European Commission

Food science and techniques

Reports of the Scientific Committee for Food

(43rd series)

OPINIONS OF THE SCIENTIFIC COMMITTEE FOR FOOD ON:

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Starch aluminium octenyl succinate (SAOS)

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Directorate-General Consumer Policy and Consumer Health Protection

1998

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Cataloguing data can be found at the end of this publication.

Luxembourg: Office for Official Publications of the European Communities, 1999

ISBN 92-828-5887-1

Printed in Belgium

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For their valuable and kind assistance with the following studies, the Scientific Committee for Food wishes to thank:

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C. Boudene	Paris (F)
J. Carstensen	Novo Nordisk A/S, Måløv (DK)
L. Castle	Central Food Laboratory, Norwich (UK)
R. Crebelli	Istituto superiore di sanità. Rome (I)
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Abbreviations used in this report

ADI	acceptable daily intake
b.w.	body weight
ED	effective dose
FSMP	foods for special medical purposes
LEL	Lowest effective level
MTDI	maximum tolerable daily intake
NOAEL	no observable adverse effect level
NS	not specified
PMTDI	provisional maximum tolerable daily intake
PTWI	provisional tolerable weekly intake
R	restriction indicated. If not otherwise indicated R: x mg/kg means mg/kg of food or food simulant.
TDI	tolerable daily intake
t-ADI	temporary ADI
t-TDI	temporary TDI

US-ATSDR	USA Agency for Toxic Substances and Disease Registry
BIBRA	British Industrial Biological Research Association (UK)
CAS N.	Chemical Abstract Service Registry Number (USA)
CIVO-TNO	Central Institute for Nutrition and Food Research (NL)
EM	electron microscopy
FAO	Food and Agriculture Organisation (UN)
HRC	Huntingdon Research Centre (UK)
IARC	International Agency for Research on Cancer (F)
JECFA	Joint FAO/WHO Expert Committee on Food Additives (UN)
NTP	national toxicology program (USA)
RIVM	National Institute for Public Health and Environmental Protection (NL)
SCC	Scientific Committee for Cosmetology
SCF	Scientific Committee for Food
US-NRC	USA National Research Council
WHO	World Health Organisation (UN)

ARSENIC, BARIUM, FLUORIDE, BORON AND MANGANESE IN NATURAL MINERAL WATERS

(expressed on 13 December 1996)

Terms of reference

To advise on the acceptability from the public health point of view of certain substances in natural mineral waters. The substances the Committee is asked to consider are those natural constituents for which the levels requested by the European industry (*) are above the levels in the drinking water directive (*) or in its proposed amendment (*).

Background

Directive 80/777/EEC (*) on natural mineral waters defines 'natural mineral water' as:

> *'...microbiologically wholesome water originating in an underground water table or deposit and emerging from a spring tapped at one or more natural or bore exits. Natural mineral water can be distinguished from ordinary drinking water, notably:*
> - *by its nature, which is characterised by its mineral content, trace elements and other constituents;*
> - *by its original state.'*

The directive states that 'natural mineral waters are protected from all risk of pollution in order to preserve intact these characteristics'. However, this directive of 1980 does not lay down individual limits for the minerals, trace elements or contaminants that may be found in natural mineral waters.

The Committee was informed that a proposal (*) to update Directive 80/777/EEC was in the process of adoption to take into account scientific and technical progress and the mandate from the European Council for rationalising the directive. Under this new directive (*), the SCF must

(*) The European organisation of producers of natural mineral waters (GESEM-UNESEM)

(*) Council Directive 80/778/EEC of 15 July 1980 relating to the quality of water intended for human consumption (OJ L 229, 30.8.1980, p. 11)

(*) Proposal for a Council directive concerning the quality of water intended for human consumption (COM(94) 612 final, OJ C 131, 30.5.1995, p. 5).

(*) Council Directive 80/777/EEC of 15 July 1980 on the approximation of the laws of the Member States relating to the exploitation and marketing of natural mineral waters (OJ L 229, 30.8.1980, p. 1)

(*) European Commission Proposal for a European Parliament and Council directive amending Council Directive 80/777/EEC on the approximation of the laws of the Member States relating to the exploitation and marketing of natural mineral waters (COM(94) 423 final, 94/0235 (COD), OJ C 1, 4.1.1995, p. 22)

(*) Directive 96/70/EC of the European Parliament and of the Council of 28 October 1996 amending Council Directive 80/777/EEC on the approximation of the laws of the Member States relating to the exploitation and marketing of natural mineral waters (OJ L 299, 23.11.1996, p. 26).

be consulted in questions relating to public health. The European industry (*) presented to the Commission a list of proposed limits for these substances which included a number which exceeded the limits in Directive 80/778/EEC (*) or in its proposed amendment(*).

Evaluation

Introduction

In preparing this report, the Committee has made use of reviews such as the SCF report on nutrient and energy intakes for the European Community (27), the WHO guidelines for drinking water quality (31, 32, 34), IARC and US-NRC, US-ATSDR, Eectoc and RIVM reports and recent papers.

The Committee was provided with information on the consumption of natural mineral waters in the Community. However, this information does not allow estimation of intakes by high level consumers of natural mineral waters. Some natural mineral waters are increasingly consumed as a replacement for tap water. In the absence of other data, the Committee considered that the conventional value of 2 litres per day and per person for the consumption figure, as used by the EU and WHO for risk assessments relating to drinking water, should also be applied to natural mineral waters. The Committee is aware that this is a conservative approach.

In making its risk assessment, the Committee has recognised that drinking water is not the sole dietary source of human exposure to the substances under consideration. For boron and manganese, the Committee has, in the absence of reliable exposure data, used the WHO default value of 10 % (31) as the proportion of the TDI (or equivalent value established in this opinion) which can be allocated to natural mineral waters. This approach could not be applied to arsenic which has been classified by the International Agency for Research on Cancer (IARC) as a human carcinogen (16). For barium and fluoride the evaluation is based on epidemiological data derived from studies involving consumption of drinking water containing known levels of these elements.

Individual substances

1. Arsenic (As)

Evaluation

Inorganic arsenic is an established carcinogen able to induce primary skin cancer and has been classified by IARC 1987 (16) in Group 1 (carcinogenic to humans). So far it has been found negative in animal carcinogenicity bioassays with one exception, but positive in tumour promotion studies. It has been found essentially non-mutagenic at gene level, but able to induce chromosomal aberrations and micronuclei in a variety of mammalian cells, including human cells. The mechanism of arsenic carcinogenic activity has not yet been clarified. Recent

epidemiological studies (7, 28, 3) seem to suggest that the carcinogenicity of arsenic requires the presence of other carcinogenic agents such as cigarette smoke.

All these facts seem to suggest that arsenic is an indirect carcinogen, with promoting and/or co-carcinogenic activity. The lack of knowledge of the precise mechanism of carcinogenic activity and the known problems in applying mathematical models makes the cancer-risk assessment for arsenic very difficult. Based on the increased incidence of skin cancer observed in Taiwan and by using a multistage model, WHO (32) has calculated that an excess lifetime skin cancer risk of 10^{-5} is associated to a concentration of 0.17 mg/l in drinking water. This value may, however, overestimate the actual risk due to a number of factors, among which is the possible indirect mechanism of arsenic carcinogenicity. In order to reduce the concentration of arsenic in drinking water, WHO has established a provisional guideline value of 10 µg/l. This value is in line with the proposed amendment of the drinking water directive (1) but it is below the maximum admissible concentration of 50µg/l in the existing directive (2).

Conclusion

In consideration of the fact that inorganic arsenic is an established human carcinogen, exposure should be as low as possible. For the time being an upper level of 10 µg/l in natural mineral waters seems reasonable and is in line with the proposed amendment (1) to the existing drinking water directive.

2. Barium (Ba)

Evaluation

Barium is not considered to be an essential element. Soluble salts of barium are known to be toxic. The acute toxic oral dose of barium chloride for humans is reported to be 0.2 -0.5 g and doses above 3 g are lethal.

Sub-chronic studies following oral exposure have been carried out in rats. The most relevant adverse effect reported was a rise in systolic blood pressure when barium was given in the drinking water at 100 mg/l (22, 23). Although even 10 mg/l induced some less marked increases in blood pressure, being only occasionally significant, a no-adverse-effect level of 0.5 mg/kg b.w./day was derived from that concentration, because the increases were deemed small enough not to constitute an adverse effect (34).

In a controlled human study, daily doses of barium up to 15 mg did not show effects on blood pressure or on the cardiovascular system (36). One retrospective epidemiological study in some communities in Illinois (USA) resulted in significantly higher age-adjusted death rates for 'all cardiovascular diseases' and 'heart disease' in the areas with high barium levels in the drinking water (2 - 10 mg/l) compared with low barium communities (< 0.2 mg/l) (5). However, this study was difficult to interpret and in a similar, better controlled, study by the same authors it was concluded that levels of barium in drinking water of 7.3 mg/l do not significantly elevate blood pressure levels in adult males or females (6).

Conclusion

Considering that the concentration of 7.3 mg/l drinking water does not affect blood pressure or incidence of cardiovascular diseases in humans and applying an uncertainty factor of 10 to account for intra-human variations, an upper level of 1 mg/l in natural mineral waters appears to be acceptable.

3. Fluoride (F)

Evaluation

In 1992 the SCF concluded that there does not appear to be a specific physiological requirement for fluoride and no specific recommendations were made (27). However, it was recognised that the element is beneficial to dental health at low intakes while on the other hand fluoride excess (fluorosis) is endemic in many parts of the world.

Fluoride has been subject to a series of acute, short-term, and long-term studies, but given the limited character of these animal studies, and the large body of data on the toxic effects of fluoride in humans, the latter data have priority in the derivation of long-term tolerable intakes for humans.

In humans, acute toxic effects have been reported at doses of 1–10 mg/kg b.w. with values of 14–140 mg/kg b.w. being reported for the acute lethal oral dose of soluble fluorides. The long-term adverse effect starts in its mild form at concentrations within the 'beneficial' range with a mild dental fluorosis prevalence of 12–33 % being reported for concentrations in drinking water of 0.9–1.2 mg/litre. The clinical picture of dental fluorosis in the mild form consists of the presence of opaque white areas on the teeth and is normally considered as a cosmetic effect rather than an adverse effect. Severe forms of this condition occur already at concentrations of 5–7 mg/litre. In such cases the tooth enamel can become brittle enough to fracture at incisal edges and cusp tips. Climate has been identified as a factor determining the degree to which dental fluorosis will develop. In areas with a temperate climate, manifest dental fluorosis occurs at concentrations above 1.5–2.0 mg/litre whereas in warmer areas, the same effect may be already present at lower concentrations i.e. 0.7 – 1.2 mg/litre. This may be attributed to greater water consumption in warmer climates (25, 9, 30, 29).

Skeletal fluorosis consists of adverse changes in bone structure due to continuous deposition of fluoride in the bone. The minimum dose required for production of skeletal fluorosis in its various degrees is not known exactly. However, various studies of population groups indicate that at levels below 4 mg/day there is no hazard of a significant degree of accumulation, while 6–20 mg/day causes skeletal fluorosis to some degree and the severe form, crippling skeletal fluorosis, requires a daily dose of 20–80 mg (25, 30, 2). Fluoride has been used in the past in the treatment of osteoporosis, however, clinical trials indicate that the effectiveness of this

treatment is questionable. Population studies on bone fracture rate in relation to fluoride in drinking water have also yielded inconclusive results (29).

Many mutagenicity studies are available, mostly carried out with NaF (29). It has been found negative in bacterial systems, and positive in cultured mammalian cells (at gene and chromosome level) only at cytotoxic concentrations, probably by an indirect mechanism. So far, no adequate *in vivo* data are available.

According to IARC (15) the limited animal data available were evaluated as inadequate. More recent NTP studies performed in rats and mice have shown an increased incidence of osteosarcomas only in male rats. This effect was evaluated by NTP as equivocal evidence (21, 29).

Numerous epidemiological studies have been carried out to investigate whether there is a relation between the occurrence of cancer and the exposure to fluoride via drinking water. IARC concluded that the studies provide inadequate evidence for carcinogenicity in humans (15, 16). More recent studies also have not supplied evidence that there is a relation between fluoride in drinking water and cancer mortality (25, 2).

Conclusion

On the basis of the data reviewed above, especially as concerns the occurrence of dental fluorosis at concentrations above 0.7 mg/l (warm climates) and 1.5 mg/l (temperate climates), the Committee has no reason to deviate from the level of 1.5 mg/l, as given in the proposed amendment (') to the existing drinking water directive, and concludes that this level should also apply to natural mineral waters.

4. Boron (B)

Evaluation

There are conflicting views about the essentiality of boron for man. In its report on nutrient and energy intakes, the SCF concluded that the evidence supporting the essentiality of boron has yet to be substantiated (27).

Boron undergoes little, if any, metabolism in the organism. It is excreted through the kidneys with a half-life of approximately 24 hours or less (18, 17, 26) and elimination is similar in rats and in man (19). There is no information available on elimination in pregnant women or in people with impaired kidney function.

The most important toxic effects of boron are on the reproductive system. In male laboratory animals testicular lesions have been observed in rats, mice and dogs given boron in the diet or in drinking water (21).

A survey of reproductive performance was carried out in 542 male workers in a borax mine using a questionnaire approach to test any anti-fertility effect of inhalation of borax. The mean exposure over one year in the highest exposure group was estimated on the basis of a mean male body weight of 70 kg. to be 0.34 mg of boron/kg b.w./day, and no adverse effect on reproduction was found by this indirect method (35).

Developmental effects have been seen in rats, mice and rabbits. The critical effect was a decreased average fetal weight per litter in the rat. Offspring body weight was decreased at 13.3 mg of boron/kg b.w./day and the NOAEL was 9.6 mg of boron/kg b.w./day (12, 24).

No adequate study on boron is available on the developmental effects in man nor is any human study available of effects of boron during pregnancy nor in persons with decreased function of the major excretory organ for boron, the kidney. The Committee, therefore, found no basis for deviating from the usual safety factor to be applied to the NOAEL in the most sensitive animal species.

The Committee noted that the two recent evaluations of boron (8, 10) had arrived at the same no-effect level from the animal studies but had differed in the rationale for the derivation of a safety factor.

Conclusion

A NOAEL of 9.6 mg/kg b.w./day was established on the basis of the rat study (decreased average fetal weight per litter). Application of the usual safety factor of 100 gives a TDI of 0.1 mg boron/kg b.w./day. Consumption of 2 litres of natural mineral water/person/day and an allocation of 10 % of the TDI to this source of exposure would lead to a guideline value of 0.3 mg/l.

5. Manganese (Mn)

Evaluation

Manganese has been shown to be an essential element for animals. Therefore, it is presumed that manganese is also beneficial or essential to humans. On the other hand, higher doses can cause adverse effects, especially on the central nervous system. In humans, neurological effects have been observed in workers following chronic inhalation exposure to manganese dust and fumes. There is, however, only limited evidence that oral exposure might be of concern.

in vitro mutagenicity studies, including tests on bacteria and mammalian cells, have shown that manganese has a genotoxic potential in the absence of metabolic activation. So far, results of *in vivo* assays have been negative (1). Carcinogenicity studies in rats and mice revealed only equivocal evidence of increased tumour incidence (13, 14).

The data on the dose-relationship of changes in the central nervous and reproductive system are insufficient and do not allow no-effect levels to be established. The lowest effective doses were seen in semi-chronic oral studies with $MnCl_2$, in which the motor activity of male rats was changed significantly at about 10 mg Mn/kg b.w./day (4) and testicular changes (20) as well as muscular weakness, rigidity of the lower limbs and marked neuronal degeneration in the region of substantia nigra (11) were noticed in male monkeys at 6.9 mg Mn/kg b.w./day.

Conclusion

From the lowest effective doses observed, the semi-chronic oral studies, a no-effect level of 1 mg/kg b.w./day can be estimated. Application of a safety factor of 100 would result in a tolerable daily intake which would be lower than the essential intake from a nutritional point of view. Therefore, an acceptable maximum level for natural mineral waters was based on the safe and adequate range of 1 - 10 mg Mn/day derived by the SCF in setting nutrient intakes (27). Taking the upper value of this range into account and assuming an allocation factor of 10 % for natural mineral waters and a daily consumption of 2 litres, an upper level of 0.5 mg Mn/l in natural mineral waters appears to be acceptable.

Assumptions concerning consumption of natural mineral waters

The Committee stresses that its evaluations have explicitly assumed that the conventional consumption value of 2 l per person per day as used by the EU and WHO for risk assessment of drinking water, also applied to natural mineral waters. The Committee considered this to be a conservative approach but one which allows a certain flexibility for risk management purposes.

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OPINION ON STARCH ALUMINIUM OCTENYL SUCCINATE (SAOS)

(expressed on 21 March 1997)

Terms of reference

To evaluate the safety in use of starch aluminium octenyl succinate (SAOS) as a powdering agent when processing micro-encapsulated vitamins and carotenoids.

