European Food Safety Authority

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# SCIENTIFIC OPINION

## Pantethine as source for pantothenic acid added as a nutritional substance in food supplements[1]

## Scientific Opinion of the Panel on Food Additives and Nutrient Sources added to Food (ANS)

**(Question No EFSA-Q-2006-227)**

## Adopted on 23 September 2008

**PANEL MEMBERS**

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**SUMMARY**

Following a request from the Commission, the Panel on Food Additives, Nutrients and Food Supplements (ANS) was asked to evaluate the safety and bioavailability of pantethine as a source for pantothenic acid when added for nutritional purposes in food supplements.

Pantothenic acid has been evaluated by the Scientific Committee on Food in 2002 (SCF, 2002) who concluded that no numerical upper limit could be derived.

The present opinion deals only with the safety and bioavailability of a particular source of pantothenic acid, intended to be added to food supplements. The safety of pantothenic acid itself, in terms of amounts that may be consumed, is outside the remit of this Panel.

Pantethine is the disulphide of pantetheine, the metabolic substrate which constitutes the active part of coenzyme A and acyl carrier proteins. After oral intake, pantethine can be metabolized into pantetheine in the intestinal lumen. Pantetheine is, in its turn, absorbed and hydrolysed in the intestinal mucosa cells into pantothenic acid.

[1] For citation purposes: Scientific Opinion of the Panel on Food Additives and Nutrient Sources added to Food (ANS) on a request from the Commission on pantethine as a source for pantothenic acid added as a nutritional substance in food supplements. *The EFSA Journal* (2008) 865, 1-20.

Pantethine is proposed to be used as a source of pantothenic acid by food supplement manufacturers as an ingredient in tablets, caplets, capsules, chewable tablets, effervescent powders and liquids that are food supplements.

The petitioner indicates that in Europe food supplement products containing pantethine have been on the market since at least 1984 and used typically up to 10 mg pantethine per day in multivitamin/mineral supplements or vitamin B complex supplements. Also EU food supplements supplying pantothenic acid in the form of calcium pantothenate can be found at levels up to around 500 mg.

In the US, pantethine is on the market in dietary supplements at much higher levels (600-900 mg/day) to reflect the different market requirements and expectations.

The petitioner does not provide actual proposed use levels. The Panel notes that if use levels of pantethine are based on use levels of pantothenic acid or on levels of pantethine used in the US, use levels up to 500 to 900 mg pantethine/day would be foreseen, equivalent to intakes of 8.3 to 15 mg/kg bw/day for a 60 kg person. Daily intakes of 10 mg pantethine/day would amount to 0.167 mg/kg bw/day for a 60 kg person.

Data in the literature indicate that pantethine is about equally rapidly absorbed as calcium pantothenate when given orally to rats (Ono *et al.,* 1974) with plasma levels being at their maximum values already upon 2 hours post dosing and only slightly (18%) higher in animals dosed with pantethine at a dose that was twice as high as that of calcium pantothenate based on pantothenic acid equivalents.

Another study reports that in healthy subjects, pantethine is retained longer in the blood and has more tissue affinity than pantothenic acid.

The toxicity studies reported by the petitioner for pantethine are based on the dossier or on limited unpublished translations of studies published in Japanese journals, performed in the period 1966-1968. No original data were available for evaluation.

The petitioner provided results of toxicity studies on pantethine from which it can be concluded that pantethine is not genotoxic, not carcinogenic and that no developmental toxicity was observed in mice and rats up to doses of 600 mg/kg bw/day, and in rabbits up to 120 mg/kg bw/day.

From subchronic toxicity studies in rats and dogs, the Panel identified NOAELs of respectively 36 and 50 mg pantethine/kg bw/day.

Daily intakes up to 8.3 to 15 mg/kg bw/day would result in margins of safety compared to the NOAELs from the animal studies of only 2.4 to 6.0. Daily intakes of 10 mg/day would amount to 0.167 mg/kg bw/day for a 60 kg person and would give rise to margins of safety of 216 to 300.

Results from clinical studies reveal that pantethine is well tolerated although there are occasional reports of heartburn, mild pruritis, gastrointestinal discomfort and diarrhea, most frequently when administered in higher doses i.e. 350-1200 mg/day and higher.

The Panel concludes that the bioavailability of pantothenic acid from pantethine upon oral intake might be comparable to or lower than that of pantothenic acid.

Given the facts that:

i) the bioavailability studies in rats indicate that part of the pantethine might be absorbed unmodified and that in first instance only part may be converted to pantothenic acid,

ii) the metabolic fate of an oral dose of pantethine in humans has not been clearly described,

iii) the margin of safety between the NOAELs from the animal studies and the exposure resulting from use levels of 500 to 900 mg/day of pantethine is less than 10 and,

iv) results from clinical studies with pantethine indicate that at dose levels between 350-1200 mg/day, occasional cases of heartburn, mild pruritis, gastrointestinal discomfort and diarrhea occur,

the Panel concludes that the safety in use of pantethine as a source for pantothenic acid in food supplements intended for the general population at levels of use of 500 to 900 mg pantethine/day is not demonstrated.

