substances in this group, 14 occur endogenously in humans, and 28 are esters or diesters that would be expected to be metabolized to innocuous products. There was evidence that the remaining substances in the group, including acetals, derivatives of β-keto and β-hydroxy acids, γ-keto and γ-hydroxy acids and aliphatic di- and tricarboxylic acids, are also metabolized to innocuous products. The evaluation of all of the substances in this group therefore proceeded via the left-hand side of the decision-tree.

Step A3

The estimated daily per capita intakes of 41 of the 47 substances in this group were below the threshold for substances in class I (1800μg), indicating that they are of no safety concern when used at current levels of intake. The estimated daily per capita intakes of the remaining six substances, ethyl acetoacetate (no. 595), fumaric acid (no. 618), (–)-malic acid (no. 619), a mixture of (+)-, (–)-, (+/–)- and meso-tartaric acid (mixture of (+)-, (–)-, (+/–)- and meso-2,3-dihydroxybutanedioic acid; no. 621), adipic acid (hexanedioic acid; no. 623) and triethyl citrate (triethyl 2-hydroxy-1,2,3-propanetricarboxylate; no. 629) were above the threshold for class I. The evaluation of these six substances therefore proceeded to step A4.

Step A4

Four of the six substances for which the intake exceeded the threshold of concern for class I are endogenous in humans. Three of these four substances, namely, fumaric acid (no. 618), (–)-malic acid (no. 619) and triethyl citrate (no. 629), are components of the tricarboxylic acid cycle. The fourth substance, ethyl acetoacetate (no. 595), is expected to be hydrolysed to acetoacetic acid, which is endogenous in humans and is formed from the condensation of two acetyl CoA units in the fatty acid pathway.

The remaining two substances, the mixture of (+)-, (–)-, (+/–)- and meso-tartaric acid (no. 621) and adipic acid (no. 623), are not endogenous and are not predicted to be metabolized to endogenous products. The evaluation of these substances therefore proceeded to step A5.

Step A5

The NOEL for tartaric acid (no. 621) in a 2-year toxicity study in rats was 1200 mg/kg of body weight per day, the highest dose tested, which provides an adequate margin of safety (>10000 in Europe and >1000 in the USA) when compared with the current levels of estimated intake of this substance. A NOEL was not available for adipic acid (no. 623), but the NOEL for a structurally related substance, dibutyl sebacate (dibutyl decanedioate; no. 625), in a 2-year study in rats was 6200 mg/kg of body weight per day, which provides an adequate margin of safety (>1×10^7 in Europe and >10000 in the USA), when compared with the current levels of estimated intake of adipic acid. Therefore, these substances were determined to be of no safety concern when used at current levels of estimated intake.

Table 3 summarizes the stepwise evaluation of the 47 aliphatic primary alcohols, aldehydes, carboxylic acids, acetals and esters containing additional oxygenated functional groups used as flavouring agents.

4.2.4 *Consideration of combined intakes*

In the unlikely event that foods containing all 47 aliphatic primary alcohols, aldehydes, carboxylic acids, acetals and esters containing additional oxygenated functional groups were consumed simultaneously on a daily basis, the estimated total daily per capita intake of these substances in Europe and the USA would exceed the threshold for flavouring agents in class I. All of these substances are expected to be efficiently metabolized via well known biochemical pathways to innocuous metabolic and/or endogenous substances; in the opinion of the Committee, the endogenous levels of these metabolites would not give rise to perturbations outside the physiological range. Accordingly, even a combined theoretical intake would be of no safety concern.

4.2.5 *Conclusions*

The Committee concluded that the substances in this group would not present safety concerns at the current levels of estimated intake.

No toxicity data were required for application of the Procedure to 45 of the 47 substances in this group. For the remaining two substances, the mixture of (+)-, (–)-, (+/–)- and meso-tartaric acid (no. 621) and

adipic acid (no. 623), the toxicity data were consistent with the results of the safety evaluation using the Procedure.