Background

The related compound starch sodium octenyl succinate (SSOS) was accepted for general food use as a food additive by the SCF (1). The Committee's current evaluation of SAOS considers the aspect of exposure to aluminium arising from the requested use of this substance.

Technological function

Since SAOS, unlike most other starches, has hydrophobic properties, it is used in micro-encapsulated vitamins and carotenoids to prevent sticking when drying at low temperature thereby preventing their decomposition.

Exposure

On the basis of the information provided by the petitioner, the Committee made the following evaluation which is based on a 25 % incorporation rate for SAOS in the micro-encapsulated formulations and an aluminium content of 1.25 µg/mg SAOS.

Micro-encapsulated vitamin A and vitamin D

Reference values for nutrition labelling of vitamin A and vitamin D are 500 µg (1 550 IU) and 5 µg (200 IU) respectively (2). The potency of coated vitamin A is 325 000 IU/g while that of vitamin D is 100 000 IU/g.

The maximum daily doses of micro-encapsulated vitamin A and vitamin D proposed by manufacturers are 10 mg and 2 mg respectively, which, at a level of incorporation of 25 % would contain 2.5 and 0.5 mg SAOS respectively or a total of 3 mg SAOS from both. On the basis of an aluminium content of 1.25 µg/mg SAOS, the maximum daily intake of aluminium from this source is thus estimated to be around 3.75 µg.

Micro-encapsulated carotenoids

Carotenoids are normally added at 30 mg/kg as food colours but because the colour potency of micro-encapsulated carotenoids is only 1 % of that of non-micro-encapsulated carotenoids, the micro-encapsulated form of carotenoids may require addition at 3 g/kg of food. At a 25 % incorporation rate, this would be equivalent to 0.75 g SAOS per kg of food or, on the basis of an aluminium content of 1.25 mg/g SAOS, approximately 1 mg of aluminium per kg of food. Although no data are available on the foods to which micro-encapsulated carotenoids may be added and therefore on the intake of such foods, taking account of the typical maximum daily intake of food from all sources (1.5 kg per day for a 60 kg adult), the intake of aluminium resulting from the use of SAOS can be expected to be far below the average dietary intake of aluminium which is of the order of a few milligrams per day.

Toxicology

No toxicological data are available for SAOS but these are available for the related compound SSOS (2). These data are satisfactory and acceptable for the evaluation of SAOS, apart from consideration of the aluminium content. The intake of aluminium from this source would be considerably below the PTWI of 7 mg/kg b.w. for aluminium established by the SCF (3).

Conclusion

Taking into consideration the small contribution of SAOS to the daily intake of aluminium, the use of SAOS as a component of micro-encapsulated vitamins and carotenoids may be regarded as acceptable.

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OPINION ON THE ADDITIONAL INFORMATION FROM THE AUSTRIAN AUTHORITIES CONCERNING THE MARKETING OF CIBA-GEIGY MAIZE

(expressed on 21 March 1997)

Terms of reference

The provisions of Directive 90/220/EEC have led the Commission to seek advice on the additional information from the three Scientific Committees that expressed opinions on Ciba-Geigy maize in December 1996 and in particular:

a) does the information submitted by Austria constitute new relevant scientific evidence which was not taken into account by the Committee at the time its opinion was delivered?

and, if so,

b) would this information thus cause the Committees to consider that this product constitutes a risk to human health or the environment?

The evaluation of the SCF was limited to consideration of risks to human health.

Evaluation of the additional Austrian information

The Austrian point of view is described most precisely in the introduction to the additional information where it is stated that 'especially new scientific results have questioned the present scientific possibility of a conclusive evaluation of the mechanism of gene transfer as well as the development of resistance to the B.t. toxin. Accordingly, possible risks are very hard to assess and should be avoided at the present state of scientific discussion'.

The new data quoted with regard to B.t. toxin relate to potential environmental risks and were not therefore considered by the SCF. With regard to the aspects considered by the Austrian authorities in the section 'Assessment of the ß-lactamase resistance', the Committee has carefully examined the scientific argument presented therein.

During its evaluation of CG-maize, the Committee was aware of, and took into account, the available scientific knowledge concerning the unexpectedly long survival of DNA in the environment and the persistence of fragments of DNA in the human body following the consumption of food. The likelihood of a possible gene transfer to micro-organisms in the intestinal tract under specific conditions was also known and taken into consideration. The potential hazard arising from the original transformation of maize by the use of the pUC plasmid harbouring the gene encoding ß-lactamase was given particular attention and was even the major topic of the *ad hoc* expert meeting organised jointly by the SCF and the Scientific Committee for Animal Nutrition (SCAN) on 6 December 1996.

In its opinion expressed on 13 December 1996, the Committee made reference to the widespread presence in the intestine and in the environment of bacteria that harbour the naturally occurring genes encoding β-lactam resistance and concluded that the possibility that the product would add significantly to the already widespread occurrence of ampicillin resistant bacteria in animals and man is remote.

The Committee notes that none of its conclusions concerning the toxicology, nutritional value, allergenicity and secondary changes are affected by the Austrian arguments.

Conclusions

The information submitted by the Austrian authorities supports the conclusion of the SCF stating that 'the Committee was conscious of the general question of the use of genes coding for antibiotic resistance in marker gene constructs in the development of novel foods and proposes to scrutinise the future needs and application of marker genes'. In the view of the Committee, the Austrian information does not provide new scientific evidence regarding the food use of Ciba-Geigy maize which was not taken into account by the Committee at the time its opinion was delivered. Thus, the Austrian information does not cause the SCF to consider that the Ciba-Geigy maize constitutes a risk to human health.

OPINION ON ACTILIGHT – A FRUCTO-OLIGOSACCHARIDE (FOS)

(expressed on 21 March 1997)

Terms of Reference

To give an opinion on the safety of Actilight (FOS) in accordance with its proposed use as a food ingredient.

Background

The Commission was asked by the petitioner to evaluate Actilight in the context of food additive use in 1988. The Committee concluded at its 83rd Meeting (10 April 1992) as follows: 'The Committee endorsed the conclusions of the Additives Working Group. Although the Committee had no concern about exposure (estimated at around 2 g/day) through the consumption of normal items of the diet in which fructo-oligosaccharides occur naturally, it noted that even from single, typical portion sizes of foods to which Actilight had been added, intakes approached those at which gastrointestinal effects in humans had been reported. Furthermore, in feeding studies with experimental animals numerous effects had been seen, in some cases at all dose levels. The Committee concluded that for these reasons, "Actilight" could not be considered acceptable for addition to food at the levels requested'.

The petitioner responded in February 1995 by providing additional information which it believes addresses the concerns expressed by the Committee. The legal status of Actilight was clarified by the Standing Committee on Food who indicated in June 1995 that it would classify fructo-oligosaccharide as a food ingredient (and not as a novel food ingredient nor as an additive). At its 100th meeting (7-8 March 1996), it was therefore decided to examine the above-mentioned additional information and more recent publications on digestion, excretion and energy value of FOS in healthy humans, and on the effects of chronic consumption of FOS by healthy subjects, in the light of the previous reservations of the Committee related specifically to digestibility.

A complementary file provided by the petitioner presented a more comprehensive analysis of toxicological studies previously submitted. In addition, the petitioner presented new data on sub-chronic toxicity (90-day oral rat study), embryotoxicity in the rat, and a study on colon carcinogenesis after treatment with FOS and a chemical inducer of carcinogenesis. This information showed that FOS has no significant effects, other than gastrointestinal symptoms, at doses from 5 to 40 times higher than the no-effect level for laxative effects in humans.

Digestion, absorption and metabolic fate of FOS

Actilight fructo-oligosaccharides (FOS) result from the action on sucrose of a fructosyl furanosidase present in *Aspergillus niger*. Sucrose plays the dual role of fructose donor and fructose acceptor. The first reaction on two sucrose molecules results in kestose (glucose-fructose-fructose, GF2). The same enzyme acts on kestose to produce nystose (GF3) and on nystose to produce fructosyl-nystose (GF4). Bonds between fructose units are *b* (1-2) (1).

There is no enzyme present in the small intestine that can specifically hydrolyse the (2-1)-*b*-glycosic linkages found in FOS. Consequently, FOS are not hydrolysed nor absorbed in the small intestine but are totally fermented in the colon. Their main nutritional properties and their value in human nutrition are related to their inability to be hydrolysed in the small intestine and to their capacity to reach the colon and to be fermented by the microflora (1).

Studies in rats showed that FOS are not hydrolysed by salivary and pancreatic amylases, and that few or none are hydrolysed by intestinal brush border enzymes (2). Moreover, FOS recovery from rat small intestine was approximately the same as that of an unabsorbable marker (3). Long-term ingestion of FOS did not cause induction or suppression of the rat intestinal brush border enzymes (2). FOS did not influence the transmural potential difference of everted sacs prepared from rat jejunum (4). In addition, when injected intravenously to rats, FOS are rapidly excreted in urine without having undergone any degradation, suggesting that FOS are not used as an energy source in the body (2). On the other hand, *in vitro* incubation of (U-^{14}C) FOS with the caecal content of rats showed that most of the label appeared in short-chain fatty acids (SCFAs). FOS fed to normal rats showed rapid fermentation, whereas germ-free animals delayed excretion of the label for many hours with substantial amounts appearing in faeces (5). Compared with other undigestible sugars (e.g. cellulose, pectins or lactulose), the fermentation of FOS produced higher percentages of propionic and butyric acid, which may be relevant to predicting their metabolic effects *in vivo* (6).

In humans, no change in blood glucose was noted after oral ingestion of FOS (7). Breath-hydrogen studies have also shown that FOS are fermentable, resulting in an amount of hydrogen in breath similar to that excreted after ingestion of an identical load of lactulose (8), suggesting again that FOS are not digested in the human small intestine. This was recently confirmed by *in vitro* and *in vivo* studies (9). Only sucrose was hydrolysed during *in vitro* incubations with homogenates from duodenal mucosa, whereas the constituent oligosaccharides of FOS were not hydrolysed at all. *In vivo*, the fate of FOS in the human gastrointestinal tract was evaluated in six healthy volunteers over an 11-day period. After an equilibration phase, 20.1 g FOS/d was given in three identical postprandial doses: distal ileal output of FOS and its constituent components were determined by intestinal aspiration after a single meal, and the amounts of FOS excreted in stools and urine were also measured. Most of the ingested FOS, 89 ± 8.3 % (x ± SEM), was not absorbed in the small intestine, and none was excreted in stools, indicating that the portion

reaching the colon was completely fermented by colonic flora. A small fraction of the ingested FOS (0.12 ± 0.04 %) was recovered in urine. The mean estimated energy value of FOS was 9.5 kJ, i.e. 2.4 kcal/g (9).

Digestive tolerance

The digestive tolerance to undigestible sugars depends on the amount ingested, on the presence of factors that reduce their osmotic load in the small intestine, and on the degree of adaptation of the colonic microflora to ferment these sugars. The importance of the osmotic effect of undigestible sugars is determined by the concentration of sugar leaving the stomach. This obviously depends on the amount of undigestible sugar ingested but also on factors reputed to slow down gastric emptying, such as the energy content of the meal, the solid content, and the viscosity. The worst conditions for testing the digestive tolerance to undigestible sugars are encountered after fasting, when sugars are tested in a single liquid load, and when the microflora of the subject has not been adapted by chronic sugar ingestion. All these factors must therefore be taken into account when comparing digestive tolerance thresholds of different undigestible sugars (1).

Animal studies

The influence of chronic intake of FOS on growth and intestinal function was investigated in rats by Tokunaga *et al.* (10). Male Wistar rats, initially weighing 40–50 g each, were fed *ad libitum* for 6–8 weeks. The only variable in the experimental diets was the carbohydrate source (corn starch only, corn starch partially replaced by 10 or 20 % FOS, or by 20 % glucomannan), daily food intake was similar in all four groups of six rats each. After feeding rats on these diets for six weeks, the body weight gain of the group receiving the 20 % FOS diet was significantly lower compared with the control group. In animals on the 10 % diet, no significant decrease in body weight gain was observed. A remarkable suppression of body weight gain was also observed in animals consuming the 20 % glucomannan diet (a kind of dietary fibre). The smaller body weight gain was interpreted as a consequence of an incomplete utilisation of FOS as an energy source.

The feeding of 10 % and 20 % FOS diets produced a significant increase in both wet weight and the ratio of caecum to colon weights: a greater effect was observed in the caecum that in the colon of animals fed on the 20 % FOS diet, as in the case of animals fed on 20 % glucomannan diet. A similar enlargment of the caecum and colon has been observed in rats fed with dietary fibres such as pectin, cellulose, guar gum and wheat bran.

The faecal wet weight increased significantly in animals fed on either the 10 % or the 20 % FOS diet ($p < 0.01$), although the range was considerable. The gastrointestinal transit time was about 28 h, 21 h and 14 h in the control, 10 % and 20 % FOS diets respectively, in inverse correlation to the faecal wet weight. The concentration of volatile acids (SCFAs) per gram of wet faeces also greatly increased in animals fed FOS or glucomannan compared with the control group, but the profile of SCFAs was different, indicating that the effects on intestinal microflora also differ.

Finally, it was pointed out by the authors that rats developed diarrhoea after starting FOS feeding. This stopped within 2 to 3 weeks, differing with individual rats. However, FOS intake of rats in the present study was more: 10 g per kg b.w. in the early period of feeding. Another study, quoted by Tokunaga *et al.*, but published only as an abstract, has shown previously that single-dose intake of FOS at less than 0.8 g per kg b.w. does not produce diarrhoea in males, but that it does at above this level, and that females are more resistant to diarrhoea than males (10).

Human studies

The gastrointestinal tolerance of the mixture of oligosaccharides, consisting of glucose linked to a series of 2, 3 or 4 fructose molecules, was evaluated in three studies.

The Japanese study
A first estimation of the maximum non-effective dose of FOS in humans and of the 50 % laxative effective dose (the amount of FOS which causes diarrhoea in 50 % of people, ED_{50}) was performed in 85 healthy Japanese volunteers (51 men, 34 women) by Hata and Nakajina (11). To test the effect on the digestive tract using diarrhoea as index, a FOS mixture (56 % FOS, 12 % sucrose, 29 % glucose, 3 % fructose) given in six different dosages (0.21, 0.27, 0.40, 0.53, 0.67 and 0.8 g/kg b.w.) in 180 ml of water after lunch was compared with a sucrose mixture (68 % sucrose, 29 % glucose, 3 % fructose). At the administration of 0.2 to 0.3 g/kg b.w. standard dose, diarrhoea did not occur in both men and women. The maximum non-effective dose of FOS on diarrhoea (expressed as pure FOS) was 0.3 g/kg (approximately 44 g of the FOS mixture) for men, and 0.4 g/kg (approximately 49 g of the FOS mixture) for women. In comparison, the maximum non-effective dose of sorbitol for men was 0.15 g/kg. The ED_{50} of FOS was 0.78 g/kg for men and 0.84 g/kg for women (ED_{50} for sorbitol: 0.5 g/kg). Incidence of diarrhoea due to doses above the maximum non-effective dose was higher in men than in women by approximately 10 %. Effects of 0.27 g FOS/kg b.w. on the abdominal symptoms and the macroscopic aspects of stools were not influenced by differences in the form of intake (FOS mixture *vs* purified FOS, with or without mixing with food), sex or age. At the same dosage (14 g/d for women and 17 g/d for men), no significant difference was noted between FOS and sucrose (11).

The US study
Clinical tolerance to regular consumption of FOS was also studied by Stone-Dorshow and Levitt (8). In volunteers receiving a constant daily amount of FOS (5 g three times a day with meals), they showed that gaseous symptoms, such as flatulence, bloating and abdominal discomfort were significantly more severe in subjects ingesting the FOS than in control subjects ingesting sucrose. Moreover, symptoms did not improve after a 12-day period on FOS. However, at this daily dose, with the exception of flatulence, symptoms were rated absent or mild and no subject experienced diarrhoea.

The French study

The French study, performed on healthy volunteers (6 females, 8 males, aged from 21 to 37 years), was a double-blind, randomised cross-over study. The tolerance to FOS was compared with that to sucrose. Both sugars as hard candies (2.5 g of sugar each) were consumed occasionally and regularly. In the first period, FOS consumption was occasional, i.e. one dose of FOS and sucrose were tested at random on Tuesdays and Thursdays of each week. To avoid any adaptation, subsequent ingestions of FOS thus were separated by at least 5 days. The starting dose was 10 g. Sugar doses were increased by 10 g until diarrhoea and/or a symptom graded 3 occurred, or when volunteers did not want to ingest more candies.