The Panel also concludes that when use levels are 10 mg/day (0.167 mg/kg bw for a 60 kg person), as reported by the petitioner to be used typically in multivitamin/mineral supplements or vitamin B complex supplements in European food supplement products, the margin of safety would be 216 to 300. In view of this margin of safety, the Panel concluded that the use of pantethine as a source of pantothenic acid under these conditions would not be of safety concern.

**Key words:**

Food supplements, pantethine, CAS Registry Number 16816-67-4, pantothenic acid.

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## BACKGROUND AS PROVIDED BY THE COMMISSION

The European Community legislation lists nutritional substances that may be used for nutritional purposes in certain categories of foods as sources of certain nutrients.

The Commission has received a request for the evaluation of pantethine added for nutritional purposes to food supplements. The relevant Community legislative measure is:

- Directive 2002/46/EC of the European Parliament and of the Council on the approximation of the laws of the Member States relating to food supplements[2].

## TERMS OF REFERENCE AS PROVIDED BY THE COMMISSION

In accordance with Article 29 (1) (a) of Regulation (EC) No 178/2002, the European Commission asks the European Food Safety Authority to provide a scientific opinion, based on its consideration of the safety and bioavailability of pantethine added for nutritional purposes in food supplements.

## ACKNOWLEDGEMENTS

The European Food Safety Authority wishes to thank the members of the Working Group B on food additives and nutrient sources of the Scientific Panel on food additives and nutrient sources added to food for the preparation of this opinion: D. Boskou, U.R. Charrondiere, B. Dusemund, D. Gott, T. Hallas-Møller, K. F. A. M. Hulshof, J. König, D. Parent-Massin, I. Rietjens, G.J.A. Speijers, P. Tobback, T. Verguieva, R. Woutersen.

**ASSESSMENT**

## 1. Introduction

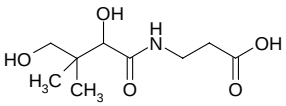
The present opinion deals only with the safety and bioavailability of a particular source of pantothenic acid intended for the general population, to be used in food supplements. The safety of pantothenic acid itself, in terms of amounts that may be consumed, is outside the remit of this Panel.

## 2. Technical data

### 2.1. Chemistry

Pantethine is the disulphide of pantetheine, the metabolic substrate which constitutes the active part of coenzyme A and acyl carrier proteins.

Pantethine is a synonym for D-bis-(N-pantothenyl-β-aminoethyl)-disulphide. Its CAS registry number is 16816-67-4 and the petitioner mentioned 10 other synonyms. The molecular weight is 554.72 g/mol, the molecular formula is $C_{22}H_{42}N_4O_8S_2$ and the molecular structure is presented below together with the structure of pantetheine and panthenoic acid for comparison.



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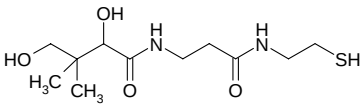
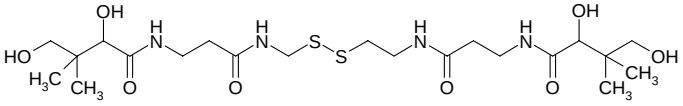


Pantetheine



Pantethine

### 2.2. Specifications

The petitioner indicates that pantethine is available in two forms, and the opinion refers to both pantethine powder (pantethine 55% powder) and pantethine liquid (pantethine 80% solution).

The pantethine in the 55% powder complies with the purity criteria in the Japanese Pharmacopoeia. The preparation contains in addition to 55% pantethine, 33% colloidal silicon

dioxide and 12% microcrystalline cellulose and is a white powder. The specifications provided for the preparation by the petitioner include heavy metals ≤ 10 mg/kg and arsenic ≤ 1 mg/kg.

The pantethine in the 80% solution also complies with the specifications of the Japanese Pharmacopoeia and is a solution of pantethine in water giving rise to a colourless to pale yellow, clear, viscous liquid of pH 7.

The specifications provided by the petitioner for the pantethine 55% powder include heavy metals ≤ 1 mg/kg and arsenic ≤ 1 mg/kg.

The specifications provided by the petitioner for the pantethine 80% solution include heavy metals ≤ 10 mg/kg, arsenic ≤ 1 mg/kg, loss on drying ≤ 0.1 %.

### 2.3. Manufacturing Process

The manufacturing process is described by the petitioner. Pantethine has been manufactured from D-pantolactone, β-aminopropionitrile and cysteamine under stringent pharmaceutical GMP in Japan. The pantethine 80% solution is manufactured first and pantethine 55% powder is as a result of a further procedure on the liquid.

### 2.4. Methods of analysis in food

The petitioner indicates that there are no analytical methods available for the determination of pantethine in food. The petitioner provided a method for analysis of pantethine in food supplements.

### 2.5. Reaction and fate in foods to which the source is added

The petitioner reports that both pantethine 55% powder and pantethine 80% solution are stable in the finished product.