The ADIs for fumaric acid (no. 618) and its salts and for triethyl citrate (no. 629) were maintained at the present meeting.

A monograph summarizing the safety data on this group of flavouring agents was prepared.

5. Peanut oil and soya bean oil

The allergenicity of foodstuffs has not previously been considered by the Expert Committee. Allergens in food have been considered by the Codex Committee on Food Labelling on a number of occasions since 1993, and that Committee has issued a list of foods and food ingredients known to cause allergy. The list, with modifications, was revised at an FAO Technical Consultation in 1995 (*11*). After debate in the Codex Committee (*8*), the list was forwarded at step 8 for adoption by the Codex Alimentarius Commission. The Joint FAO/WHO Expert Committee on Food Additives was asked by the Codex Committee on Food Labelling (*8*) to provide scientific advice on this issue and to develop criteria for identifying food products on the list for which labelling of the food source is unnecessary (see section 2.4). An ad hoc Panel on Food Allergens was convened for this purpose; its report is attached as Annex 4.

The criteria identified by the Panel included:

- evidence from a double-blind clinical study that challenge with the specific product does not elicit allergic reactions in a group of patients with clinical allergy to the parent foodstuff, and
- the existence of specifications for the product and its manufacturing process that ensure that the process yields a consistently safe product.

The Panel identified only two foodstuffs that it considered may currently fulfil the criteria for inclusion in a list of products for which labelling of the allergen-containing food source is not necessary: refined peanut oil and refined soya bean oil. The Panel recommended that these substances be considered by the Committee at its present meeting.

The Committee recognized that the allergenicity of vegetable oils is heavily dependent on the processes used to extract and refine the oils. It was aware that several steps are involved in the refining process and that different producers may use variations of the basic procedures. In addition, in clinical trials of the oils, the mode of administration, the

allergic sensitivity of the subjects to the source material and the use of double-blind protocols can affect the outcome.

The Committee was aware of a number of studies conducted using the double-blind procedure in which people sensitive to peanuts had been challenged with various grades of peanut oil. In a study from the USA, all 10 male and female patients with known sensitivity to peanuts gave a positive reaction in skin-prick tests with peanut extracts and were found to have elevated serum titres of antibodies to peanut allergens. A cross-over challenge with commercial peanut oil and olive oil did not elicit adverse reactions, although the Committee noted that the use of gelatin capsules to administer the oils may have masked reactions of the lips and oral cavity.

In a double-blind study in France, 11 children with symptoms possibly due to allergies were found to react to skin-prick tests with peanut or peanut protein extracts. Four of the patients reacted to an oral challenge with peanut oil. The origin and grade of the peanut oil used were not defined, and the Committee recognized that it may have been obtained before adoption of a revised code of practice for the refining of vegetable oils by the food industry in continental Europe. Earlier studies of infants in France had suggested that peanut oils used as a vitamin carrier or in infant formulas may have contained allergenic proteins.

The results of a randomized, double-blind, cross-over challenge study involving a group of 62 patients from the United Kingdom who had reacted to skin-prick tests with peanut extracts were reviewed. None of the subjects reacted to challenge with refined peanut oil, although six reacted to challenge with crude peanut oil. Of the 62 patients, 60 also reacted to an oral challenge with peanuts. The Committee considered that the study was well designed, with adequate statistical power, and recognized the value of confirming the sensitivity of the subjects to peanuts after the double-blind challenge had been completed. Nevertheless, although the study provided adequate evidence for a lack of allergenicity of the oil used, appropriate descriptions of the manufacturing process and the consequent specifications of the oil were not provided and the results cannot be extrapolated to other oils.