In the second period, volunteers were asked to consume the same sugar (either FOS or sucrose) every day according to an increasing schedule lasting at most 18 days. In the same manner, they consumed the other sugar after a washout period of 15 days. According to the schedule, subjects should reach the threshold found in the first period (diarrhoea and or grade 3 symptom) on the 15th day. On days 16, 17 and 18 they should ingest this dose plus 10, 20 and 30 g, respectively. As in the first period, subjects were asked to stop sugar ingestion if diarrhoea and/or a grade 3 symptom occurred.

At each dose, the mean scores for each symptom (flatus, borborygmi, bloating, cramps, diarrhoea) experienced with FOS and sucrose were compared and the 50 % effective dose (ED_{50}) was determined graphically. H_2 excretion in breath was measured on the 15th day of FOS and sucrose consumption. Fifteen days after the conclusion of the study, breath H_2 excretion was again assessed before and after FOS ingestion, to evaluate if H_2 excretion was higher in these conditions than H_2 excretion measured for the same daily dose during chronic consumption of FOS.

Symptomatic responses and laxative thresholds were roughly similar during the occasional and regular consumption of FOS. The threshold FOS dose was reached in all subjects: 13 experienced diarrhoea and one severe abdominal pain in the first period. During the second one, 9 had diarrhoea, 1 complained of severe flatus and 4 did not want to take more candies. In both studies, approximately 10 % of the volunteers complained of flatus and borborygmi with 10 g/d. For excess flatus, the first dose at which the severity of the symptom was significantly higher with FOS than with sucrose was 30 g; for borborygmi, bloating and abdominal cramps, this dose was 40 g; the mean threshold and laxative doses were approximately 50–60 g, as ED_{50} for diarrhoea. The mean volume H_2 excreted in breath was not significantly higher during the occasional consumption of FOS than during chronic consumption (183 ± 103 vs 120 ± 104 ml/16 h, $p = 0.16$). H_2 excretion was 45 ± 18 ml/16 h during the sucrose consumption.

The authors concluded that chronic consumption of FOS initiated cautiously with subsequent gradual increase did not improve tolerance, nor reduce excretion of hydrogen. Their results confirm that adaptive response of the microbial flora may be different with various unabsorbable sugars (12, 13).

Conclusions

Metabolic studies on Actilight (a mixture of oligosaccharides consisting of glucose linked to 2, 3 or 4 fructose units) have shown that FOS are poorly absorbed in the human small intestine, but completely fermented by colonic flora.

Diarrhoea has been reported in rats fed a 10 % or 20 % FOS diet (more than 10 g/kg b.w. in the early period of feeding) but the laxative effect of FOS stopped within 2–3 weeks. However, the faecal wet weight and the concentration of SCFAs per gram of wet faeces remained higher than in control rats at the beginning of the sixth week of 10 or 20 % FOS diet. Gastrointestinal transit time was inversely correlated with the faecal wet weight. In animals on the 10 % FOS diet, no significant decrease in body weight gain was observed. The body weight gain of the group receiving the 20 % FOS diet was significantly lower, compared with the control group.

In humans, the maximum non-effective dose of FOS (expressed as pure FOS) on diarrhoea was 0.3 g/kg b.w. (approximately 24 g/day) for men and 0.4 g/kg b.w. (approximately 28 g/day) for women. At the administration of 0.2–0.3 g/kg b.w., diarrhoea did not occur in either men or women.

With 15 g (5 g three times a day with meals) gaseous symptoms, such as flatulence, bloating and abdominal discomfort were significantly more severe in subjects ingesting the FOS than in control subjects ingesting sucrose. At this daily dose, with the exception of flatulence, symptoms were rated absent or mild and no subject experienced diarrhoea.

The amount of FOS which causes diarrhoea in 50 % of healthy adult volunteers (ED_{50}) was estimated in two different studies as 0.8 g/kg b.w., i.e. approximately 50–60 g/day. Gastrointestinal symptoms did not improve after a 12 to 15 day period on FOS. In comparison the maximum non-effective dose of sorbitol and ED_{50} for sorbitol were lower (0.15 g/kg b.w. and 0.5 g/kg, respectively) than those for FOS.

In accordance with its previous opinion on polyols (16th Series), the Committee concluded that, although laxation may be observed at high intakes (more than 30 g/day) a consumption of the order of 20 g a day of FOS is unlikely to cause more undesirable laxative symptoms than isomalt, lactitol, maltitol, mannitol, sorbitol and xylitol. The Committee has no objection to the use of Actilight provided the limitations due to its laxative action are kept in mind.

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OPINION ON DIACETYLTARTARIC ACID ESTERS OF MONO- AND DIGLYCERIDES (DATEM E-472e)

(expressed on 13 June 1997)

Terms of Reference

To re-evaluate the use of DATEM (E-472e) for general food uses and to consider the use of DATEM (E-472e) in foods for special medical purposes (FSMPs) for infants and young children based on protein hydrolysates and amino-acids, and in infant formulae and follow-on formulae based on partial protein hydrolysates and amino acids for infants in good health.

Background

An application was received in 1992 for the use of E-472e as an emulsifier in mother's milk substitute preparations suitable for infants allergic to cow's milk and/or soya protein and also in FSMPs in children suffering from various metabolic disorders. The Committee has been informed that E-472e is the only technologically useful emulsifier in preparations which do not contain any protein. Clinical information additional to the other data already available in 1974 has now been submitted to the Committee.

This emulsifier was also evaluated by JECFA in 1966 and 1974 (1). In 1966 JECFA established for E-472e an ADI with an unconditional zone of acceptance of 0–25 mg/kg b.w. and a conditional zone of acceptance of 25–50 mg/kg b.w. separately from the ADI for other tartaric esters of mono- and diglycerides because diacetyltartaric acid does not occur in nature. In 1974 JECFA confirmed an ADI of 0–50 mg/kg b.w. for E-472e on the basis of then available data (2). In 1978 the SCF reviewed these data and confirmed an ADI of 0–50 mg/kg b.w. in its first report on emulsifiers (3).

The SCF agreed at its 107th Meeting (13 June 1997) to the temporary use of E-472e for two years in FSMPs for infants and young children at levels up to 0.3 g/l (as reconstituted from dry powders), up to 0.4 g/l (liquids) and up to 5 g/kg in gluten-free bakery products for gluten sensitive patients (4).

Specification

The recent submission contains only the JECFA specification of 1978 (5) for E-472e and quotes a saponification value of 490–520, an acid value of 80–100 and an iodine value of approximately 40. There exists a new JECFA specification of 1995 (6) which is, however, not mentioned in the new submission.

Evaluation

When the Committee first assessed the safety to health of E-472e it considered only its use as an emulsifier in food generally. A re-evaluation of the available safety data has become necessary, because requests have now been received for the additional use of E-472e in FSMPs for infants and young children and in infant formulae and follow-on formulae for children in good health.

Biological data

No biological data additional to those evaluated by the Committee in its first report on emulsifiers (3) have currently been submitted to the Committee for evaluation except for some recent clinical studies with a mother's milk substitute containing E-472e. The Committee is also aware that a long-term study has recently been completed. The original biological data included a metabolism study in rats fed E-472e labelled with ^{14}C in the 2 carboxyl groups to determine absorption, distribution and excretion through the lungs and in the urine (8). A 22-month feeding study in dogs was used to determine digestibility and the body distribution of the tartaric acid moiety (8). *In vitro* studies demonstrated hydrolysis of E-472e in aqueous media into mono- and di-glycerides, acetylated tartaric acid, acetic acid and tartaric acid, particularly in the presence of pancreatic enzymes (9).

The toxicological data examined in the earlier review included acute toxicity studies in rats, rabbits and dogs. These showed absence of any significant acute toxic effects (8). A 30-day study in dogs given E-472e i.v. and a feeding study in dogs exteeding over 25½ months, also showed no adverse effects even at the 20 % dose level in the feed (10). In a two-year feeding study in rats, no adverse effects attributable to E-472e even at 20 % incorporation in the diet, the highest dose level tested, were noted (8). Furthermore, a reproduction study in rats extending over 22 months also showed no adverse findings related to the feeding of E-472e up to a dose level of 20 % in the diet (8). E-472e was found to be non-mutagenic in *in vitro* assays using *Salmonella typhimurium* and *Saccharomyces cerevisiae* (14).

Human clinical data

Several recent tolerance trials with a mother's milk substitute preparation containing 2 000 mg/kg E-472e were carried out in new-born infants at high risk for atopy and in infants and children known to suffer from allergy to cow's milk. The parameters examined included weight gain, growth, titre of IgE specific for cow's milk, incidence of positive skin prick tests for allergy to cow's milk and incidence of allergy to cow's milk. In none of these prospective studies were any adverse effects noted in the parameters examined other than those due to the existing underlying allergy or atopic predisposition (11, 12, 13).

Predicted intakes

Given the requested levels of use of E-472e in foods for infants and their low body weights, it is relevant to compare the predicted intakes with the ADI for E-472e of 50 mg/kg b.w. and the possible formation of one of its hydrolysis products, tartaric acid, with the ADI for that substance of 30 mg/kg b.w.

The requested levels of use of E-472e in formulae and follow-on formulae based on hydrolysed proteins for infants in good health and for use in FSMPs vary considerably. One group of products requires levels up to 0.175 g/100 g of dry product, while others require 1.2–3 g/100 g of dry product (15). Intake data have been provided for three typical products in the lower use range of E-472e and these all give estimated intakes of E-472e which are at or below an ADI of 50 mg/kg b.w. (16). Intake data have also been provided for a typical product in the upper use range of E-472e, containing 1.6 g/100 g of dry product. These intake data range from 194–274 mg/kg b.w./day for infants between 0 and 12 months of age (15), implying they would regularly exceed the ADI.

Considering tartaric acid, the Committee has been informed that the E-472e preparations requested for use in formulae and follow-on formulae based on hydrolysed proteins for infants in good health and for use in FSMPs would conform to Food Chemical Codex limits, i.e., would contain up to 20 % tartaric acid (15). Thus any intake of more than 150 mg/kg b.w./day of this type of E-472e preparation could exceed the ADI for tartaric acid. This suggests that use of E-472e at the requested higher use range of 1.2–3 g/100 g of dry product would result in the ADI for tartaric acid being regularly exceeded. (It should also be noted that the EU specification for E-472e allows preparations containing up to 40 % tartaric acid to be used in the European Community).

Conclusions

The Committee has reviewed the total information on E-472e currently available to it and has noted that a new specification for E-472e, aligned with the one of 1995 has not been supplied. The biological data show that hydrolysis in the mammalian gut is sufficiently slow to allow the absorption of some unhydrolysed E-472e. The identity of the urinary metabolite remains undetermined. The compound has no acute toxicity by the oral route. The available short-term, long-term and reproduction studies, though inadequate by present-day standards, provide collateral evidence for the absence of serious adverse effects, when E-472e is ingested in high doses. Genotoxicity has not been adequately studied as neither *in vitro* nor *in vivo* tests for chromosomal aberrations have been provided. However, the clinical studies with a mother's milk substitute containing E-472e demonstrate that E-472e is well tolerated by infants and children suffering from allergy to cow's milk.

In these circumstances the Committee is prepared at present to set a temporary ADI of 25 mg/kg b.w. for general food uses which represents half the ADI established in 1978. but wishes to see the full presentation of the recently completed long-term feeding study in laboratory animals within three months. In the absence of these data the Committee may have to consider further restrictions on the use of E-472e.

The Committee is unable to express a view on whether E-472e is acceptable for use in formulae and in follow-on formulae based on hydrolysed proteins for infants in good health and in the meantime considers that it should not be used in such products. As mentioned previously, the Committee has already agreed to the temporary use of E-472e for two years in FSMPs.

The Committee will reconsider the request for use in infant formulae and in follow-on formulae based on hydrolysed proteins for infants in good health, its recent temporary acceptance for use in FSMPs and the temporary ADI now set for general food use once the following information is supplied. or within two years of publication of this opinion, whichever is the earlier:

i. an adequate specification which also includes a limitation for tartaric acid to 20 %,
ii. full submission of the recently completed long-term study in laboratory animals,
iii. studies on reproduction and teratology conducted to modern standards,
iv. a test for chromosomal aberrations in mammalian cells *in vitro*.

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OPINION ON CANTHAXANTHIN

(expressed on 13 June 1997)

Terms of Reference

To re-evaluate the safety in use of canthaxanthin as a food additive in the light of additional information received.

Background

Canthaxanthin was considered by the Committee for the first time during its comprehensive review of the colouring matters in food in 1983 (1), when the Committee established an ADI of 0-25 mg/kg b.w./day. Subsequently new data, derived from studies on individuals taking canthaxanthin for therapeutic, dermatological or cosmetic purposes (e.g. as sun-tan producing agent), showed that the high doses employed for these purposes could induce the formation of crystalline deposits in the retina, eventually identified as canthaxanthin. These data pointed to a lowest effect level in man of 30 mg/day (0.5 mg/kg b.w./day). Consequently, in 1989, the Committee changed the existing ADI to a temporary ADI of 0-0.05 mg/kg b.w./day, using a safety factor of 10 because of the availability of human data (2). The Committee also requested the provision of certain additional information during the next five years to elucidate more precisely the pharmacokinetics underlying this deposition of canthaxanthin in the retina of both animals and man and to establish whether any functional visual effects were associated with these findings. Further information supplied by the manufacturer to a national advisory committee together with their assessment of this information was later communicated to the Committee. Following its own assessment in 1992 of this new information, the Committee decided to leave unchanged the temporary ADI of 0-0.05 mg/kg b.w./day (3), the validity of which it had limited in 1990 to a period of five years. Additional information has now been submitted to support a reconsideration of the Committee's 1992 assessment. The same information was reviewed by JECFA in 1995, which established an ADI of 0.03 mg/kg b.w (4).

Information submitted since the 1989 review

Animal studies

Long-term feeding studies in rats, extending over 104 weeks, were carried out recently as two separate studies in males and females using dose levels ranging from 5 to 250 mg/kg b.w./day (5). Earlier studies in the mouse extended over 98 weeks (6) and in the dog over 52 weeks (7). None of these studies showed any evidence of crystal deposition in the retina of the test animals

or any other ocular pathology. The rat studies showed increased liver weights both in males and females with associated biochemical changes in the males and histological changes in the liver of males and females interpreted as evidence of a possible hepatotoxic effect. The NOAEL in these rat studies was 5 mg/kg b.w./day. No specific adverse effects were seen in the mouse apart from pigment deposition in hepatic sinusoidal cells. No changes in tumour incidence were noted even at 1 000 mg/kg b.w., the highest dose level tested. The dog study showed only that the adipose tissue contained the highest canthaxanthin concentration, followed by the adrenals and the liver.

A three-year feeding study in cynomolgus monkeys, using doses ranging from 0.2 to 1 000 mg/kg b.w./day, revealed no treatment-related effects on body weight gain, food consumption, haematology, clinical biochemical parameters and urinalysis. The only treatment-related macroscopic changes were orange-red discolourations of the gastrointestinal mucosa, the adipose and the connective tissues. No unusual findings were noted in organ weights and the histopathology of all organs examined showed no evidence of any systemic target organ toxicity. However, examination of the retinae showed birefringent inclusions especially in the peripheral regions. At the lowest dose of 0.2 mg/kg b.w./day no inclusions were seen. Canthaxanthin concentrations in the retina correlated with the presence of birefringent inclusions. These and the microscopic crystalline inclusions in the liver were shown by chemical analysis to be associated with canthaxanthin. No functional visual impairment was associated with these inclusions. The NOAEL for systemic toxicity was therefore 1 000 mg/kg b.w./day and for observable inclusions in the retina it was 0.2 mg/kg b.w./day (8).

Recent pharmacokinetic and metabolism studies in the cynomolgus monkey, using ^{14}C-labelled canthaxanthin showed plasma concentrations to be higher than those in the blood with peak values occurring in males within 4 hours and females within 6 hours. Faecal excretion was the major route of elimination amounting to 85–90 %, urinary excetion accounted for 1.6–3.6 %, while tissue retention ranged from 1.6 to 4.6 % and occurred preferentially in the adrenals. Monkeys therefore appeared to absorb from 3 to 8 % of orally administered canthaxanthin. (9).

In a study comparing the pharmacokinetics in rats and monkeys under steady state conditions the urinary excretion was faster in the rat, some 4.6 % being found in the rat urine after 96 hours compared with 2.1 % in the monkey urine over the same period. Some 91.4 % of the administered dose was recovered from the rat faeces after 96 hours compared with 87.2 % after the same period in monkey faeces. Overall tissue residues after 96 hours amounted in the rat to about 0.8 %, accumulating mostly in the spleen and liver, whilst the equivalent figure for the monkey after 96 hours was 5.1 % with accumulation being highest in the adrenals. Excretion and metabolism were thus faster in the rat than in the monkey. In the rat the urinary metabolites contained some very polar compounds which were present only in trace amounts in monkeys, while monkey urine contained some less polar compounds absent from rat urine. In the monkey retina 4^1-OH-echinenone and isozeaxanthin were identified as metabolites. The urinary metabolites have not been identified so far in both species. The mean plasma level of canthaxanthin in the monkey at the NOAEL of 0.2 mg/kg b.w. was 156 µg/l (10).