### 2.6. Case of need and intended levels of use

The petitioner indicates that pantethine is to be used as a source of pantothenic acid by food supplement manufacturers as an ingredient in tablets, caplets, capsules, chewable tablets, effervescent powders and liquids that are food supplements. The method of incorporation is determined by the individual manufacturers as appropriate for the particular type of finished products.
The petitioner also indicates that the quantity of pantothenic acid (as pantethine) to be added to food supplements will be determined by individual formulators.

### 2.7. Exposure

The petitioner indicates that in Europe food supplement products containing pantethine have been on the market since at least 1984 and used typically up to 10 mg pantethine per day in

multivitamin/mineral supplements or vitamin B complex supplements. Also EU food supplements supplying pantothenic acid in the form of calcium pantothenate can be found at levels up to around 500 mg.

In the US, pantethine is on the market in dietary supplements at much higher levels (600-900 mg/day) to reflect the different market requirements and expectations.

The petitioner does not provide actual proposed use levels. The Panel notes that if use levels of pantethine are based on use levels of pantothenic acid or on levels of pantethine used in the US, use levels up to 500 to 900 mg pantethine/day would be foreseen, equivalent to intakes of 8.3 to 15 mg/kg bw/day for a 60 kg person. Daily intakes of 10 mg pantethine/day would amount to 0.167 mg/kg bw/day for a 60 kg person.

According to the SCF (2002), average intakes of pantothenic acid in adults range between 3-12 mg/day. Few data on the distribution of intakes from dietary and supplement sources are available. In Ireland, the 97.5$^{th}$ percentile of intakes from all sources (food and food supplements) was reported as 12.5 mg in men and 14.4 mg in women, and from food sources only, as 10.4 mg and 7.2 mg per day, respectively (IUNA, 2001). For the UK the 97.5$^{th}$ percentile of intakes reported for all sources (food and food supplements) was 11.2 mg in men and 9.1 mg in women, and from food sources only as 10.5 mg and 7.7 mg per day, respectively (Gregory *et al.*, 1990).

## 2.8. Information on existing authorisations and evaluations

The Scientific Committee on Food concluded in 1993 that there is no information on intake below which deficiency is likely, nor adequate evidence to establish Population Reference Intakes (PRI) (SCF, 1993) In the US, new guidelines recommend an adequate intake for adults of 5 mg/day (IOM, 2006). The societies for Nutrition of Germany, Austria and Switzerland jointly established an adequate daily intake of 6 mg pantothenic acid for adults (D-A-CH, 2000).

The SCF has issued an opinion (SCF, 2002) on the Tolerable Upper Intake level of pantothenic acid and concluded that:

> "Owing to the lack of systematic oral response intake studies and the very low toxicity of pantothenic acid (calcium pantothenate or panthenol) no LOAEL or NOAEL can be established and no numerical UL can be derived."
>
> "Although it is not possible to derive a numerical UL for pantothenic acid evidence available from clinical studies using high doses of pantothenic acid indicates that intakes considerably in excess of current levels of intake from all sources do not represent a health risk for the general population."

The UK Expert Group on Vitamins and Minerals (EVM) concluded that there are insufficient data from human or animal studies to establish a Safe Upper Level for pantothenic acid. They indicated that for guidance purposes only, a supplemental daily intake of 200 mg (equivalent to 3.3 mg/kg bw/day for a 60 kg adult), in addition to that present in the diet, would not be expected to produce adverse effects in the general population. Assuming a maximum dietary intake of 10 mg/day, this would equate to a total intake of 210 mg/day, or 3.5 mg/kg bw/day for a 60 kg adult (EVM, 2003).

Also the US Food and Nutrition Board did not establish a Tolerable Upper Intake Level (UL) for adults (FNB, 1998)

In the Council Directive 90/496/EEC on nutrition labelling for foodstuffs, the Recommended Daily Allowances given for pantothenic acid is 6 mg (EEC, 1990).

## 3. Biological and toxicological data

### 3.1. Bioavailability of pantothenic acid from its pantethine source

Ingested pantethine is reduced to pantetheine with reduced glutathione (GSH) as cofactor (Durr and Cortas, 1964). One equivalent of pantethine results in two equivalents pantetheine in the intestinal lumen. Pantetheine is in turn absorbed from the intestinal lumen by mucosa cells (van den Berg, 1997; Bender, 2003). Although in earlier studies simple diffusion was reported to be the main transport system of pantetheine, there is now ample evidence that transport is affected in mammals through a saturable, sodium dependent transport system in the jejunum (SCF, 2002; Bender, 2003). In the intestinal mucosa cells pantetheine is hydrolysed rapidly to pantothenic acid by pantotheinase (Bender, 2003).

The absorption and distribution of pantethine was studied by Tachizawa *et al.* (1979) using jejunum and ileum rat intestinal loops. The results indicated that pantethine is absorbed from the intestine in its unmodified form and as pantothenic acid. $^{14}$C-labelled pantethine was given at a dose of 200 mg/kg bw orally to rats, and two hours after the administration, radioactivity was detected in whole blood, aorta, kidney and liver.