The Committee also reviewed the results of a double-blind, cross-over challenge study of the allergenicity of soya bean oils involving seven individuals who had experienced allergic reactions up to 10 years previously. All the individuals had positive reactions to a skin-prick test with soya bean extract. The titres of serum immunoglobulin E (IgE) binding to soya bean proteins were increased in six of the seven patients. None of the subjects reacted to increasing volumes of

hydrogenated, partially hydrogenated or cold-pressed soya bean oils, although the Committee noted that the use of gelatin capsules to administer the oils may have masked reactions of the lips and oral cavity. Although the study provided some evidence that the oils used were not allergenic, appropriate descriptions of the manufacturing process and the consequent specifications of the oils were not provided, and the results cannot be extrapolated to other oils.

The Committee noted the absence of clear descriptions of the processes that had been used to refine the peanut and soya bean oils tested. Additionally, comparable data on the protein content of those oils that were clinically tested were not available. Furthermore, the Committee expressed reservations about the quality and the lack of validation of the analytical methods used to determine the concentration of residual protein in the oils. In view of these considerations, the Committee concluded that refining processes that would consistently yield safe products have not been defined.

The Committee therefore concluded that the results of studies of immunological tolerance to representative refined peanut and soya bean oils would be required for a full evaluation. Such studies should provide extensive information on a wide range of oils, representing refining procedures used throughout the world. Full descriptions of the refining processes used and evidence for a lack of allergenicity of the oils as determined by appropriately designed clinical studies should be provided. Information on the nature and quantity of protein in the oils would be essential for defining the level of refinement of the oils tested, with a view to identifying representative oils.

6. Contaminants

The Committee evaluated one contaminant for the first time and re-evaluated two contaminants considered at previous meetings. The results of the evaluations are summarized in Annex 2.

6.1 Lead

The Committee first evaluated lead at its sixteenth meeting (Annex 1, reference *30*), when a provisional tolerable weekly intake (PTWI) of 3mg per person, equivalent to 50μg/kg of body weight, was established. This PTWI was reconfirmed at the twenty-second meeting (Annex 1, reference *47*). At its thirtieth meeting (Annex 1, reference *73*), the Committee assessed the health risks of lead to infants and children and established a PTWI of 25μg/kg of body weight for this population group. The Committee again evaluated lead at its forty-

first meeting (Annex 1, reference *107*), when the previous PTWI of 50μg/kg of body weight for adults was withdrawn and the existing PTWI of 25μg/kg of body weight for infants and children was reconfirmed and extended to all age groups. The review of the health effects of lead at the forty-first meeting was based on an assessment of lead that had been performed by an IPCS Task Group, which was published as Environmental Health Criteria, No. 165 (*12*).

At its present meeting, the Committee was requested to assess the health risks of dietary exposure of infants and children to lead, with special emphasis on the most critical effect, which was considered to be impaired neurobehavioural development. The Committee considered several models that had been developed to define the relationship between the effects of current levels of exposure to lead and the impact on health that might be anticipated from a reduction in exposure. The PTWI was not reconsidered and was maintained at its present value.

The most widely used biomarker of exposure to lead is the concentration in blood (blood lead concentration, measured in μg/dl). The most critical effect of lead at low concentrations is reduced cognitive development and intellectual performance in children. A number of studies in which various tests of behavioural performance were used have shown an association between blood lead concentration and reduced intelligence quotient (IQ) in children exposed pre- and postnatally. The effects of confounding variables and limits to the precision of analytical and psychometric measurements increase the uncertainty of any estimate of the effect of blood lead concentrations below 10–15μg/dl. If a threshold does exist, it is unlikely to be detected because of these limitations; nevertheless, there was some evidence of an association between cognitive deficits and blood lead concentrations below 10μg/dl.

6.1.1 *Exposure*

Exposure to lead can occur as a result of ingestion of lead in foodstuffs and water and from other sources, such as air. All these sources make important contributions. Although the assessment reported here was limited to dietary intake, a complete analysis would require the inclusion of all sources of lead.