In a search for appropriate animal model species to study crystal deposition of canthaxanthin in the retina, various other species were exposed to this colourant. Thus, the ferret was found to be unsuitable as no canthaxanthin residues could be detected in any of its tissues (11). Five different mouse strains were examined but little deposition in the retina could be detected and the distribution differed from that found in humans. However, some functional ERG effects were noted which were reversible (12). Birefringent crystals accumulated in a dose-related manner in the retina of broiler chicken exposed to canthaxanthin, correlating also with the plasma concentrations (13). Guinea-pigs accumulated the colourant in their retina after some 10 months treatment (14).

Canthaxanthin administered to rats at a dose of 300 mg/kg diet (15 mg/kg b.w.) for 15 days increased the concentration of hepatic P_{450} and thereby the activities of the hepatic isoenzymes P_{450}1A1 and P_{450}1A2. There was co-induction of the microsomal Phase II enzyme activities of p-nitrophenol- and 4-hydroxybiphenyl-UDP glucuronyl transferases as well as quinone reductase, an effect reminiscent of 3-methylcholanthrene inducing activity (16).

***In vitro* studies**

Neuronal retina reaggregate cultures, derived from chick embryos, were found to be a suitable system to study cellular crystal deposition. Red-brown birefringent entities formed in these cells in proportion to the amount of canthaxanthin in the culture medium. No accumulation occurred at 120 µg/l medium but crystals appeared at 1200 µg/l medium (15).

Human ophthalmological data

A retrospective biostatistical analysis of 411 cases of canthaxanthin treatment either for cosmetic or therapeutic reasons, for which sufficient relevant information was available, indicated that 95 individuals had a crystal retinopathy. This finding occurred only when high doses had been ingested over long periods and a dose-response relationship was discernible. A dose of 30 mg (0.5 mg/kg b.w.)/day or a cumulative intake over a prolonged period of 3 g/person appeared to be the lowest effective dose (LEL) causing crystal deposition in the retina. No such deposits were reported with exposures of less than 30 mg/day. Doses above 105 mg (1.75 mg/kg b.w.)/day led to crystal deposition in the retina in 50 % of cases (17).

Some reversibility of the deposits of canthaxanthin in the retina was noticed in patients who, over 12 years, had accumulated these deposits and had thereafter discontinued treatment for at least five years (19).

No hepatotoxicity was detected in 11 patients treated for up to 12 years for erythropoeitic protoporphyria with cumulative doses up to 150 g (20).

An earlier review of the ocular toxicity of canthaxanthin in persons with a high intake of canthaxanthin and deposits in their retina had demonstrated that this phenomenon was not associated clinically with any significant adverse functional visual defects. However, a

reproducible and reversible small decline in the scotopic ERG b wave amplitude at higher light intensities without any change in peak latency was detected. This small functional effect was:

- not detectable by psychophysical tests;
- always associated with normal visual acuity and colour vision but occasionally with minimally reduced threshold static perimetry and mild reversible delayed dark adaptation;
- not due to retinopathy with loss of retinal sensitivity because the ERG kinetics and the ERG a waves (parameter for photoreceptor activity) were unaffected and weak stimuli did not induce any ERG b waves (parameter for general retinal damage);
- due to a depolarisation of the glial Müller cells, because crystals were found near the end feet of the Müller cells near the inner surface of the retina. These crystals were eventually voided into vacuoles and either phagocytosed by cells of the retinal pigment epithelium or disappeared very slowly from the retinal surface due to the absence of an active removal process.

These changes were not considered to be of pathological significance or indicative of significant functional damage to the retina. In this review the LEC for effects on the ERG b wave was estimated to lie between 0.25 and 1 mg/kg b.w (18).

Human intake estimates

According to Directive 94/36/EEC the use of canthaxanthin as a food additive is restricted to the colouring of saucisse de Strasbourg. The Committee is aware that it is also used as an animal feed additive for poultry and fish and that residues may therefore be present in egg yolks and farmed trout and salmon. Canthaxanthin also occurs naturally in certain wild mushrooms. Reliable intake estimates are not available at present but data suggest that 0.2 mg/egg and 0.1 mg/100 g fish are representative residue levels. In view of the low ADI it is particularly important that intakes from all sources should be taken into account in any future risk assessment.

Conclusion

The lowest effect level for ERG b wave changes in man was 0.25 mg/kg b.w./day but in view of the fact that these changes were not of pathological significance or indicative of significant functional damage to the retina, a safety factor of 10 is considered appropriate. This is supported by the finding of a one order of magnitude difference between the plasma level (156 µg/l) at the NEL in monkeys and the *in vitro* concentration (1 200 µg/l medium) first showing the presence of cellular microcrystal formation in neuronal retina reaggregate cultures. An ADI of 0.025 mg/kg b.w., rounded up to 0.03 mg/kg b.w., can therefore be established.

The Committee considers that up-to-date information should be obtained on human intake from the use of canthaxanthin in animal feeds to give assurance that total exposure by this route would not exceed the ADI.

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OPINION ON A REQUEST FOR THE USE OF ALGAL BETA-CAROTENE AS A FOOD COLOUR

(expressed on 13 June 1997)

Terms of reference

To advise on the safety in use as a food additive of a beta-carotene preparation produced from *Dunaliella salina*.

Background

A first petition has been introduced for the addition to the list of food colours authorised in the EU of beta-carotene produced in a bioreactor by a unicellular alga, *Dunaliella salina*. The Committee had given a first opinion on 10 December 1987 (21st series of reports) in which it asked for more information about the composition of the product and on the process used for its preparation. In 1991, the additional information provided was not considered satisfactory to judge on the safety of the product and this first petition was withdrawn. In 1992, a new request was made by another petitioner for the same 'natural beta-carotene' also produced by *Dunaliella salina*, but grown in large saline lakes located in Whyalla, South Australia. By 1997 the information available was considered sufficient by the Committee to give the following opinion.

Safety evaluation

Composition

The beta-carotene preparation is a dispersion in a soya bean oil (containing 0.3 % natural tocopherol) at a concentration of 20–30 %. It is composed of all-trans- and of cis- (mainly 9- but also 13- and 15-) isomers. The respective concentrations of trans- and cis-isomers have been found to be in the range of 50/50 to 71/29. The other major components are alpha-carotene and oxygen containing carotenoids. It also contains minor constituents such as sterols, hydrocarbons, free fatty acids, oxidised carotenoids and chlorophyll. 0.5 % of the product before the addition of soya oil remains unidentified. The heavy metals content is very low (less than 10 ppm expressed as Pb). Microbiological contamination could not be found in the samples analysed (total count, *E. coli*, yeast and mould not detected in 0.1 g, *Salmonella* not detected in 10 g).

Stability

The Committee was satisfied that the beta-carotene preparation was stable.

Production process

Dunaliella salina grows in 30 % salt concentration, coupled with high light intensity and high temperature. The Committee was informed that beta-carotene content of the culture, pH and brine density are measured on a daily basis. Weekly controls include chemical analysis of the algal cells and of the brine as well as a microscopic examination of the cells. The data provided demonstrate a batch-to-batch consistency. Harvesting of the algal cells and concentration of the beta-carotene from the cellular components involve only physical processes. Isolation of the beta-carotene is obtained after several steps: breakdown of protein and carbohydrate into soluble components by food-grade enzymes; extraction of the beta-carotene using an essential oil; saponification; partial crystallisation without isolation of carotene crystals; and finally oil dispersion of the beta-carotene material and adjustment of its concentration.

Toxicological data

Dunaliella salina belongs to the *Chlorophyceae*. It is considered that this unicellular alga, like 25 other species of *Chlorophyceae* which are classified as food sources, does not produce toxins.

Acute toxicity:
LD_{50} of a preparation containing 30 % of beta-carotene is higher than 20 g/kg b.w. in the mouse.

Short-term toxicity:
Several studies, lasting between two and eight weeks, have been conducted on rats and chicken with a powder of the alga *Dunaliella bardawil* (which the Committee has been informed is identical to the species *Dunaliella salina*) without signs of toxicity up to the equivalent of 0.1 % of beta-carotene in the diet.

Multigeneration reproduction study:
No toxicity has been assigned to a powder of *Dunaliella bardawil* ingested by the rat up to 10 % in the diet (equivalent to 0.2 % of beta-carotene).

Genotoxicity:
The algal beta-carotene was unable to induce gene mutation or chromosomal aberrations in two Ames tests and one chromosome aberration assay in human lymphocytes.

Data obtained on the algal powder provide reassurance of the absence of toxicity of the components other than beta-carotene. This is important for the safety evaluation of the beta-carotene preparation in the absence of a total crystallisation of beta-carotene.

Conclusion

On the basis of the information provided, the Committee consider that the use of a dispersion of beta-carotene produced by the alga *Dunaliella salina* growing in large, shallow saline lakes in Whyalla, South Australia, is acceptable as a food additive. This opinion is expressed on the basis of a maximum use level of this preparation around 50 ppm (equivalent to around 10 ppm of beta-carotene).

This conclusion is valid only for the beta-carotene produced in the conditions and at the sites described in the dossier provided by the petitioner which provided reassurance on the composition and relative purity of the material. A specification should be developed which complies with all the above information and recommendations.

OPINION ON CERTAIN ADDITIVES FOR USE IN FOODS FOR INFANTS AND YOUNG CHILDREN IN GOOD HEALTH AND IN FOODS FOR SPECIAL MEDICAL PURPOSES FOR INFANTS AND YOUNG CHILDREN

(expressed on 13 June 1997)

Terms of reference

To consider the safety-in-use of certain additives in infant formulae, follow-on formulae and weaning foods for infants and young children in good health and in foods for special medical purposes (FSMP) for infants and young children.

Background

The use of additives in foods for infants and young children in good health and in FSMP for the same age group is controlled under European Parliament and Council Directive 95/2/EC on food additives other than colours and sweeteners (1). This restricts the additives permitted in infant formulae, follow-on formulae and weaning foods for infants and young children in good health to those listed in Parts 1–3 of Annex VI to the directive. The use of additives in FSMP for infants and young children is controlled under Part 4 of Annex VI of the same directive, which permits all the additives listed in Parts 1–3 to be used in FSMP, with no additional additives currently permitted exclusively for FSMP.

The Association of the Food Industries for Particular Nutritional Uses of the European Union (IDACE) has requested an amendment of Directive 95/2/EC to include additional additives for use in foods for infants and young children in good health and for use in FSMP for the same age group (2, 3, 4, 5). In some cases particular additives have been requested for both categories (good health and FSMP) and in other cases for just one category or the other. Some of the requests to use the same additive in both categories relate to use in products containing hydrolysed proteins which have special technical requirements (e.g. products for cow's milk allergy). Such products are marketed as FSMP and/or as foods for infants and young children in good health.

The Scientific Committee for Food (SCF) has recently given an opinion on the use of certain colours for FSMP for young children aged 12–36 months of age (6). The present opinion covers certain additives other than colours, most of which are emulsifiers, stabilisers or thickening agents. The additives requested and the recommendations of the Committee are listed in the table 'Summary of Recommendations' at the end of this opinion and are considered individually below.

General considerations of need

In considering the request to use additional additives, the Committee was mindful of the principle set out in its earlier reports that it is prudent to keep the number of additives used in foods for infants and young children to the minimum necessary (7, 8). The Committee has stressed in the past that there should be strong evidence of need as well as safety before additives can be regarded as acceptable for use in infant formulae and foods for infants and young children. The Committee continues to endorse these principles, but is aware, for reasons explained below, that the nature of FSMP is such that they may require a wider range of additives than those already permitted for foods for infants and young children in good health and/or they may require a higher level of addition of an additive already permitted for foods for infants and young children in good health. The Committee is also aware that, for historical reasons, different manufacturers of FSMP have developed products over long periods of time utilising different additives which may nevertheless have the same technological function. However, the Committee wishes to record that it is not always easy to reach a view on the case of need.

FSMP for infants and young children encompass a wide variety of different products in powdered, liquid or semi-solid forms, each with a specific formulation and hence each with its own technological requirements. They are generally either of the 'elemental' type (formulae containing free amino acids, glucose syrup or maltodextrin and a low fat content) or 'semi-elemental' type (containing hydrolysed proteins, maltodextrin and fat), together with vitamins, minerals and trace elements. The fats and starches used may also be unusual compared with those used in foods for infants and young children in good health. Thus the technological requirements for additives for FSMP may differ considerably from those for foods for infants and young children in good health.

Normal infant formulae based on cow's milk benefit from the inherent properties of the ingredients they contain, in particular whole proteins which act as efficient emulsifiers. Products containing hydrolysed proteins, peptides or free amino acids lack the normal emulsifying properties of whole protein and so the use of additives such as emulsifiers with a high hydrophylic-lipophylic balance (HLB) value is necessary to enable lipids and carbohydrates of very high molecular weight to interact. This limits the separation of the fatty phase in ready-for-consumption liquid products, notably when products are formulated with modified fats.

The only thickening agent or stabiliser which is permitted in infant formulae for infants in good health is starch. Native starches tend to lose their thickening properties when processed, cooked and stored. Products formulated with protein hydrolysates, peptides or free amino acids require added thickening agents and stabilisers to influence viscosity both during sterilisation and at room temperature, to avoid separation of the various elements such as fibres in liquid feeds, and to reduce the coalescence of fat globules, especially in products in liquid, ready-to-consume form

which may be stored for several months. The particular oils and fats used in certain products may also require the addition of thickening agents which are adapted to their special properties.

Against this background, the specific considerations of need for each of the additives requested for foods for infants and young children in good health and for FSMP for the same age group have been carefully considered by the Committee. For each additive requested for FSMP, the Committee was also informed about the range of medical conditions requiring such foods which might contain that additive and the estimated levels of intake of each additive in the various age groups (2, 3, 4, 5).

It is to be noted that, unless otherwise stated, reference to FSMP in the following evaluations is to be understood as covering FSMP intended for infants and young children from birth onwards.

Consideration of individual additives

Distarch phosphate (E-1412)

Distarch phosphate has been requested for use up to 10 g/l (reconstituted dry powders) and 22 g/l (liquids) in infant formulae and follow-on formulae for infants and young children in good health and in FSMP. It is a chemically modified starch containing less than 0.02 % phosphorus and is used as a stabiliser, thickening agent and binder. Infant formulae containing modified milk proteins or soy proteins and insoluble salts (such as calcium) require a stabilising agent to prevent protein precipitation during sterilisation or storage. The Committee has been informed (2) that starches cross-linked with phosphorus, such as distarch phosphate, have higher stability during shear stress and heat sterilisation. The Committee notes that distarch phosphate is included in the list of additives permitted by the Codex Alimentarius in soya-based infant formulae (5 g/l) and in hydrolysed protein and/or amino acid based infant formulae (25 g/l).

Distarch phosphate was assigned an acceptable daily intake (ADI) 'not limited' by the FAO/WHO Joint Expert Committee on Food Additives (JECFA) in 1973 (10). Modified starches for general food use and for use in baby food were also considered by the SCF in 1976 and in 1981 (11). Both JECFA and the SCF have considered the kidney lesions which occur in rats fed high levels of modified starches. They concluded that the rat is a particularly sensitive species for pelvic nephrocalcinosis and the finding has little relevance for safety evaluation of modified starches in man, and JECFA confirmed an ADI 'not specified' (11, 12). In its report on weaning foods (13), the SCF concluded that use of distarch phosphate singly or in combination with other starches was acceptable up to a limit of 5 g/100 g. In 1992, the SCF reviewed the suitability of distarch phosphate for use in infant formulae and concluded that distarch phosphate should not be permitted in infant formulae (8).

Consideration of the digestibility of starches is also relevant. Adults digest starch mainly by the action of pancreatic amylase but infants have a very low activity of this enzyme (14). Instead, mucosal glucoamylase and salivary amylase are sufficient in some infants to digest long-chain glucose-polymers (15). It has also been shown that infants are capable of digesting cooked native starches from the age of one month (16). However, large amounts (40 g/day) of starches in the diet given to one-month old infants resulted in malabsorption and fermentative diarrhoea (16). It has also been suggested that undigested starch in the gastrointestinal tract may interfere with other food ingredients resulting in failure to thrive in some infants (17). The SCF has concluded that pre-cooked or gelatinised starch (but not native or chemically modified starches) can be added to infant formula up to 2 g/100 ml, but not exceeding 30 % of the total carbohydrate content (7). An addition of 2 g starch/100 ml corresponds to an estimated intake of 10 g/day. Consequently, there is no wide margin between the permitted level of native starches and the level of 40 g starch per day, where effects have been observed.