In another study the changes in blood concentration and the urinary excretion of free and bound pantothenic acid were studied after oral and intravenous administration of pantethine (21.6 µmoles/kg equivalent to 43.2 µmoles pantothenic acid equivalents/kg) to rats (Ono *et al.,* 1974). The results were compared with those obtained after administration of calcium-pantothenate (Ca-pantothenate, 21.6 µmoles/kg).

The concentration of total pantothenic acid in blood expressed in nmoles pantothenic acid equivalents/ml reached its maximum levels 2-4 hours after administration and amounted at 2 hours after administration to 2.52 $\pm$ 0.03 nmoles pantothenic acid equivalents/ml for the control, 4.06 $\pm$ 0.16 nmoles pantothenic acid equivalents/ml for the pantethine group and 3.43 $\pm$ 0.07 nmoles pantothenic acid equivalents/ml for the Ca-pantothenate group. Thus, the concentration of total pantothenic acid in blood was statistically significantly but only 18% higher in the pantethine group than in the Ca-pantothenate group after oral administration of vitamins, whereas on the basis of pantothenic acid equivalents the dose of pantethine was 2-fold higher than that of pantothenic acid. The excretion rates of total pantothenic acid in 24-hours urines were 29±3% and 18±2% of the dose rates of the pantethine and Ca-pantothenate groups, respectively. The authors conclude that these findings indicate that pantethine is more absorbable through the gastrointestinal wall of the rat than Ca-pantothenate. No significant difference was, however, found in the amounts of bound pantothenic acid in blood between these two groups. Pantethine was found to be converted to some extent to pantothenic acid by an enzyme during passing the intestinal mucosa.

After an intravenous injection of pantethine, about 50% of total pantothenic acid in blood was pantethine and the rest was free pantothenic acid, while little bound pantothenic acid was detected after injection of Ca-pantothenate. The decrease in total pantothenic acid in blood was significantly delayed in the early period after injection in the pantethine group when compared with that of Ca-pantothenate group, although both compounds were almost completely eliminated in 24 hour urines after injection (Ono *et al*., 1974).

Three Wistar rats fed with solid feed containing radioactive pantethine (500 microgram 1.24 x $10^4$ dpm) excreted 15-35% of the radioactivity within a week with the amount of radioactivity excreted into urine being about double of that excreted into feces (Nakamura and Tamura, 1972).

Shigeta *et al.* (1966), studying urinary excretion of pantothenic acid and pantethine in nine healthy human subjects aged 20-25 years following intramuscular injection of 20 mg each of calcium pantothenate (equivalent to 17 mg of pantothenic acid) or pantethine (equivalent to 15 mg of pantothenic acid), have concluded that pantethine, in healthy subjects, is retained longer in the blood and has more tissue affinity than pantothenic acid. Intact pantethine was not detected in the urine.

The petitioner indicates that it is generally accepted that a portion of pantethine is converted to pantetheine and a portion is degraded to pantothenic acid prior to intestinal absorption (Kelly, 1997). The pantetheine and pantothenic acid can subsequently enter normal biochemical pathways in which pantetheine can be phosphorylated to provide the active moiety of Coenzyme A (CoA) and acyl carrier protein (ACP) (Kelly, 1997).

The metabolic activity of pantethine is due to its role in the synthesis of CoA and acyl carrier proteins, ACP. CoA is a cofactor in over 70 enzymatic pathways, including fatty acid oxidation, carbohydrate metabolism, pyruvate degradation, amino acid catabolism. ACP is an essential component of the fatty acid synthase complex required for fatty acid elongation.

The metabolic fate of an oral dose of pantethine in humans has not been clearly described (Kelly, 1997).

### 3.2. Toxicological data

The toxicity studies reported by the petitioner for pantethine are based on the dossier from the petitioner or on limited unpublished translations of studies published in Japanese journals, performed in the period 1966-1968 (Oshima *et al.,* 1966; Akimoto *et al.,* 1966; Morita *et al.,* 1968). The description of the studies below in section 3.2.1 to 3.2.5 is based on the text provided by the petitioner and no original data were available for evaluation.

#### 3.2.1 Acute toxicity

The acute oral toxicity of pantethine was determined in male and female dd-YF mice and shown to be higher than 10 g/kg bw (Akimoto *et al.,* 1966).

#### 3.2.2 Sub-chronic toxicity

The petitioner reports a six-month oral toxicity study of pantethine was conducted in male rats (gavage) (10 rats per group) at doses ranging from 4 to 18,000 mg/kg bw/day (Akimoto *et al.,* 1966). Rats receiving 8,000 and 18,000 mg pantethine/kg bw/day all died prematurely within 14 weeks of treatment due to diarrhea and malnutrition. No gross abnormalities were observed at 800 mg pantethine/kg bw/day. Dose related clinical signs of intolerance included diarrhea, emaciation and ruffled coat. Decreased body weight gain, decreased food consumption and increased water intake were noted at the two highest doses. In the 18,000 mg pantethine/kg bw group, basophilic granules were found in 3 to 5 % of neutrophils; no

other changes were observed in the other hematologic parameters. The weight of the spleen, adrenals, kidneys and testes was slightly increased in the 800 mg/kg bw/day group. In the 8,000 and 18,000 mg pantethine/kg bw group, the spleen and thymus were reduced in weight, the adrenals were increased in weight, the intestinal mucosa was thin and the number of lymphocytes was decreased in the spleen and thymus.