The Committee reviewed data on lead intake in 25 countries and assessed several diets on the basis of the assumption that the Global Environment Monitoring System–Food Contamination Monitoring and Assessment Programme (GEMS/Food) regional diets contain "typical" levels of lead in the food categories for which limits have

been proposed by the Codex Committee on Food Additives and Contaminants. The GEMS/Food diets have been used by the Codex Committee on Pesticide Residues and other committees to estimate intakes of pesticides and contaminants since 1987. At its present meeting, the Expert Committee also used the regional diets to estimate lead intake under three sets of assumptions:

- All foods contain lead at the limits proposed by the Codex Committee on Food Additives and Contaminants.
- All foods contain lead at a "typical" average concentration.
- All foods contain lead at "typical" high levels.

When levels at the limits proposed by the Codex Committee on Food Additives and Contaminants were used in the assessment, the estimated intakes were 13–20 μg/kg of body weight per week. The "typical" average and high levels were derived from monitoring studies in the USA and were similar to those reported in other countries. The intakes ranged from 1 to 2 μg/kg of body weight per week for "typical" lead levels and from 2 to 4 μg/kg of body weight per week for "typical high" levels. The narrow ranges in estimated intake reflect the fact that the data submitted to the Expert Committee did not include foodstuffs that contained particularly high levels of lead, and no food group predominated. Virtually no data were submitted on foods containing levels above the limit proposed by the Codex Committee. The Expert Committee noted that similar intakes were estimated on the basis of the three sets of assumptions for the five GEMS/Food regional diets.

The Expert Committee consulted the GEMS/Food database and found that foods that were sampled in the 1980s contained much higher concentrations of lead than those measured recently and decided to base its conclusions on current data.

The potential intake of lead by children was reported by seven countries in which the general food supply, infant formulas and other foods commonly consumed by children had been monitored. Several countries provided information on consumption of foods by children. On the basis of this information, the estimated range of intake of lead by children was 0.6–30 μg/kg of body weight per week, which was generally two to three times the intake by adults in the same country when evaluated on the basis of body weight.

Tap water is a significant potential source of lead intake, particularly for bottle-fed infants, but the data submitted were inadequate to permit estimation of the range of levels found.

6.1.2 *Quantitative risk assessment*

Exposure assessment

Several simulation models were developed to estimate the distributions of dietary lead intake in regional diets. The first involved a scenario in which the regional populations covered by the GEMS/Food database consumed food with lead concentrations corresponding to those found in a survey conducted in the USA. The second was designed to evaluate the impact of inclusion of a food commodity from a source with a much higher distribution of lead superimposed on the background of other regional dietary and non-dietary exposures. In a third simulation, the effects of several theoretical regulatory interventions on dietary intake were evaluated.

Estimates of the blood lead concentrations from dietary intake

In order to predict the biological effects of lead intake, the Committee used simple empirical models to relate the concentration of lead in the diet to changes in the biomarker, the concentration of lead in blood. Most of the older data refer to relatively constant exposure to lead, usually as a consequence of contamination of drinking-water by lead plumbing. These data have limited predictive value for levels of exposure that result in blood lead concentrations higher than 25 µg/dl. In addition, other dietary components or atypical physiological states may alter the rate of absorption of lead from the intestine to the blood.

In order to infer a relationship between ingestion of lead and an increase in the blood lead concentration of infants and young children, data from studies of bottle-fed infants were fitted into several models. Reasonable fits required the assumption that a lead concentration in drinking-water of zero corresponds roughly to a blood lead concentration of 15 µg/dl, perhaps reflecting exposure from the environment or in utero. The results attributable to dietary intake of lead by infants correspond roughly to a change in blood lead concentration of 0.05–0.1 µg/dl per µg of lead intake per kg of body weight per day. For a 10-kg infant, this corresponds to a blood lead concentration of 0.5–1.0 µg/dl per µg of lead in the diet per day.

The Committee used data from another study to calculate the relationship between the blood lead concentration of pregnant women and intake of lead from drinking-water. The sample size was large enough to allow characterization of variation in the population. When the raw data were introduced into several simple models, all showed poor fits. When concentrations of lead in drinking-water below 300 µg/l were fitted separately, a linear model with a lognormal population distribution produced a very good fit. A background blood lead