In vitro studies using human and rabbit pancreatic amylases and *in vivo* studies in adult rabbits show that the digestibility of waxy corn distarch phosphate and native starches is comparable (18). This study may be of little relevance for the digestibility of modified starches in infants due to low levels of pancreatic amylase, but as pointed out in the opinion by the Committee in 1992 (8), the spectrum of activity of alpha-amylase in saliva and pancreatic secretion is the same since the amino-acid sequence is identical (19). Nevertheless, it is not known how the efficacy of the alpha-amylase in saliva compares with the pancreatic form due to possible effects on the enzyme of, for example, gastric juice.

Conclusion:
In its 1992 opinion (8) the SCF recommended distarch phosphate should not be permitted in infant formulae because the Committee would prefer to see direct evidence indicating that infants can tolerate the 2.5 % level of modified starches then requested. The current request is for use up to 2.2 %. A concern was also raised that infants could develop fermentative diarrhoea or modification of the gut flora. No new information on these aspects has been found. Furthermore, the Committee is not persuaded there is a need for use of distarch phosphate in infant formulae generally. A case of need could be valid for formulae containing hydrolysed proteins (whether for children in good health or FSMP), but at present the Committee has insufficient information to reach a conclusion on this point. In the meantime the Committee does not consider that the use of distarch phosphate is acceptable in infant formulae, follow-on formulae for infants and young children in good health or in FSMP.

Sodium citrate (E-331) and potassium citrate (E-332)

Sodium citrate and potassium citrate have been requested for use from birth, either singly or in combination at levels up to 2 g/l, in infant formulae and follow-on formulae for infants and young children in good health and in FSMP (2). The heating procedure for infant formulae or follow-on formulae made from cow's milk results in denaturation and aggregation of proteins. In extreme cases the result is a phase separation of fat and proteins.

The Committee has been informed (2) that the addition of sodium citrate and potassium citrate improve heat stability. During heat treatment casein in milk coagulates due to surplus ionised calcium. The addition of sodium or potassium citrate complexes free calcium ions, resulting in decreased coagulation. The same technological function can also be performed by sodium and potassium phosphates (see below) and all four additives have been requested so that alternatives are available (e.g. in cases where no additional phosphate load can be tolerated). Sodium and potassium citrates are already permitted under Directive 95/2/EC in weaning foods at *quantum satis* levels for pH adjustment only (1), and under Directive 91/321/EEC as sources of nutrients in infant formulae and follow-on formulae for infants and young children in good health (9).

Conclusion:
The Committee considers the use of sodium and potassium citrates are acceptable up to 2 g/l, either singly or in combination, in infant formulae and follow-on formulae for infants and young children in good health and in FSMP.

Sodium phosphate (E-339) and potassium phosphate (E-340)

Sodium phosphate and potassium phosphate have been requested for use from birth, either singly or in combination, at levels up to 1 g/l (expressed as P_2O_5), in infant formulae and follow-on formulae for infants and young children in good health and in FSMP (2). Their technological function is the same as that for sodium and potassium citrates (see above). The use of sodium and potassium phosphates in weaning foods and cereals for infants and young children is permitted under Directive 95/2/EC, either singly or in combination up to a maximum of 1 g/kg (expressed as P_2O_5) (1). These additives are also permitted as nutrients in infant formulae and follow-on formulae under Directive 91/321/EEC (7, 9). In 1992, the Scientific Committee for Food also concluded that the use of phosphoric acid (E-338) in infant formulae was acceptable (8).

Sodium phosphate and potassium phosphate were evaluated by JECFA in 1982 (12). Of greatest concern was the toxicity arising from excess of phosphorus (P) in the diet and deficiencies in calcium (Ca). A high dietary phosphate intake in animals results in hypocalcaemia which stimulates secretion of parathyroid hormone. This hormone inhibits tubular reabsorption of phosphates by the kidney and increases demineralisation of bone tissue. P is mainly excreted in faeces as calcium phosphate; so that intake of excessive amounts of sodium phosphate and phosphoric acid may cause a loss of Ca. The main toxicological finding in animal feeding studies with phosphates is nephrocalcinosis, the rat being highly susceptible. An estimate of the lowest level of dietary intake of phosphates (expressed as P) that might cause nephrocalcinosis in man is about 7 000 mg P/day. Since P is an essential nutrient, JECFA allocated a maximum tolerable daily intake (MTDI) rather than an ADI. The MTDI of 70 mg/kg b.w. applies to the sum of phosphates naturally present in food and derived from additives in the diet, for diets that are nutritionally adequate with respect to Ca (12).

Old data indicate that high-phosphorus human milk substitutes may contribute to hypocalcaemic tetany in infants. However, when artificial formulae simulated breast milk, the occurrence of

tetany decreased (20). Cow's milk contains more Ca and more P than human milk. The Ca:P ratio in cow's milk is 1.35:1 and in human milk 2.25:1. On the other hand, infants absorb 85–90 % of the P in human milk and 65–70 % of that in cow's milk (21).

The RDA (recommended dietary allowance) for P for formula-fed infants from birth to six months of age is 300 mg/day, and 500 mg/day for infants aged 6–12 months (21). Allowances for P are based on a Ca:P ratio of 1.3:1 (the same as in cows' milk) during the first six months of life and 1.2:1 for the following six months. Thus, for newborn infants, the RDA is around the same level as the MTDI for adults. The Committee therefore considers that the MTDI as an upper limit is not applicable to infants.

At the requested level of 1 g/l for E-339 and E-340 (expressed as P_2O_5), the P content of formula would be 440 mg P/l (P is 40 % of phosphate). The mean consumption of infant formula is 700–850 ml/day in infants aged 1–3 months, with a range of 500–1100 ml/day (22), and the 90th percentile consumption is about 1 000 ml/day. Thus, the estimated 90th percentile intake of P from use of E-339 and E-340 would be 440 mg P/day. This represents 150 % of the RDA.

Conclusion:
The estimated 90th percentile intake of P, although above the RDA for infants, is unlikely to be harmful. While knowledge about toxicity from P intake in infants is limited, there are no indications that there would be adverse health effects at the intakes resulting from the use of E-339 and E-340 at the requested level, provided the Ca:P ratio is kept within the recommended limits set out in Annex 1 to Directive 91/321/EEC (9). The Committee considers the use of sodium phosphate and potassium phosphate, either singly or in combination, up to 1 g/l (expressed as P_2O_5) is acceptable in infant formula, follow-on formula and FSMP.

Locust bean gum (E-410)

Locust bean gum has been requested for use in infant formulae and follow-on formulae, both for infants and young children in good health and in FSMP, at levels of 4–10 g/l, to reduce gastro-oesophageal reflux (GOR). Locust bean gum, also called carob bean gum, is refined from the endosperm of the carob tree, *Ceratonia siliqua*. It contains tannins and the carbohydrate component is a galactomannan polymer consisting of linked D-mannose units with side chains of D-galactose. It is used as a stabiliser and thickening agent. It is currently allowed in follow-on formulae at a maximum level of 1 g/l, and in weaning foods at a maximum level of 10 g/kg under Directive 95/2/EC (1). The SCF has previously considered a request to use locust bean gum at a level of 1 g/l in infant formulae but at that time there was no confirmation of need (8).

The Committee is aware that some medical specialists (Espgan) recommend that thickening of foods is useful in the treatment of GOR, and that in cases of uncomplicated GOR, treatment with thickening agents may be started without complementary investigations (23). Clinical

observations have shown that the clinical efficacy is best when locust bean gum is added to infant formula in the concentration range 4–10 g/l (2). However, there are few controlled studies of the efficacy of use of thickened infant formula in reducing GOR. It is believed that the increased viscosity of thickened feed will reduce the episodes of reflux, but it has been shown that the effects are unpredictable (24, 25, 26). Thickeners added to infant formula may reduce the number of reflux episodes, but may also prolong the duration of remaining episodes (27). Increased coughing in infants after thickened feedings compared with after unthickened feedings has also been reported (28).

Locust bean gum was evaluated by JECFA in 1981 (29). An ADI not specified was allocated due to lack of toxicity. In an unpublished three-generation reproduction study referred to by JECFA, significant decreases in premating body weight gain in the F0 females fed 2 % locust bean gum, and final body weight in the females fed 5 % locust bean gum were observed. No significant effect on body weight gain was observed in newly weaned rats fed a diet containing 2 % locust bean gum for 36 days (30). On the other hand growth was depressed in chickens fed a diet containing 2 % locust bean gum for 24 days (30). Thus there are indications of growth depression in animals fed locust bean gum, although these are equivocal.

Bean gum preparations are fermented in the colon, providing a small energetic gain. They can cause abdominal pain and diarrhoea (23). Absorption of minerals and trace elements may be reduced by dietary fibre and tannins. Although a study on adults ingesting locust bean gum has shown no evidence of impaired absorption of minerals and trace elements (31), it is not always appropriate to use results from adults when evaluating health effects in infants in cases where growth may be affected. In rapidly growing healthy infants, even minor effects on gastrointestinal absorption of trace elements and minerals may have growth retarding effects. Studies on growth in healthy infants chronically exposed to locust bean gum are lacking.

Conclusion:
The Committee is not persuaded that it is necessary to give thickened infant formula to infants in good health. It therefore recommends that the use of locust bean gum is not acceptable, at the doses requested, for use in infant formulae and follow-on formulae intended for infants in good health. The Committee does, however, accept that there is a case of need for its use in FSMP to treat GOR and recommends that its use in these products up to 10 g/l is acceptable.

Mono- and diglycerides of fatty acids (E-471)

Use of this emulsifier has been requested for FSMP at levels up to 5 g/l in powdered and liquid formulations intended for infants and young children from birth onwards (2). It is currently permitted under Directive 95/2/EC in infant formulae and follow-on formulae for infants in good health up to 4 g/l and in weaning foods for infants and young children in good health up to 5 g/kg (1). The Committee has been informed (2) that a slightly higher level of E-471 is required, for use

in combination with other emulsifiers, for amino acid and peptide-based nutritionally complete FSMP which contain fat requiring an emulsifier of intermediate HLB value.

This emulsifier has not previously been individually considered by the SCF. The basic EC directive on emulsifiers, stabilisers, thickeners and gelling agents for use in foodstuffs (32) was agreed in 1974, before the SCF was constituted. That directive permitted over 65 additives, some temporarily. The first SCF report on emulsifiers, stabilisers, thickeners and gelling agents (33) published in 1978, explains that the Committee was asked to review only certain of the additives in the directive. The review included the E-472 group of emulsifiers but did not include E-471. However, since the E-472 group are derived from E-471 (they are the acetic, lactic, citric, tartaric and diacetyl tartaric acid esters of mono- and diglycerides of food fatty acids), it can be assumed that the SCF was content to go along with the acceptable daily intake (ADI) 'not limited' for E-471, confirmed by the FAO/WHO Joint Expert Committee on Food Additives (JECFA) in 1974 (34).

Conclusion:
The Committee considers that the use of E-471 is acceptable at levels up to 5 g/l in all FSMP.

Citric acid esters of mono- and diglycerides of fatty acids (E-472c)

This emulsifier has been requested for use from birth onwards in dry formulae up to 7.5 g/l and in UHT-liquid formulae up to 9 g/l which contain partially or extensively hydrolysed proteins both for infants and young children in good health and for FSMP (2). Under Directive 95/2/EC, it is not currently permitted for use in infant formulae or follow-on formulae for children in good health but is permitted up to 5 g/kg in weaning foods for children in good health (1). The Committee has been informed (2) that E-472c is needed, in addition to the emulsifiers already permitted, for products based on hydrolysed proteins or amino acids with high levels of fat, requiring emulsifiers with high HLB value. The E-472 series of emulsifiers has high HLB values.

In its 1978 review (33), the SCF concluded that the use of E-472c was acceptable for foods generally and, in effect, endorsed the ADI set by JECFA in 1974, allocating an ADI 'not specified' for E-472c (34).

Conclusion:
The Committee considers that the use of E-472c is acceptable in products which contain partially hydrolysed proteins for infants and children in good health and for FSMP containing extensively hydrolysed proteins or amino acids at levels up to 7.5 g/l in ready-to-feed products made from dry powder and up to 9 g/l in ready-to-use UHT-liquid formulae.

Diacetyl tartaric acid esters of mono- and diglycerides of fatty acids (E-472e)

This emulsifier, also known as DATEM, has been requested for use from birth onwards up to 3 g/l in reconstituted powders and up to 4 g/l in liquid formulations which are devoid of whole protein, both for children in good health and for FSMP, and up to 5 g/kg in gluten-free bakery

products for coeliac patients (2). It is not currently permitted in infant formulae, follow-on formulae or weaning foods for infants and children in good health (1). The Committee has been informed (2) that it is a suitable emulsifer, in combination with other already permitted emulsifers, for hydrolysed protein, peptide and amino acid-based products, having very large hydrophilic polar groups which are particularly suitable for multi-component systems.

In its 1978 review, the SCF concluded that the use of DATEM was acceptable for foods generally and, in effect, endorsed the ADI set by JECFA in 1974, allocating an ADI of 0-50 mg/kg b.w. (33). The SCF has now updated its view on DATEM and, for reasons explained in a separate opinion (57), has withdrawn the ADI of 0--50 mg/kg b.w. and set a temporary ADI of 0 25 mg/kg b.w.

Conclusion:
The Committee temporarily accepts the use of DATEM in FSMP, but at a lower level than that requested for foods which are given as liquid formulations (whether reconstituted from dry powder or from liquids). The use of DATEM is temporarily acceptable for a period of two years at levels up to 0.4 g/l, as consumed, in FSMP which are devoid of protein, and up to 5 g/kg in gluten-free bakery products for coeliac patients. The Committee is aware that a limit of 0.4 g/l may still mean that some infants and young children could exceed the temporary ADI of 0 25 mg/kg b.w. However, the Committee has also taken into account the essential nature of these FSMP for infants and young children with certain diseases and the considerable technical problems in manufacturing these FSMP. Furthermore, the Committee will review the situation in two years' time. In the meantime the use of DATEM in foods for infants and young children in good health is not acceptable.

Sucrose esters of fatty acids (E-473)

Sucrose esters of fatty acids have been requested for use as an emulsifier up to 120 mg/l in powdered and liquid FSMP containing partially or fully hydrolysed proteins or amino acids for infants and young children from birth onwards, and in liquid infant formulae and follow-on formulae containing partially hydrolysed or fully hydrolysed proteins for infants and young children in good health (2). They have been permitted as an emulsifier in the EU in foods generally (except for bread unless authorised for that use nationally) since 1974 (32) and in dietetic foods (35). They are not currently on the list of permitted additives for foods for infants and young children in good health.

Sucrose esters of fatty acids were evaluated by the SCF in 1992, when a group ADI of 0 - 20 mg/kg b.w. (expressed as sucrose monostearate) was allocated for sucroglycerides and sucrose esters derived from palm oil, lard and tallow fatty acids, subject to a specification which would limit the presence of tetra- and higher esters to no more than 7 % (36). This opinion was based on information that the lower fatty acid esters of sucrose are hydrolysed in the gut to the constituent fatty acids and sucrose, which are well-known components of food. It has been considered several times by JECFA and, in its most recent evaluation in 1995, also given an ADI of 0 -20 mg/kg b.w.

expressed as sucrose ester content (37). The only adverse effects noted in humans from sucrose esters of fatty acids are gastrointestinal disturbances (soft stools, diarrhoea, flatulence and bloating) seen at intakes of 70 mg/kg b.w./day, with a no-effect level of 35 mg/kg b.w./day.

Intake estimates from the use of E-473 in ready-to-use liquid formulae used as FSMP for infants and young children from birth up to three years of age range from 15 - 20 mg/kg b.w./day, which is just within the ADI. It is unlikely that the ADI for E-473 would be exceeded on a regular basis since E-473 is not permitted in other foods specially prepared for infants and young children.

Conclusion:
The Committee considers that the use of sucrose esters of fatty acids is acceptable up to 120 mg/l in powdered and liquid FSMP containing extensively or fully hydrolysed proteins or amino acids for infants and young children, and in liquid infant formulae and follow-on formulae containing partially hydrolysed proteins intended for infants and young children in good health.

Sodium alginate (E-401)

Sodium alginate has been requested as a stabiliser to be used in FSMP up to a level of 1 g/l, in products intended for infants and young children with metabolic disorders and for general tube-feeding from six months of age onwards (2). The Committee has been informed (2) that its use in combination with other stabilisers and emulsifiers reduces the overall intake of any one additive in products for metabolic disorders, and in tube feeds its use in combination with other thickeners and gelling agents prevents separation of fibre in the liquid feed.