The petitioner also reports that a 54-week chronic oral toxicity study of pantethine was conducted in male and female rats (15 animals per group). The average daily drug administered in the drinking water was 35.8, 143.7 and 279.3 mg/kg bw/day for male and 58.0, 213.8 and 411.8 mg/kg bw/day for females. The petitioner indicates that no adverse effects were noted in clinical signs, bodyweight and histopathology. Lower haemoglobin concentration and decreased number of erythrocytes were observed in males receiving 143.7 mg/kg bw/day and females receiving 411.8 mg/kg bw/day of pantethine. Lower cholesterol concentration was considered to be a pharmacological effect of pantethine in the two higher groups. The Panel identifies a NOAEL of 36 mg/kg bw/day.

A one year chronic toxicity study of pantethine conducted in male and female beagle dogs at daily doses of 50, 200 and 800 mg/kg bw/day was also reported by the petitioner. Pantethine was given mixed in chicken meat. No animal died and no effect of treatment on body weight was observed during the study. The animals administered 800 mg/kg bw/day demonstrated translucent excretion of whitish soft stool or diarrhea. There was a tendency toward lower haemoglobin concentration and haematocrit level in the animals receiving the 800 mg/kg bw/day dose. The blood chemistry, urinalysis, ECG, eyeground and indocyanin green tests revealed no abnormality in any of the animals. The relative liver weights in males receiving 200 mg/kg bw/day and the relative kidney weights of females administered 800 mg/kg bw/day of pantethine were significantly increased. Hemosiderosis and hematopoiesis in the spleen of the animals receiving 800 mg/kg bw/day of pantethine was noted. Extramedullary haematopoiesis was also observed in a few animals administered 200 mg/kg bw/day. Hemosiderin deposits in the liver were detected in one male and one female receiving 800 mg/kg bw/day of pantethine. The Panel concludes that the NOAEL in dogs was 50 mg/kg bw/day when administered daily for one year.

### 3.2.3 Reproductive and developmental toxicity

The petitioner describes a study of Oshima *et al*. (1966) in which the effects of orally administered pantethine to pregnant ddYF mice and Wistar-Imaichi rats during the organogenesis period of their fetuses were investigated (Oshima *et al.,* 1966). Pantethine was administered once daily by gastric incubation for seven consecutive days from day 6 to day 12 of gestation to rats and mice. The doses in mice were 0 (control), 60 to 600 mg/kg bw/day and in rats 0 (control), 60, 300 and 600 mg/kg bw/day. The following parameters were recorded in this study in both species: body weight gain of mothers, number of implantations, number of live fetuses, number of dead fetuses, body weight of fetuses and pups up to 21 days postnatally, body length and tail length of live fetuses, external and internal abnormalities, fusion of occipital bone and first cervical vertebra, split of first and second cervical vertebral arch, absence or incomplete formation of the ribs, asymmetry of the ribs, asymmetry of sternebrae, number of ossified caudal vertebrae, length of major and minor axes of the femur, and rate of rearing of pups.

In mice that received 60 mg/kg bw/day of pantethine, the mean body weight at birth and at 21 days and the mean body length of live fetuses were statistically significantly greater when compared to the control group. The petitioner indicates that there were 5.6% of fetuses with

incomplete formation of ribs, this was statistically significantly lower than in the control group (17.1%). The mean number of ossified caudal vertebrae was statistically significantly greater when compared with the control group. In mice that received 600 mg/kg bw/day of pantethine, the mean body weight, body length and tail length of live fetuses at birth were all statistically significantly greater than those of the control group. There were 8.2% of fetuses with incomplete formation of ribs, this was statistically significantly lower than in the control group (17.1%). This mean number of ossified caudal vertebrae and the length of the major axis of the femur were statistically significantly greater than those of the control group. There were no significant differences in all the other parameters measured in the mice.

In rats that received 60 mg/kg bw/day of pantethine, the tail length of live fetuses was statistically significantly longer than those in the control group. No external abnormality was observed in any of the groups. In rats that received 300 mg/kg bw/day of pantethine the mean body weight, mean body length, mean tail length of live fetuses at birth and the number of ossified caudal vertebrae were statistically significantly increased when compared to the control group. In rats that received 600 mg/kg bw/day of pantethine the mean body weight, mean body length and mean tail length of live fetuses at birth and the mean body weight at 21 days were statistically significantly increased when compared to the control group. None of the other parameters measured in the rats were significantly different between the various groups.

It is concluded by the authors of the study that orally administered pantethine to pregnant female mice and rats up to dose levels of 600 mg/kg bw/day has no teratogenic effects.

The petitioner describes a study performed by Morita *et al.* (1968) (paper in Japanese, English translation provided) who studied the effects of calcium pantothenate and pantethine on the delivery rate and litter size of adult ddYF mice. Mice were continually fed on a basal diet (control), a calcium pantothenate (258 mg/kg) basal diet and a pantethine -added (300 mg/kg) basal diet, respectively. The addition of calcium pantothenate or pantethine to the basal diet did not decrease but rather increased the delivery rate and litter size.

The effects of oral administration of pantethine to pregnant Japanese albino rabbits during the organogenesis period were investigated. Pantethine was administered once a day by gavage as an aqueous solution to four groups of 10 to13 pregnant rabbits from day 6 to day 18 of gestation. The doses were 0, 12, 40, and 120 mg/kg bw/day. The various parameters recorded were: Food intake, body weight, number of corpora lutea, number of implantations, live births, fetal mortality, viability of newborns, body weight of fetuses, gross external and internal anomalies, and ossification of skeletons of fetuses.

There was a trend toward decreased food intake in the pantethine treated groups but this was not statistically significant except for one brief period (days 5 and 16) in the group that received the highest dose. The mean body weight of rabbits that received 12 mg/kg bw/day of pantethine was compared to the control group. There was a slight decrease in mean body weight in the 40 mg/kg bw/day group and a statistically significantly lower mean body weight in the group that received 120 mg/kg bw/day of pantethine when compared to the control group.

One rabbit in the 40 mg/kg bw/day group and three rabbits each in the 12 mg/kg bw/day, 120 mg/kg bw/day and the control groups did not get pregnant. The implantation rate in all groups was similar, but the mean number of corpora lutea and of implants in the 120 mg/kg bw/day group was slightly lower than those in the control group. The average number of live fetuses and their mean body weights were also slightly deceased in the 120 mg/kg bw/day group, but

these differences were not statistically significant. The survival rate of the fetuses was the same in all four groups.

One case of a single nostril was observed in the control group and one case of spinal bifida with deformities of the forelimbs was recorded in the 40 mg/kg bw/day group.

No visceral anomalies were detected in any of the groups.

Incomplete ossification of the sternebrae was noted in all groups and non-ossified metacarpals were observed in the control group as well as in the group that received 120 mg/kg bw/day, group that demonstrated an increase in the number of ossified caudal vertebrae. There were no statistically significant differences between groups in the incidence of formation of a 13$^{th}$ rib, separated or dislocated sternebrae, hypoplasia or separation of cervical vertebrae, shortening of cervical vertebral arches and asymmetry of vertebral arches. There were a few skeletal anomalies such as shortening of the first ribs in the control groups, two case of fusion of the cervical vertebral arched with deformation of the forelimbs in the 40 mg/kg bw/day group.

It is concluded that decreased food intake and mean body weight occurred in pregnant rabbits when pantethine was administered orally between day 6 to day 18 of gestation. There were no statistically significant differences noted between the fetuses of the different groups. It is concluded by the petitioner that orally administered pantethine has no adverse effects on the development of rabbit fetuses at doses up to 120 mg/kg bw/day, the highest dose tested.

The effects of intramuscular injections of pantethine to pregnant ICR-JCL mice and Wistar rats during the organogenesis period of their fetuses were investigated. Pantethine was injected intramuscularly as an aqueous solution into pregnant mutiparous ICR-JCL mice and Wistar rats. The doses in mice were 0, 20, and 1500 mg/kg bw/day from day 7 to day 12 of gestation. In rats the doses were 0, 20, and 500 mg/kg bw/day from day 9 to day 14 of gestation.

The average number of implants, live fetuses and surviving weanlings were within normal limits. No statistically significant adverse effects were observed in the average litter size and stillborn fetuses in treated groups as compared to control. The mean body weight at birth of both treated groups was slightly but significantly lower in mice; weight gain was also slightly but significantly lower in the group of mice treated with 20 mg/kg bw/day of pantethine.

The mean weight at birth of rats treated with 500 mg/kg bw/day of pantethine was slightly but significantly lower than in control rats: weight gain was also slightly but significantly lower in both treated groups. A small number of isolated abnormalities were seen in all groups in both species with comparable incidences. Female mice treated with 1500 mg/kg bw/day of pantethine delivered fetuses with a higher number of incomplete formation of a 13$^{th}$ rib and non-ossified calcanei. However, mice treated with 20 mg/kg bw/day delivered fetuses with a higher number of ossified caudal vertebrae. Female rats treated with 500 mg/kg bw/day of pantethine delivered fetuses with a higher number of non-ossified calcanei vertebrae. It is concluded by the petitioner that the intramuscular administration of pantethine at dosages of up to 1500 mg/kg bw/day in pregnant mice and 500 mg/kg bw/day in pregnant rats resulted in slightly decreased body weight of pups at birth and at weanling, but no adverse effects on mice or rat fetal development were observed at these dosages or at a lower dose of 20 mg/kg bw/day.

### 3.2.4 Genotoxicity

Pantethine (PTSS lot no. 2591) was tested in a modified Ames mutagenicity assay. The petitioner reports that pantethine showed no mutagenicity in strains TAI535, TA537, TAI538, TA98, TA100 of *Salmonella typhimurium* and in strain Wos uvrA of *Escherichia coli* either with or without metabolic activation.