The SCF has already recommended that sodium alginate is acceptable as an additive in weaning foods for infants and young children in good health, for use in desserts and puddings at levels up to 0.5 g/kg (8). It is permitted for these uses under Annex VI of Directive 95/2/EC (1). It has a JECFA group ADI 'not specified' for alginic acid and its ammonium, calcium, potassium and sodium salts (38). No estimates of intake for E-401 are given in the various submissions but intakes from FSMP would almost certainly be higher than that from its use in desserts and puddings for infants and young children in good health. A worst case estimate, assuming that the products concerned are the sole source of nutrition and consumption of 1 000 ml of product at six months rising to 2 000 ml of product by three years of age, gives intakes of 130 - 140 mg/kg b.w./day for E-401.

Conclusion:
The Committee considers that the use of sodium alginate is acceptable up to a level of 1 g/l in FSMP used from four months of age onwards.

Propane-1,2-diol alginate (E-405)

Propane-1,2-diol alginate has been requested as a stabiliser for use up to levels of 0.2 g/l in FSMP intended for young children from one year of age onwards who have cow's milk intolerance or inborn errors of metabolism (2). The Committee has been informed (2) that it has

stabilising properties and is synergistic with other emulsifiers. It enables the development of acidic products containing fat, which means that citrus flavouring may be added to mask the bitter taste of amino acids. It is permitted under Annex IV of Directive 95/2/EC in a variety of foods including FSMP (for children over 3 years of age and adults) up to 1.2 g/l, but is not listed in Annex VI which covers food additives permitted for infants and young children (1).

E-405 was listed in the basic 1974 EEC directive on emulsifiers, stabilisers, thickeners and gelling agents for use in foodstuffs (32) but the SCF did not review E-405 until 1990 when an ADI of 0–25 mg/kg b.w. was allocated (39). JECFA allocated it an ADI of 0–70 mg/kg b.w. in 1993 (40). Estimated intakes of E-405 from nutritionally complete FSMP are 26 27 mg/kg b.w./day for children aged 12–36 months (2), which is marginally above the SCF ADI and below the JECFA ADI. Alginates in general and propane-1,2-diol are of low toxicity and regular consumption just above the ADI would not be expected to pose any health problems.

Conclusion:
The Committee considers that use of propane-1,2-diol alginate is acceptable up to levels of 0.2 g/l in FSMP intended for young children from one year upwards.

Carrageenan (E-407)

Carrageenan has been requested for use in FSMP at levels up to 0.3 g/l in ready-to-use liquid formulae given from birth onwards which contain extensively hydrolysed protein and/or amino acids (2). In these products it acts to increase viscosity, delaying fat separation and so giving a longer shelf life. The products concerned are for children with intractable diarrhoea and other malabsorption conditions and with inborn errors of metabolism (2).

In 1994 carrageenan was regarded by the Committee as not acceptable for use in infant formulae for infants in good health (8) and is currently being reviewed again by the SCF in relation to food use generally. The Committee currently has serious reservations about its use in very young infants because of possible uptake by the immature gut and the possibility of effects on the immature immune system. In the context of FSMP, the Committee notes that medical conditions affecting the permeability of the gut may be of particular importance in any safety consideration. So while the SCF did agree in 1983 that the use of carrageenan was acceptable at levels up to 0.3 g/l in follow-up milks which may be given from four months of age onwards (7), it has more recently stated (8) that it wishes to defer its opinion on the use of carrageenan in weaning foods and may wish to reconsider its earlier opinion on follow-on formulae in the light of the ongoing review mentioned above.

Conclusion:
The Committee is unable to offer an opinion on the acceptability of the use of carrageenan in FSMP until its current review on carrageenan is completed.

Guar gum (E-412)

Guar gum has been requested for use in FSMP at levels up to 10 g/l from birth and 20 g/l from 6 months of age in ready-to-use liquid formulae which contain extensively hydrolysed protein and/or amino acids, and for use at levels up to 1 g/l in ready-to-use liquid formulae containing extensively hydrolysed proteins used from birth for infants in good health. In these products it acts to increase viscosity, delaying fat separation and so giving a longer shelf life. The FSMP are intended for children with intractable diarrhoea, other malabsorption conditions, impairment of the gastrointestinal tract, protein intolerance or inborn errors of metabolism.

Guar gum has a JECFA ADI 'not specified' (34). The SCF has already agreed that the use of guar gum is acceptable at levels up to 1 g/l in follow-up milks which may be given from four months of age onwards (7) and at levels up to 10 g/kg in all weaning foods and up to 20 g/kg in gluten-free, cereal-based weaning foods (13). These uses are all included in Annex VI of Directive 95/2/EC (1). In its 1994 opinion on infant formulae, the SCF reserved its view pending confirmation by the industry of essential need and also noted that no multi-generation study was available (8). The Committee further notes now that guar gum contains a significant protein fraction and that true allergy to guar gum via inhalation and subsequently the oral route has been recorded in exposed workers. The Committee therefore considers that guar gum is not suitable for use in liquid formulae for any age group in infants and young children suffering from protein intolerance, unless it can be shown that the products concerned comply with the conditions set out for products claiming reduced allergenic properties in Annex IV of Directive 91/321/EC (9) as amended by Commission Directive 96/4/EC (56).

Conclusion:
Guar gum is acceptable for use in FSMP from birth onwards, at levels up to 10 g/l in ready-to-use liquid formulae which contain extensively hydrolysed protein and/or amino acids, and at levels up to 1 g/l in ready-to-use liquid formulae containing partially hydrolysed proteins for infants in good health, provided that in the case of products intended for infants and young children with protein intolerance, the addition of guar gum does not jeopardise compliance with the conditions set out for products claiming reduced allergenic properties in Annex IV of Directive 91/321/EC (9) as amended by Commission Directive 96/4/EC (56). The Committee requires further technical justification for the need to use amounts higher than 10 g/l in FSMP and in the meantime no more than 10 g/l should be used.

Xanthan gum (E-415)

Xanthan gum has been requested for use in FSMP at levels up to 1.2 g/l in products based on amino acids and peptides given from birth onwards (2). The Committee has been informed (2) that in these products, some of which are fed by tube, it acts in combination with guar gum to prevent sedimentation of components with low water solubility, such as fibre. The products concerned are for children with impairment of the gastrointestinal tract, protein malabsorption or inborn errors of metabolism.

The SCF changed its ADI of 10 mg/kg b.w. set in 1978 (33) to an ADI 'not specified' in 1989 (41). This was based on lack of toxicity in animal feeding studies and observations in man and the then proposed levels of use (1–5 g/kg in foods and 0.5 g/l in beverages). The SCF has also agreed that the use of xanthan gum is acceptable at levels up to 10 g/kg in all weaning foods and up to 20 g/kg in gluten-free, cereal-based weaning foods (13) and these uses are all included in Annex VI of Directive 95/2/EC (1). It has a JECFA ADI 'not specified' (34). There are no reasons to indicate it is unsuitable for infants from birth onwards.

Conclusion:
The Committee considers that xanthan gum is acceptable for use at levels up to 1.2 g/l in FSMP.

Pectin (E-440)

Pectin has been requested for use in FSMP in liquid and dry formulae at levels up to 10 g/l in ready to feed form for products given from birth onwards for those with diarrhoea resulting from gastrointestinal disorders (2). The Committee has been informed (2) that in these products it acts both as a gelling/thickening agent to delay gastric emptying and serves a physiological function as soluble dietary fibre.

The SCF has already agreed that the use of pectin is acceptable at levels up to 10 g/kg in all weaning foods and up to 20 g/kg in gluten-free, cereal-based weaning foods (13) and is acceptable in follow-on formulae up to 5 g/l (8). These uses are all included in Annex VI of Directive 95/2/EC (1). In its opinion on infant formulae where the level of addition requested was 1 g/l, the SCF has reserved its view pending confirmation by industry of essential need (8). The SCF has endorsed (33) the JECFA ADI 'not specified' (10) and there are no reasons to indicate it is unsuitable for infants from birth onwards.

Conclusion:
The Committee considers that pectin is acceptable for use at levels up to 10 g/l in FSMP.

Sodium carboxymethyl cellulose (E-466)

Sodium carboxymethyl cellulose has been requested for use in FSMP at levels up to 10 g/l in liquid foods given from birth onwards and at levels up to 10 g/kg in solid foods. These products are intended for metabolic disorders, such as inborn errors of fatty acid metabolism (2). The Committee has been informed (2) that it acts as a thickening, gelling and solvation agent resulting in a less 'sandy' mouth feeling.

The SCF has allocated an ADI 'not specified' for modified celluloses as a group which included E-466 (42). The Committee earlier reserved its opinion on a request to use E-466 in weaning foods pending completion of its work on persorption of macromolecular additives but noted that otherwise the toxicological data did not indicate any effects likely to be of concern for infants and young children over weaning age (8). However, the Committee has since been informed that sodium carboxymethyl cellulose in water is in colloidal form and hence is not likely to be persorbed.

Conclusion:
The Committee considers that use of sodium carboxymethyl cellulose is acceptable in FSMP at levels up to 10 g/l in liquid foods and at levels up to 10 g/kg in solid foods.

Glycerol (E-422)

The use of glycerol at *quantum satis* levels has been requested as a non-cariogenic substitute for sugar as a sweetener in concentrated juices based on vegetables and fruits, especially intended for young children. Proposed levels are 5- 15 % in ready-to-drink products. Glycerol would also play a role as a preservative and as a solvent in such drinks (5).

Sweeteners are regulated under Directive 94/35/EC and glycerol is not included in the list of permitted sweeteners (43). Moreover, Article 2.3 of 94/35/EC states that sweeteners shall not be used in foods intended for infants or young children. Other low-calorie sweeteners, such as polyols, are not permitted in food for children under three years of age. The use of food additives in fruit juice in general is also restricted under Directive 95/2/EC, Annex II (1).

JECFA allocated an ADI 'not specified' in 1976, based on the daily intake of glycerol arising from its use, at the levels necessary to achieve the desired effect (44). The SCF agreed with this view and considered glycerol acceptable for use as a solvent in food (45).

Glycerol is rapidly absorbed after ingestion and readily metabolised. It can be converted to fat, shunted into glucose or glycogen synthesis or broken down by glycerokinase in the liver to carbon dioxide and water. Glycerol is reabsorbed in proximal tubuli, and is thus not normally present in urine. However, in the case of a glycerol overload it appears in the urine (46).

Due to its pharmaceutical uses, the adverse effects of glycerol in humans have been studied. They are primarily due to its dehydrating action. The major effects of oral glycerol in otherwise healthy patients are headache, nausea and vomiting (47). Less frequently diarrhoea, thirst, dizziness, and mental confusion can occur. Cardiac arrhythmias have also been reported. Mild osmotic diuresis occurs after oral doses of 1.0- 1.3 g glycerol/kg b.w. (46).

Following oral administration of 0.75- 1.5 g/kg b.w. in a 50- 75 % solution, a relatively rapid increase in serum osmolality occurs, leading to a reduction of intraocular and intracranial pressure (48, 49). This dose level is used as treatment for acute glaucoma in children as well as adults. The effect on intraocular pressure may last for 4 to 10 hours (48).

Thirty minutes after ingestion of glycerol at a dose of 1 g/kg b.w. the intracranial pressure was four times lower than before glycerol administration, and normalised after 4 hours (50).

Medical specialists recommend that glycerol should be used cautiously in diabetic patients (47, 49). The absorption of glycerol is insulin-independent (51). In an insulin-deficient organism glycerol is converted to glucose. Two thirds of this glucose is metabolised in the Kreb's cycle and the remaining one third is released into the circulation. Glycerol administered orally increases the blood glucose level slightly in diabetic and minimally in non-diabetic patients. In addition, when glycerol is substituted as energy equivalent for glucose, it may reduce the blood glucose level and cause hypoglycaemia in diabetics receiving insulin (52). An insulin-dependent man was given 120 ml of 50 % glycerol-solution (i.e. 0.9 g glycerol/kg b.w.) twice during 24 hours as treatment for neovascular glaucoma (51). The patient developed hyperosmolar non-ketonic coma due to dehydration. Since potassium deficiency may accompany diuresis, it is also advised that individuals with congestive heart disease, hepatic or renal disorders should be monitored carefully when exposed to glycerol.

The proposed level of addition of glycerol is 5–15 % in ready-to-drink products. At the higher level of addition, a child three years of age, weighing 15 kg, and drinking 200 ml juice would ingest 2 g glycerol/kg b.w. Thus the amount of glycerol that might be consumed from the proposed use is comparable to the doses used when treating acute glaucoma. The juice concentrate would contain 65 g glycerol/100 ml according to the applicant (5). A three-year-old child ingesting one to two tablespoonfuls of juice concentrate would be exposed to about 1 g glycerol/kg b.w.

Conclusion

The adverse effects reported in non-diabetic patients occur at oral exposure levels which are similar to those which could be consumed as a result of the proposed concentrations in ready-to-drink products. The safety margin, if any, is small compared to exposure levels where the dehydrating effects of glycerol are apparent. In addition, oral glycerol at these intakes in combination with diabetes may have severe consequences. Since low-sugar products are often selected by patients with diabetes, its proposed use seems particularly undesirable. The reported mild diuretic effect of glycerol might also worsen the condition of a child already dehydrated by infection or other diseases. The Committee considers that the use of glycerol is not acceptable in juices intended for young children.

Sodium-L-ascorbate (E-301)

Sodium-L-ascorbate has been requested for use as an antioxidant in coatings of nutrient preparations containing polyunsaturated fatty acids which are used in infant formulae and follow-on formulae for infants and young children in good health and in FSMP for the same age group. The amounts used would give levels up to 75 mg/l (expressed as ascorbic acid) in final foods (5, 53). The maximum level of addition requested is the same as that already permitted for addition of sodium-L-ascorbate as a dietetic additive to infant formulae and follow-on formulae. It is already permitted in certain weaning foods up to 0.3 g/kg under Directive 95/2/EC (1).

Conclusion:
The Committee considers sodium-L-ascorbate to be acceptable as an antioxidant in coatings of nutrient preparations containing polyunsaturated fatty acids in infant formulae and follow-on formulae for infants and young children in good health and in FSMP for the same age group, provided total levels in final food do not exceed 75 mg/l.

L-ascorbyl palmitate (E-304)

L-ascorbyl palmitate has been requested as an antioxidant, for use in combination with tocopherols, at up to 10 mg/l in infant formulae and follow-on formulae for infants and young children in good health and at corresponding levels in FSMP for the same age group([2]). It is already permitted in certain weaning foods up to 0.1 g/kg under Directive 95/2/EC (1). The Committee has previously examined the use of L-ascorbyl palmitate in the context of approved nutritional substances and technological additives (7, 13).

Conclusion:
The Committee considers L-ascorbyl palmitate to be acceptable as an antioxidant up to 10 mg/l in infant formulae and follow-on formulae for infants and young children in good health and at corresponding levels in FSMP for the same age group.

Calcium citrate (E-333)

Calcium citrate has been requested at *quantum satis* levels for use as a firming agent in fruit-based products, with a low sugar content in which low ester pectins have to be used, for infants and young children in good health. The Committee has been informed that addition of extra calcium ions achieves proper gel formation with low ester pectins. Calcium citrate dissolves slowly in the product donating calcium ions and is preferable to other, more rapidly dissolving calcium salts. The actual use level would depend on the calcium present in the fruit and could range from 100–1 000 mg/kg of product, corresponding to 1-0-25 mg added Ca/g added pectin (5). Calcium citrate is already permitted at *quantum satis* levels as a pH adjuster in weaning foods under Directive 95/2/EC (1). It is also allowed as a source of nutrients in cereal based weaning food and baby foods under Directive 96/5/EC (54).

Conclusion:
The Committee considers the use of calcium citrate in fruit-based baby foods with a low sugar content is acceptable provided the amounts added do not exceed the maximum limit for calcium for this category of foods set out in Article 5 of 96/5/EC (54).

Tricalcium phosphate (E-341)

Tricalcium phosphate has been requested as a firming agent for use in combination with pectin to improve gel formation in fruit desserts for infants and young children in good health (4). Calcium salts of orthophosphoric acid are permitted sources of nutrients in weaning foods under Directive 91/321/EEC (9, 13) and in cereal foods and baby foods under Directive 96/5/EC (54). It is also

permitted as an additive in weaning foods up to 1 g/kg (expressed as P_2O_5), either singly or in combination with other phosphates, under Directive 95/2/EC (1).

Conclusion:
The Committee considers that the use of tricalcium phosphate up to a maximum level of 1 g/kg is acceptable as a firming agent in fruit desserts for infants and young children in good health.

Acacia gum (E-414)

Acacia gum, also known as gum arabic, has been requested for use in nutrient preparations, up to a level of 150 g/kg, as a coating for vitamins and trace elements added to infant formulae and follow-on formulae for infants and young children in good health and for FSMP for the same age group (2). It is required in the coating to prevent oxidation of the vitamins and in the case of certain trace elements to prevent them causing peroxidation of unsaturated fatty acids with which they may come in contact (2). The Committee has already considered the use of acacia gum/gum arabic in coatings for vitamin preparations and concluded that use resulting in a low level of carry-over of 10 mg/kg in infant formulae, follow-on formulae and weaning foods was acceptable (55). The same level of addition to the coating of trace elements has been requested as was previously requested for the coating of vitamins (150 g/kg of nutrient preparation). Acacia gum is currently permitted as a direct additive in weaning foods up to 10 mg/kg and in gluten-free, cereal based foods up to 20 mg/kg (9, 1).