The mutagenic potential of pantethine (PTSS lot no. 2591) was tested in the chromosomal aberration test using Chinese hamster cells. Based on the outcomes the petitioner concludes that pantethine is not mutagenic in the chromosome aberration test with mammalian cells in culture.

The mutagenic potential of pantethine (PTSS lot no. 2591) was also tested by the rec-assay using recombination repair deficient *Bacillus subtilis* M45(rec-) and wild type H17($rec^+$). The pantethine concentrations used were 10,100, 1000 and 10000 µg/ml. Pantethine showed no growth inhibition with either strain of *B. subtilis* M45 or H17 at a concentration of up to 10000 µg/ml. It is therefore concluded by the petitioner that pantethine caused no primary damage to DNA under the protocol conditions.

### 3.2.5 Long term carcinogenicity study

The petitioner reports that pantethine was given in drinking water at levels of 0, 0.8 and 3.2 g/l and administered to three groups each of 55 males and 55 females Fisher 344 rats for 104 weeks to evaluate its carcinogenicity. The average pantethine consumption calculated from the water consumption was 32.8 mg/kg bw/day for males and 44.9 mg/kg bw/day for females in the 0.8 g/l group, and 124.2 mg/kg bw/day for males and 175.1 mg/kg bw/day for females in the 3.2 g/l group.

Clinical signs, mortality, body weights and water consumption were comparable between control and pantethine treated rats. No statistically significant differences in various types of neoplastic and proliferative lesions were seen between the control and treated groups.

It is concluded by the petitioner that under the conditions of this study, pantethine is not carcinogenic in rats.

### 3.2.6 Human studies

A number of clinical trials with pantethine have been reported in the literature and the review of McRae (2005) presents an overview. The purpose of this review was to investigate the effectiveness and tolerability of pantethine in the treatment of hyperlipoproteinemia. Twenty-eight clinical trials were identified providing a pooled population of 646 hyperlipidemic subjects. The average study trial length was 12.7 weeks with pantethine dosages of 600-1200 mg/day. The parameters studied were the mean percent decrease from baseline for total serum cholesterol, low-density lipoprotein cholesterol and serum triacylglycerols and the mean percent increase from baseline for high-density lipoprotein cholesterol. The mean percent decrease from baseline for total serum cholesterol across months 1 through 4 was 8.7%, 11.6%, 12.6% and 15.1%. The mean percent decrease from baseline for low-density lipoprotein cholesterol across months 1 through 4 was 10.4%, 15.2 %, 17.7%, and 20.1%. The mean percentage decrease from baseline for serum triacylglycerols across months 1 through 4 was 14.2%, 15.8%, 23.7% and 32.9%. The mean percent increase from baseline for high-density lipoprotein cholesterol across months 1 through 4 was 6.1%, 7.8%, 10.7% and 8.4%.

It was concluded by the author that pantethine offers an effective therapeutic option in treating patient populations with total serum cholesterol levels greater than 200 mg/dL and/or serum triacylglycerol levels greater than 150 mg/dL.

The review also presents information about adverse effects during the clinical trial experiments McRae (2005) concluded that over the long term, there does not appear to be any severe adverse reactions associated with pantethine administration, even in patients who have been receiving treatment for more than 1 year. However, it was also indicated that very mild adverse reactions have been reported, such as heartburn, mild pruritis, and diarrhea. Although the percentage of subjects experiencing adverse effects was 3.6%, two thirds of these were observed in one single study. It is not evident why 12 of the 35 subjects in this single study experienced such a diverse array of adverse reactions. It is also important to note that the majority of these adverse reactions were resolved, and therefore resulted in only 1 subject of 646 withdrawing from study protocol. The dose range in the experiments was 600-1200 mg/day.

Guillams and Pins (2005) reported that pantethine has been shown in a number of trials to reduce the serum triglycerides. They state: 'pantethine is well tolerated with occasional reports of gastrointestinal discomfort and diarrhea, most frequently when administered in higher doses (i.e. ≥1200 mg/day)'.

## 4. Discussion

The present opinion deals only with the safety and bioavailability of pantethine as a particular source of pantothenic acid intended to be used in food supplements. The safety of pantothenic acid itself, in terms of amounts that may be consumed, is outside the remit of this Panel.

After oral intake pantethine can be metabolized into pantetheine in the intestinal lumen. Pantetheine is, in its turn, absorbed and hydrolyzed in the intestinal mucosa cells into pantothenic acid, which will enter the portal vein. The petitioner indicates that animal experiments favour nearly complete hydrolysis of pantetheine, formed from pantethine by reduction (Wittwer, 1985). Results reported by Tachizawa *et al.* (1979) using jejunum and ileum rat intestinal loops indicated that pantethine is absorbed from the intestine in its unmodified form and as pantothenic acid. Kelly (1997) reports that it is generally accepted that a portion of pantethine is reduced to pantetheine and a portion is further hydrolysed to pantothenic acid prior to intestinal absorption and also that the metabolic fate of an oral dose of pantethine in humans has not been clearly described.