Conclusion:
The Committee considers that the use of acacia gum/gum arabic in coatings for nutrient preparations containing trace elements is acceptable provided carry-over levels in infant formulae, follow-on formulae or FSMP do not exceed 10 mg/kg.

Discussion

IDACE proposed that the additives requested be considered according to three categories of product, that is, those which may be used from birth, those from the age of six months and those from the age of one year, reflecting the current market. FSMP can be introduced into the diet of infants, alone or in association with other foods, from the first days of life. The Committee has considered each additive requested for use in FSMP from the health point of view in relation to whether their use is justified in the particular circumstances of feeding infants and young children suffering from particular medical conditions, taking due account of the immaturity of physiological systems in the very young infant, especially in those below 12 weeks of age. For example, since protein hydrolysates may have to be used from birth, there should be no age restriction on the use of acceptable emulsifiers with respect to infants who may be given them. Similarly the Committee sees no reason to make a special distinction at six months of age instead of four months of age. According to Council Directive 91/321/EC, 'infants' means those below 12 months of age and 'young children' those between 12 and 36 months of age (9).

FSMP are intended for use for a very heterogeneous group of medical conditions and it should be noted that the views on safety which the Committee offers in this opinion can only be based on general considerations rather than considerations of the precise nature of the various illnesses and conditions for which they are intended for use.

Since the required levels of addition of emulsifiers in FSMP may be relatively high for some products, the Committee has given careful consideration to whether it would be possible regularly to exceed the ADI for any of the additives requested, in cases where a numerical ADI has been allocated. This does not appear to be the case.

Conclusions and recommendations

The Committee's recommendations have been reached after consideration of each request with respect to the case of need, safety and nutritional requirements, bearing in mind the need to keep the number of additives in foods for infants and young children, whether sick or in good health, to the minimum necessary. In the case of additives requested for use from birth onwards, the Committee was particularly conscious of the physiological situation in infants and very young children. The Committee draws attention to the fact that in some countries, products made with hydrolysed proteins for infants and young children with protein intolerance are not marketed as FSMP but as products for infants and young children in good health.

The Committee's recommendations on the requested additives are summarised in the table below. The opinion on acceptability applies only to the uses and the maximum levels of use specified. It should be noted that in cases where the Committee has recommended that the use of an additive in FSMP is acceptable, this does not imply that its use in foods for infants and young children in good health is also acceptable, unless so stated. Equally, a recommendation that an additive is acceptable for use in foods for infants and young children in good health does not necessarily imply that it is acceptable in FSMP. It follows that should the recommendations in the present opinion be implemented in due course by amendment of Directive 95/2/EC, this may require a change in the wording of Part 4 of Annex VI to the Directive, which currently permits all additives for infants and children in good health to be used in FSMP.

Summary of recommendations

Additive	Requested products and maximum level of use	Opinion: foods for infants and young children in good health	Opinion: FSMP for infants and young children
Sodium-L-ascorbate E-301 (Page 51)	In coatings of NP containing PUFA added to IF, FOF, FSMP. Level not to exceed 75 mg/l (expressed as ascorbic acid) in final foods	Acceptable from birth onwards for the requested conditions of use	Acceptable from birth onwards for the requested conditions of use
L-ascorbyl palmitate E-304 (Page 52)	IF, FOF 10 mg/l, FSMP at same levels	Acceptable from birth onwards for the requested conditions of use	Acceptable from birth onwards for the requested conditions of use
Sodium citrate E-331 (Page 40)	IF, FOF, FSMP 2 g/kg	Acceptable from birth onwards for the requested conditions of use	Acceptable from birth onwards for the requested conditions of use
Potassium citrate E-332 (Page 40)	IF, FOF, FSMP 2 g/kg	Acceptable from birth onwards for the requested conditions of use	Acceptable from birth onwards for the requested conditions of use
Calcium citrate E-333 (Page 52)	WF *quantum satis* in low-sugar fruit-based products	Acceptable up to limit for Ca in 96/5/EC for the requested conditions of use	N/R
Sodium phosphate E-339 (Page 41)	IF, FOF, FSMP 1 g/l expressed as P_2O_5, provided Ca:P ratio within limits in 91/321/EEC	Acceptable from birth onwards for the requested conditions of use	Acceptable from birth onwards for the requested conditions of use
Potassium phosphate E-340 (Page 41)	IF, FOF, FSMP 1 g/l expressed as P_2O_5, provided Ca:P ratio within limits in 91/321/EEC	Acceptable from birth onwards for the requested conditions of use	Acceptable from birth onwards for the requested conditions of use

Additive	Requested products and maximum level of use	Opinion: foods for infants and young children in good health	Opinion: FSMP for infants and young children
Tricalcium phosphate E-341 (Page 52)	WF 1 g/kg in fruit-based desserts	Acceptable for the requested conditions of use	N/R
Sodium alginate E-401 (Page 46)	FSMP 1 g/l	N/R	Acceptable from four months onwards for the requested conditions of use
Propane-1.2-diol alginate E-405 (Page 46)	FSMP 0.2 g/l	N/R	Acceptable from 12 months onwards for the requested conditions of use
Carrageenan E-407 (Page 47)	FSMP 300 mg/l	N/R	Opinion deferred, not acceptable in the meantime
Locust bean gum E-410 (Page 42)	IF, FOF, FSMP 10 g/l in products for reduction of gastro-oesophageal reflex	Not acceptable	Acceptable from birth onwards for the requested conditions of use
Guar gum E-412 (Page 48)	IF, FOF 1 g/l FSMP 10 g/l in liquid formulae containing hydrolysed proteins, peptides or amino acids. 20 g/l from six months of age	Acceptable from birth onwards for the requested conditions of use provided infant formulae comply with conditions in Annex IV of 91/321/EEC as amended by 96/4/EC	Acceptable from birth onwards for the requested conditions of use up to 10 g/l. (20 g/l not acceptable pending technical justification)
Acacia gum E-414 (Page 53)	In coatings of NP containing vitamins and trace elements added to IF, FOF and FSMP. 150 g/kg inNP, carry-over level not to exceed10 mg/kg	Acceptable from birth onwards for the requested conditions of use	Acceptable from birth onwards for the requested conditions of use
Xanthan gum E-415 (Page 48)	FSMP 1.2 g/l	N/R	Acceptable from birth onwards for the requested conditions of use

Additive	Requested products and maximum level of use	Opinion: foods for infants and young children in good health	Opinion: FSMP for infants and young children
Glycerol E-422 (Page 50)	WF *quantum satis* in concentrated juices based on vegetables and fruits	Not acceptable	N/R
Pectin E-440 (Page 49)	FSMP 10 g/l	N/R	Acceptable from birth onwards for the requested conditions of use
Sodium carboxy-methyl cellulose E-466 (Page 49)	FSMP 10 g/l (liquid) 10 g/kg (solid)	N/R	Acceptable from birth onwards for the requested conditions of use
Mono- and di-glycerides of fatty acids E-471 (Page 43)	FSMP 5 g/l	N/R	Acceptable from birth onwards for the requested conditions of use
Citric acid esters of mono- and di-glycerides of fatty acids E-472c (Page 44)	IF, FOF, FSMP 7.5 g/l (dry) 9 g/l (liquid) containing hydrolysed proteins, peptides or amino acids	Acceptable from birth onwards for the requested conditions of use	Acceptable from birth onwards for the requested conditions of use
Diacetyl tartaric acid esters of mono- and di- glycerides of fatty acids E-472e (Page 44)	IF, FOF, FSMP containing hydrolysed proteins, peptides or amino acids. 0.4 g/l (as consumed). WF 5 g/l in gluten-free bakery products for coeliac patients	Opinion deferred. not acceptable in the meantime	Temporarily acceptable from birth onwards for the requested conditions of use
Sucrose esters of fatty acids E-473 (Page 45)	IF, FOF, FSMP containing hydrolysed proteins, peptides or amino acids, 120 mg/l	Acceptable from birth onwards for the requested conditions of use	Acceptable from birth onwards for the requested conditions of use

Additive	Requested products and maximum level of use	Opinion: foods for infants and young children in good health	Opinion: FSMP for infants and young children
Distarch phosphate E-1412 (Page 39)	IF, FOF, FSMP 10 g/l (dry) 22 g/l (liquid)	Opinion deferred, not acceptable in the meantime	Opinion deferred pending submission of case of need, not acceptable in the meantime

Key to summary table

IF = infant formulae } for infants and young
FOF = follow-on formulae } children in
WF = weaning foods } good health
FSMP = foods for special medical purposes
NP = nutrient preparations
N/R = not requested
PUFA = polyunsaturated fatty acids

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AN ADDITIONAL LIST OF MONOMERS AND ADDITIVES USED IN THE MANUFACTURE OF PLASTIC MATERIALS INTENDED TO COME INTO CONTACT WITH FOODSTUFFS

(expressed on 13 June 1997)

1. During its 73rd meeting, the SCF was informed that, in principle, it was the Commission's intention to submit for adoption, at every meeting, a list of substances (monomers and additives) used in the manufacture of plastic materials and articles intended to come into contact with foodstuffs, evaluated or re-evaluated by the Working Group 'Food contact materials (FCM)' of the SCF (SCF-WG) between each plenary session of the SCF.

2. This document contains the evaluations on monomers (Annex 2) and on additives (Annex 3) adopted by the SCF-WG on FCM during the 69th (26–28 February 1997) and the 70th meetings (20–23 May 1997). The polymeric additives were excluded because they will be added in a specific report submitted later.

3. The substances are listed in alphabetical and PM/Ref. No order and the allocation into the various SCF lists is summarised in Annexes 4 and 5. The column headed 'Explanation' gives, for the substances for which the SCF-WG changed a previous classification, a brief explanation for the reason for the change.

4. The SCF endorses the evaluations given by the SCF-WG during the abovementioned meetings. As a memo, the definition of the different lists established by the SCF is included in Annex 1.

5. Because other additives are now under examination and will probably be evaluated in a few meetings, the Commission services intend to postpone the publication of the evaluations given in this document until the remaining substances have been evaluated. However, the evaluations of this document will be communicated to the interested parties (governments, professional organisations etc.).

6. It has to be stressed that the SCF-WG classified these substances according to the general criteria described in the document CS/PM/2147 without any exception and that these general criteria have already been endorsed by the SCF in the past.

Note

This is the first report distributed on the Internet and therefore some changes have been made compared to the previous reports. In fact until now the reports did not contain any substance classified by the SCF-WG in SCF-lists 7–9, because the management of these lists was delegated to the SCF-WG.

To avoid any misunderstanding, for the first time this report contains all the substances classified by SCF into SCF-lists 0–6 and also the substances classified by the SCF-WG into lists 7–9.

It has been noted that the SCF-WG classified into SCF-lists 7–9 during the 69th and 70th meetings the following substances: 15030 (monomer) and 38565/47540/76665/83598 (additives).

The polymeric additives were excluded because they will be added in a specific report to be submitted later for the approval of the SCF. The current classification of these polymeric additives made by the SCF-WG during the 70th meeting (in meeting 69 no polymeric additive was examined) can be found in the minutes of these meetings, which will be issued only after confirmation by the SCF-WG (probably after 15 october 1997).

ANNEX 1

DEFINITION OF THE SCF LISTS

Substances for which the Committee was able to express an opinion

List 0

Substances, e.g. foods, which may be used in the production of plastic materials and articles, e.g. food ingredients and certain substances known from the intermediate metabolism in man and for which an ADI need not be established for this purpose.

List 1

Substances, e.g. food additives, for which an ADI, a temporary ADI (t-ADI), an MTDI, a PMTDI, a PTWI or the classification 'acceptable' has been established by this Committee or by JECFA.

List 2

Substances for which a TDI or a t-TDI has been established by this Committee.

List 3

Substances for which an ADI or a TDI could not be established, but where the present use could be accepted.

Some of these substances are self-limiting because of their organoleptic properties or are volatile and therefore unlikely to be present in the finished product. For other substances with very low migration, a TDI has not been set but the maximum level to be used in any packaging material or a specific limit of migration is stated. This is because the available toxicological data would give a TDI which allows that a specific limit of migration or a composition limit could be fixed at levels very much higher than the maximum likely intakes arising from present uses of the additive.

List 4

Section 4A (for monomers)

Substances for which an ADI or TDI could not be established, but which could be used if the substance migrating into foods or in food simulants is not detectable by an agreed sensitive method.

Section 4B (only for monomers)

Substances for which an ADI or TDI could not be established, but which could be used if the levels of monomer residues in materials and articles intended to come into contact with foodstuffs are reduced as much as possible.

List 4 (for additives)

Substances for which an ADI or TDI could not be established, but which could be used if the substance migrating into foods or in food simulants is not detectable by an agreed sensitive method.

List 5

Substances which should not be used.

Substances for which there were insufficient toxicological or technological data to enable the Committee to express an opinion

List 6

Substances for which there exist suspicions about their toxicity and for which data are lacking or are insufficient.

The allocation of substances to this list is mainly based upon similarity of structure with that of chemical substances already evaluated or known to have functional groups that indicate carcinogenic or other severe toxic properties.

Section 6A
Substances suspected to have carcinogenic properties. These substances should not be detectable in foods or in food simulants by an appropriate sensitive method for each substance.

Section 6B
Substances suspected to have toxic properties (other than carcinogenic). Restrictions may be indicated.

List 7

Substances for which some toxicological data exist, but for which an ADI or a TDI could not be established. The required additional information should be furnished.

List 8

Substances for which no or only scanty and inadequate data were available.

List 9

Substances and groups of substances which could not be evaluated due to lack of specifications (substances) or to lack of adequate description (groups of substances).

Groups of substances should be replaced, where possible, by individual substances actually in use. Polymers for which the data on identity specified in 'SCF Guidelines' are not available.

List W

'Waiting list'. Substances not yet included in the Community lists, as they should be considered 'new' substances, i.e. substances never approved at national level. These substances are not susceptible to be included in the Community lists, lacking the data requested by the Committee.

On the basis of the data lacking the waiting list is subdivided into:

List W7

Substances for which some toxicological data exist, but for which an ADI or a TDI could not be established. The required additional information should be furnished.

List W8

Substances for which no or only scanty and inadequate data were available.

List W9

Substances and groups of substances which could not be evaluated due to lack of specifications (substances) or to lack of adequate description (groups of substances).

APPENDIX

DEADLINES FOR THE TRANSMISSION OF TOXICOLOGICAL DATA

List 2		For substances for which a temporary TDI (t-TDI) has been fixed, the data shall be transmitted as soon as possible.
List 6		1 year after the evaluation of SCF* NB: The deadlines for the transmission of the results of the tests requested are the same of those indicated for list 7.
List 7		1 year after the evaluation of SCF*
	Data on:	
	- hydrolysis	2 years after the evaluation of SCF
	— metagenesis	2 years after the evaluation of SCF
	— migration	2 years after the evaluation of SCF
	Studies:	
	— 28-days	2 years after the evaluation of SCF
	- - 90-days	2 years after the evaluation of SCF
	reproduction	3 years after the evaluation of SCF
	- - - teratogenesis	3 years after the evaluation of SCF
	- - long-term	5 years after the evaluation of SCF
List 8		1 year after the evaluation of SCF* N.B. The deadlines for the transmission of the results of the tests requested are the same of those indicated for list 7.
List 9		1 year after the evaluation of SCF

* The dates labelled with an asterisk refer to the time inside which the Commission has to receive a letter in which the interested person declares his/her intention to transmit the requested data and encloses, for example, a copy of the contractual engagement with the laboratory(ies) of analysis.

ANNEX 2

LIST OF MONOMERS EVALUATED IN THIS REPORT AND THEIR CLASSIFICATION

REF. No	CAS No	NAME	SCF List	SCF OPINION	EXPLANATION
11245	02156-97-0	ACRYLIC ACID, DODECYL ESTER	3	R: 0.05 mg/kg of food. Available: migration data, three (negative) mutagenicity studies. (ISS/TNO SDS, March 1997 = CS/PM/2996/11245). Remark: since high migration into fat has been demonstrated the WG recommends that the Commission take the necessary measures so that the restriction proposed is not exceeded.	Transferred from SCF list 8 to list 3 because new data were available.
12578	-	ALKYL(C1-C4)PHENOLS (consisting of a mixture of mono-, di-, tri- and tetra-alkyl substituted phenols and up to 2 % phenol)	5	Substance which should not be used. Available: inadequate migration data, acute toxicity data on several components of mixture. Evaluation is not possible from the data provided. Sources for the mixture mentioned do not exclude presence of toxic components. Quantitative ranges are lacking for many substances. (RIVM/TNO SDS, February 1997 = CS/PM/2993/12578).	

REF. No	CAS No	NAME	SCF List	SCF OPINION	EXPLANATION
13780	02425-79-8	1,4-BUTANEDIOL BIS(2,3 EPOXY PROPYL) ETHER	4A	Genotoxic similar to glycidylethers. (CS/PM/2958).	Re-examined because in the previous evaluation was classified on the basis of the similarity to the BADGE. The previous classification has been confirmed but the reason was changed from 'similar to BADGE' into 'similar to glycidy-lethers'.
15030	00931-88-4	*CYCLO-OCTENE	W7	Available: inadequate migration data. three (negative) *in vitro* mutagenicity studies. Needed: detailed information on the analytical method for the determination of cyclo-octene in aqueous food simulants, particularly on establishment of detection limit. Migration experiments should be carried out with blends of polymer with maximum concentration of Vestenamer. (RIVM/TNO SDS, February 1997 = CS/PM/2529 REV II/15030).	
15780	00111-90-0	DIETHYLENE-GLYCOL MONOETHYL ETHER	2	Group t-TDI: 0.05 mg/kg b.w. (with 16993=53765, 16996 = 53820, 16999, 17002=53860, 30120, 48030). See references for 16996.	Confirmed.

REF. No	CAS No	NAME	SCF List	SCF OPINION	EXPLANATION
16690	01321-74-0	*DIVINYL BENZENE	6A	R; not detectable in food. Available: two (negative) Ames tests and a mouse inhalation study showing weak clastogenic activity. Needed: detailed description of analytical method to determine residual content. (RIVM/TNO SDS, January 1995 = CS/PM/2572). (ISS SDS, 1997 = CS/PM/2959/16690). Remark: for the toxicity part (based on very low migration) there is no need to ask for further data (awaiting the needed non-tox data to be listed in List 4A).	Confirmed in list 6A but the data requested have been modified. See the SCF opinion.
19990	00079-39-0	METHACRYLAMIDE	4A	The classification is based on the decision made on the structurally related compound acrylamide classified in List 4A and because it is not detected in food/food simulants. Available: migration data, Ames test, dominant lethal assay, two generation reproduction study, teratogenicity study in mice and three neurotoxicity studies. (RIVM/TNO SDS, November 1996 = CS/PM/2961).	Transferred from List 6A to 4A on the basis of the structurally related compound acrylamide, classified in List 4A.

LIST OF ADDITIVES EVALUATED IN THIS REPORT AND THEIR CLASSIFICATION

REF. No	CAS No	NAME	SCF List	SCF OPINION	EXPLANATION
30120	00111-15-9	ACETIC ACID, 2-ETHOXY ETHYL ESTER	3	R: 3 mg/kg of food based on the group t-TDI: 0.05 mg/kg b.w. for it6996.	Confirmed.
38565	90498-90-1	*3,9-BIS(2-(3-(tert.BUTYL-4-HYDROXY-5-METHYL PHENYL)PROPIONYLOXY)-ter.BUTYL)-2,4,8,10-TETRAOXA SPIRO(5.5)UNDECANE	W7	Available: migration data, three month oral rat study, three (negative) *in vitro* mutagenicity studies, high bioaccumulation potential. Needed: data demonstrating absence of bioaccumulation *in vivo* especially in liver and fat. (RIVM SDS, = CS/PM/2994/38565).	
39090	-	N,N-BIS(2-HYDROXY ETHYL)ALKYL (C8-C18) AMINE	2	Group t-TDI: 0.02 mg/kg b.w. (as "free" amine) (with 39120). See references for 39120. Needed: three *in vitro* mutagenicity studies according to guidelines on either PM/Ref No 39090 or 39120). (RIVM SDS, September 1996 = CS/PM/2902. Remark: on the basis of the available studies it remains included in the group t-TDI pending the results of the required mutagenicity studies. If negative it will be classified in List 3 with a restriction of 5 mg/kg of food.	Confirmed but the needed data have been specified.

REF. No	CAS No	NAME	SCF List	SCF OPINION	EXPLANATION
45450	68610-51-5	p-CRESOL-DICYCLO-PENTADIENE-ISOBUTYLENE, COPOLYMER	3	R: 5 mg/kg of food. Available: Migration data, four (negative) *in vitro* mutagenicity studies, 28-day and 3-month oral rat studies, possible bioaccumulation potential. (RIVM/TNO SDS, May 1997 = CS/PM/2802 REV I/45450). Remark: despite the indication of possible bioaccumulation (i.e. log Po/w >4) the WG finds the substance acceptable for the use as described under 3.1 of the document (e.g. in ABS for freezers or very short contact).	Transferred from SCF list 7 into list 3 because new data were available.
47540	27458-90-8	*DI-tert.DODECYL DISULFIDE	W7	Available: adequate migration data, three (negative) *in vitro* mutagenicity studies and a 4-week oral rat study on a closely related compound (di-tert-dodecyl pentasulfide). Needed: the information provided is insufficient to understand the procedure for the determination of the purity of the di-tert-dodecyl disulphide. Information on the method of quantification is missing. (RIVM/TNO SDS, December 1996 = CS/PM/2964).	
48030	00112-34-5	DIETHYLENE-GLYCOL MONOBUTYL ETHER	2	Group t-TDI: 0.05 mg/kg b.w. (with 15780=48050, 16993=53765, 16996=53820, 16999, 17002=53860, 30120, 48050). See references for 16996.	Confirmed.
48050	00111-90-0	DIETHYLENE-GLYCOL MONOETHYL ETHER	2	Group t-TDI: 0.05 mg/kg b.w. (with 15780=48050, 16993=53765, 16996=53820, 16999, 17002=53860, 30120, 48030). See references for 16996.	Confirmed.

REF. No	CAS No	NAME	SCF List	SCF OPINION	EXPLANATION
76665	-	*POLYCYCLOOCTENE	7	Awaiting data on the monomer with PM/Ref. No 15030). Needed: migration of monomer from relevant plastic materials. (RIVM/TNO SDS, February 1997 = CS/PM/2995/76665). Remark: migration of this polymeric additive is covered by the limit for global migration. No need for bioaccumulation data, since no effects were seen in the lymphonodes and Kupfer cells in the 90-day study.	
83598	181314-48-7	*Reaction product of 3-HYDROXY-5,7-DI-tert.BUTYL BENZOFURAN-2-ONE with OXYLENE consisting of 5,7-DI-tert.BUTYL-3-(2,3-DIMETHYL PHENYL)-(3H)-BENZOFURAN-2-ONE AND 5-7-DI-tert. BUTYL-3-(3,4-DIMETHYL PHENYL)-(3H)-BENZOF URAN	W7	Available: Adequate migration data, three (negative) *in vitro* mutagenicity studies, 90-day oral rat study (including examination of the activity of the peroxisome associated enzymes), metabolism studies (*in vitro* and *in vivo*) and an *in vitro* gene mutation assay in bacteria with the dimer, high bioaccumulation potential. Needed: data demonstrating the absence of bioaccumulation *in vivo*. (RIVM/TNO SDS, December 1996 = CS/PM/2965/83598).	

ANNEX 4

LIST OF MONOMERS EVALUATED IN THIS REPORT AND THEIR CLASSIFICATION

REF No	CAS No	NAME	SCF List
11245	02156-97-0	ACRYLIC ACID, DODECYL ESTER	3
12578	-	ALKYL(C1-C4)PHENOLS (consisting of a mixture of mono-, di-, tri and tetra-alkyl substituted phenols and up to 2 % phenol)	5
13780	02425-79-8	1,4-BUTANEDIOL BIS(2,3-EPOXYPROPYL) ETHER	4A
15030	00931-88-4	*CYCLO-OCTENE	W7
15780	00111-90-0	DIETHYLENEGLYCOL MONOETHYL ETHER	2
16690	01321-74-0	*DIVINYLBENZENE	6A
19990	00079-39-0	METHACRYLAMIDE	4A

ANNEX 5

LIST OF ADDITIVES EVALUATED IN THIS REPORT AND THEIR CLASSIFICATION

REF. No	CAS. No	NAME	SCF List
30120	00111-15-9	ACETIC ACID, 2-ETHOXYETHYL ESTER	3
38565	90498-90-1	*3,9-BIS(2-(3-(tert.BUTYL-4-HYDROXY-5-METHYLPHENYL)PROPIONYLOXY)-tert.BUTYL)-2,4,8,10-TETRAOXASPIRO(5,5)UNDECANE	W7
39090	-	N,N-BIS(2-HYDROXYETHYL)ALKYL(C8-C18)AMINE	2
45450	68610-51-5	p-CRESOL-DICYCLOPENTADIENE-ISOBUTYLENE. COPOLYMER	3
47540	27458-90-8	*DI-tert.DODECYL DISULPHIDE	W7
48030	00112-34-5	DIETHYLENEGLYCOL MONOBUTYL ETHER	2
48050	00111-90-0	DIETHYLENEGLYCOL MONOETHYL ETHER	2
76665	-	*VOLYCYCLO-OCTENE	7
83598	181314-48-7	*Reaction product of 3-HYDROXY-5,7-DI-tert.BUTYL BENZOFURAN-2-ONE with OXYLENE consisting of 5,7-DI-tert.BUTYL-3-(2,3-DIMETHYL PHENYL)-(3H)-BENZOFURAN-2-ONE AND 5-7-DI-tert. BUTYL-3-(3,4-DIMETHYL PHENYL)-(3H)-BENZOFURAN	W7

CLARIFICATION AND EXPLANATION OF THE SCF's OPINION OF 7 JUNE 1996 ON BADGE

(expressed on 13 June 1997)

Since the Committee expressed its opinion on BADGE on 7 June 1996 (SCF, 40th series of reports, 1997), a request by the Commission has now been received to provide a more detailed explanation of the following items:

1. the background of the change in the classification from SCF list 4 to list 7, with special emphasis on the mutagenicity data;
2. the characterisation of the identity of the hydrolysis products, which are included in the upper limit of 1 mg/kg of food as a temporary restriction for specific migration of BADGE and its hydrolysis products.

The change in the classification from SCF list 4 to list 7, with special emphasis on the mutagenicity data

RIVM evaluation (Doc. CS/PM/10, February, 1987):

The following studies formed the basis of the first evaluation on BADGE delivered by the WG:

Salmonella reversion assay (Ames test)	Positive	(1)
Gene mutations in mouse lymphoma cells	Positive	(2)
Chromosomal aberrations in Chinese hamster bone marrow *in vivo*	Negative*	(3)
Nuclear anomalies in Chinese hamster bone marrow *in vivo*	Negative*	(4)
Chromosomal aberrations in mouse spermatogonia *in vivo*	Negative	(5)
Chromosomal aberrations in mouse spermatogonia *in vivo*	Equivocal	(6)
Chromosomal aberrations in mouse spermatocytes *in vivo*	Negative	(7)
Dominant lethal test in mouse	Negative	(8)

* results negative but the study had a limited experimental design

The data evaluated showed a mutagenic potential *in vitro*, but were inadequate to demonstrate the lack of activity *in vivo*, because of limitations of the studies on somatic cells (3, 4). The genotoxic profile was judged to be similar to that displayed by other glycidyl ethers. Therefore, in view of

the carcinogenicity of some glycidyl ethers, the substance was classified as a suspected carcinogen (SCF list 4).

First updated evaluation on BADGE (SCF opinion 7 June 1996, 40th series of reports).

For this updated evaluation some additional mutagenicity studies were considered in the technical document prepared by the WG on BADGE (CS/PM/2787).

Test for the induction of unscheduled DNA synthesis in *vitro*, in human white blood cells	Negative	(9)
Test for the induction of structural chromosomal aberrations in rat liver cells *in vitro*	Positive	(10)
Test for neoplastic transformation *in vitro* in baby hamster kidney cells	Positive	(11)
Micronucleus test in female B6D2F1 mice	Negative	(12)
Analysis of DNA single strand breaks in rat liver cells after single oral administration	Negative	(13)

BADGE was evaluated as mutagenic *in vitro*, and negative in *in vivo* studies which were again judged as insufficient to demonstrate lack of activity on somatic cells (because of the limited experimental design of the studies described in (3) and (4), or because the full data were not available for independent evaluation as for (12) and (13), but were only quoted in the form of summaries in a review, (14)). Therefore, further *in vivo* studies (chromosomal aberrations in bone marrow and DNA damage in liver) were requested.

Metabolic data provided evidence for extensive and rapid metabolic detoxification, and a more recent DNA binding study showed very low covalent binding to skin DNA after topical application (15). Accordingly it was expected that the genotoxic activity of BADGE *in vivo* would be weak, or non-existent. Consequently, BADGE was provisionally reclassified in list 7 (SCF opinion, 7 June 1996).

Since, the opinion on BADGE of 7 June 1996 the full reports of the studies (12, 13) have been made available and were considered by the WG (CS/PM/2967, February 1997). The data showed absence of mutagenic activity in liver and bone marrow *in vivo*.

Characterisation of the identity of the hydrolysis products, which are included in the upper limit of 1 mg/kg of food as a temporary restriction for specific migration of BADGE and its hydrolysis products

In order to reach a view on which hydrolysis products should be included in the upper limit of 1 mg/kg of food as a temporary restriction for BADGE and its hydrolysis products, the following substances have to be considered:

A: BADGE
B: monoadduct, in presence of water and acid: 2-(4-(2,3-epoxypropanyloxy) phenyl)-2-(4-(2,3-dihydroxypropanyloxy)phenyl) propane
C: diadduct: 2,2-bis(4-(2,3-dihydroxypropanyl)phenyl)propane
D: monoadduct of BADGE with HCl
E: diadduct of BADGE with HCl

Recent and ongoing studies have shown that the following substances can be found in the coatings, foodstuffs and simulants:

Coating:	A, D, E (D, E in some special coatings)
Foodstuffs:	A, B, C (A → B → C) and D, E (as migrants from special coatings)
Simulants:	A, B, C (A → B → C)

The Committee noted that in coatings residual amounts of compound A and, depending on the type of coating, also the compounds D and E have been detected. Analyses have shown that these residuals A, D and E may migrate into any type of food whether aqueous or fatty. Compound A has been shown to hydrolyse in aqueous foods to B and subsequently to C. However, in fatty media compound A is stable. If food contains chloride ions, it is theoretically possible that compound A may be transformed into compound D and subsequently E.

The Committee therefore recommends as follows on which substances should be included in the limit for BADGE and its hydrolysis products. There is some experimental evidence that compound C may be of minor toxicological relevance and therefore need not be considered when assessing compliance of foodstuffs with the SCF group restriction. Consequently, determination of compliance with the SCF group restriction would require that only the sum of A and B be considered in the case of examination of foodstuffs. However, in the case of examination of food contact materials by aqueous food simulants, the sum of A, B and C should be determined, because in this case more A and B would be converted to C as compared with the situation in foodstuffs.

The compounds D and E in foodstuffs are of concern because of their structural analogy to the genotoxic monochloropropanediol (MCPD). The Committee will address this issue as soon as it has confirmation of their presence and their levels in food.

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European Commission

Reports of the Scientific Committee for Food
(43rd series)

Luxembourg: Office for Official Publications of the European Communities

1999 — vi, 80 pp. —16.2 x 22.9 cm

Food science and techniques series

ISBN 92-828-5887-1

The Scientific Committee for Food was established by Commission Decision 74/234/EEC of 16 April 1974 (OJ L 136, 20.5.1974, p. 1), replaced by Commission Decision 95/273/EC of 6 July 1995 (OJ L 167, 18.7.1995, p. 22), to advise the Commission on any problem relating to the protection of the health and safety of persons arising or likely to arise from the consumption of food, in particular on nutritional, hygienic and toxicological issues.

The members are independent persons, highly qualified in the fields associated with medicine, nutrition, toxicology, biology, chemistry, or other similar disciplines.

Responsibility for the Secretariat of the Scientific Committee for Food was transferred form Directorate-General III 'Industry' to Directorate-General XXIV 'Consumer Policy and Consumer Health Protection' with effect from 1 April 1997.

The present report deals with:

- Arsenic, barium, fluoride, boron and manganese in natural mineral waters
- Starch aluminium octenyl succinate (SAOS)
- The additional information from the Austrian authorities concerning the marketing of Ciba-Geigy maize
- Actilight – a fructo-oligosaccharide (FOS)
- Diacetyltartaric acid esters of mono- and diglycerides (DATEM E-472e)
- Canthaxanthin
- A request for the use of algal beta-carotene as a food colour
- Certain additives for use in foods for infants and young children in good health and in foods for special medical purposes for infants and young children
- An additional list of monomers and additives used in the manufacture of plastics materials intended to come into contact with foodstuffs
- Clarification and explanation of the SCF's opinion of 7 June 1996 on BADGE