Data in the literature indicate that pantethine is about equally rapidly absorbed as calcium pantothenate when given orally to rats (Ono *et al.,* 1974) with plasma levels being at their maximum values already upon 2 hours post dosing and only slightly (18%) higher in animals dosed with pantethine at a dose that was twice as high as that of calcium pantothenate based on pantothenic acid equivalents. Based on this information the Panel concludes that the bioavailability of pantothenic acid from pantethine upon oral intake might be comparable to or lower than that of pantothenic acid.

Another study, using intramuscular injection, reports that in healthy subjects, pantethine is retained longer in the blood and has more tissue affinity than pantothenic acid (Shigeta *et al.*, 1966).

The toxicity studies reported by the petitioner for pantethine are based on the dossier or on limited unpublished translations of studies published in Japanese journals, performed in the

period 1966-1968 (Oshima *et al.,* 1966; Akimoto *et al.,* 1966; Morita *et al.,* 1968). No original data were available for evaluation.

The petitioner provided results of toxicity studies on pantethine from which it can be concluded that pantethine was not genotoxic, not carcinogenic and that no developmental toxicity was observed in mice and rats up to doses of 600 mg/kg bw/day, and in rabbits up to 120 mg/kg bw/day.

From subchronic toxicity studies in rats and dogs, NOAELs of respectively 36 and 50 mg/kg bw/day were identified.

The petitioner indicates that in Europe food supplement products containing pantethine have been on the market since at least 1984 and used typically up to 10 mg pantethine per day in multivitamin/mineral supplements or vitamin B complex supplements. Also EU food supplements supplying pantothenic acid in the form of calcium pantothenate can be found at levels up to around 500 mg.

In the US, pantethine is on the market in dietary supplements at much higher levels (600-900 mg/day) to reflect the different market requirements and expectations.

The petitioner does not provide actual proposed use levels. The Panel notes that if use levels of pantethine are based on use levels of pantothenic acid or on levels of pantethine used in the US, use levels up to 500 to 900 mg pantethine/day would be foreseen, equivalent to intakes of 8.3 to 15 mg/kg bw/day for a 60 kg person. Daily intakes of 10 mg pantethine/day would amount to 0.167 mg/kg bw/day for a 60 kg person.

Daily intakes up to 8.3 to 15 mg/kg bw/day would result in margins of safety compared to the NOAELs from the animal studies of only 2.4 to 6.0. Daily intakes of 10 mg pantethine/day amounting to 0.167 mg/kg bw/day would give rise to margins of safety of 216 to 300.

Results from clinical studies reveal that pantethine is generally well tolerated although there are occasional reports of heartburn, mild pruritis, gastrointestinal discomfort and diarrhea, most frequently when administered in higher doses (i.e. 350-1200 mg/day and higher (McRae, 2005 and references therein; Wittwer *et al.,* 1985; Gulliams and Pins, 2005).

## CONCLUSIONS

The bioavailability of pantothenic acid from pantethine upon oral intake might be comparable to or lower than that of pantothenic acid.

Given the fact that:

i) the bioavailability studies in rats indicate that part of the pantethine might be absorbed unmodified and that in first instance only part may be converted to pantothenic acid,

ii) the metabolic fate of an oral dose of pantethine in humans has not been clearly described,

iii) the margin of safety between the NOAELs from the animal studies and the exposure resulting from use levels of 500 to 900 mg/day of pantethine is less than 10 and,

iv) results from clinical studies with pantethine indicate that at dose levels between 350-1200 mg/day, occasional cases of heartburn, mild pruritis, gastrointestinal discomfort and diarrhea occur,

the Panel concludes that the safety in use of pantethine as a source for pantothenic acid in food supplements intended for the general population at levels of use of 500 to 900 mg pantethine/day is not demonstrated.

The Panel also concludes that when use levels are 10 mg/day (0.167 mg/kg bw for a 60 kg person), as reported by the petitioner to be used typically in multivitamin/mineral supplements or vitamin B complex supplements in European food supplement products, the margin of safety would amount to 216 to 300. In view of this margin of safety, the Panel concluded that the use of pantethine as a source of panthenoic acid under these conditions would not be of safety concern.

## DOCUMENTATION PROVIDED TO EFSA

1. Dossier on pantethine, application for derogation. July 2005. Submitted by Solgar Vitamin and Herb.

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## GLOSSARY / ABBREVIATIONS

| | |
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| ANS | Scientific Panel on Food additives and nutrient sources added to food |
| EVM | Expert Group on Vitamins and Minerals |
| GMM | Genetically Modified Micro-organisms |
| GRAS | Generally Recognized As Safe |
| JECFA | Joint FAO/WHO Expert Committee on Food Additives |
| NOAEL | No-Observed-Adverse-Effect Level |
| PRI | Population Reference Intake |
| SCF | Scientific Committee on Food |
| SUL | Safe Upper Level |
| UL | Upper Level |