SCIENTIFIC OPINION

Manganese ascorbate, manganese aspartate, manganese bisglycinate and manganese pidolate as sources of manganese added for nutritional purposes to food supplements

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


Adopted on 4 June 2009

PANEL MEMBERS


SUMMARY

Following a request from the European Commission, the Panel on Food Additives and Nutrient Sources added to Food (ANS) was asked to provide a scientific opinion on the safety of manganese ascorbate, manganese aspartate, manganese bisglycinate and manganese pidolate, as sources of manganese, added for nutritional purposes in food supplements, and on the bioavailability of manganese from these sources.

The present opinion deals only with the safety of manganese aspartate, manganese ascorbate, manganese pidolate and manganese bisglycinate, as sources of manganese, intended to be used in food supplements and with the bioavailability of manganese from these sources. The safety of manganese itself, in terms of amounts that may be consumed, is outside the remit of this Panel.

1 For citation purposes: Scientific Opinion of the Panel on Food Additives and Nutrient Sources added to Food on manganese ascorbate, manganese aspartate, manganese bisglycinate and manganese pidolate as sources of manganese added for nutritional purposes to food supplements following a request from the European Commission. The EFSA Journal (2009) 1114, 1-23
No specific studies on bioavailability were made available on any of the manganese sources considered in this opinion. However it can be assumed that manganese sources will dissociate in the stomach and/or in the gastrointestinal fluids into their constituents and that bioavailability in the gastrointestinal tract would be at least similar to that from other dissociable sources of manganese.

The Panel considers that exposure from these sources to aspartate, ascorbate, pidolate, and glycinate at the levels considered in this opinion would be of no safety concern.

Using the highest proposed use level of 5 mg/day of manganese from the manganese sources considered in this opinion, the Panel noted that the total anticipated exposure to manganese from food intake and from food supplements would be 6.4-9.9 mg/day at the mean and 9.8-13.2 mg/day at the 97.5th percentile for adults. For children 3-14 years old, the total anticipated exposure to manganese from food intake and from food supplements would be 6.8-7.2 mg/day at the mean and 8.9-9.2 mg/day at the 97.5th percentile.

The Panel considers that supplemental intake of 4 mg for the general population and 0.5 mg manganese/day for older people, respectively, are unlikely to produce adverse effects. This supplementation would result in total intakes of 12.2 for the general population and 8.7 mg manganese/day for older people, respectively, taking into consideration a level of dietary manganese intake of 8.2 mg/day.

The Panel concludes that the use of manganese aspartate, manganese L-ascorbate, manganese pidolate and manganese bisglycinate, as sources of manganese in food supplements, are not of safety concern provided that the guidance levels for manganese supplementation set by the EVM are not exceeded, 4 mg manganese/day for the general population and 0.5 mg manganese/day for older people.

The Panel concurs with the SCF considerations that exposure to manganese should remain low and should not exceed that found in the diet, taking into consideration that due to its ubiquity, the evidence for manganese deficiency in humans is limited, that manganese is an essential element found naturally in foods and that the margins of exposure between oral manganese levels showing neurotoxic effects and the estimated levels of manganese from the diet remain low.

**Key words:**
Manganese ascorbate, CAS Registry No 16351-10-3, manganese bisglycinate, CAS Registry No 14281-77-7, manganese L-aspartate, manganese pidolate, CAS Registry No 29193-02-0, food supplements.
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BACKGROUND AS PROVIDED BY 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 manganese ascorbate, manganese aspartate, manganese bisglycinate, manganese glycinate and manganese pidolate added for nutritional purposes to food supplements. The relevant Community legislative measure is:


TERMS OF REFERENCE AS PROVIDED BY 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 manganese ascorbate, manganese aspartate, manganese bisglycinate, manganese glycinate and manganese pidolate added for nutritional purposes in food supplements.

ACKNOWLEDGEMENTS


ASSESSMENT

1. Introduction

The present opinion deals only with the safety of manganese ascorbate, manganese aspartate, manganese bisglycinate and manganese pidolate, as sources of manganese, added for nutritional purposes in food supplements, and with the bioavailability of manganese from these sources. The safety of manganese itself, in terms of amounts that may be consumed, is outside the remit of this Panel.

Manganese is a naturally occurring element and an essential nutrient, being a component of several enzyme systems (e.g. glycosyl transferases). Manganese is the twelfth most abundant element and the fifth most abundant metal comprising approximately 0.1 % of the earth’s crust. Manganese exists in both inorganic and organic forms, with manganese dioxide (pyrolusite) being the most abundant naturally-occurring form of manganese. Its ubiquity in soils results in vegetable and animal foods readily containing varying amounts of this element.

2. Technical data

2.1. Chemistry

**Manganese ascorbate**

The molecular formula of manganese ascorbate is Mn(C_{6}H_{7}O_{6})_{2}, its molecular mass is 405.18 g/mol and its CAS Registry Number is 16351-10-3.

![](Figure 1. Structure of manganese ascorbate)

Manganese ascorbate was described, by one petitioner, as a mixture of manganese ascorbate (86%), manganese carbonate (6%) and water (8%). No detailed information was provided by the other petitioner.

**Manganese L-aspartate**

The molecular formula of manganese L-aspartate is Mn(C_{4}H_{8}NO_{4})_{2} and its molecular mass is 319.14 g/mol. No CAS Registry Number was available.

![](Figure 2. Structure of manganese L-aspartate)
The synonyms proposed by the petitioner include the following: manganese di-L-aspartate, manganese di[(S)-2-aminohydrogenobutane-1,4-dioate], and manganese L-aminosuccinate.

**Manganese bisglycinate**

One petitioner presented a dossier on manganese bisglycinate, whereas another petitioner presented a dossier on manganese glycinate, as a synonym of manganese bisglycinate. Therefore only the term manganese bisglycinate is used in the current evaluation.

The molecular formula of manganese bisglycinate is Mn(C$_2$H$_4$NO$_2$)$_2$, its molecular mass is 203.06 g/mol and its CAS Registry Number is 14281-77-7.

![Figure 3. Structure of manganese bisglycinate](image)

The synonyms proposed by the petitioners include the following: glycine manganese salt, manganese glycinate chelate, manganese glycine complex, and manganese diglycinate.

One of the petitioners described this source as a mixture containing manganese bisglycinate at 74%.

**Manganese pidolate**

The molecular formula of manganese pidolate is Mn(C$_5$H$_6$NO$_3$)$_2$, its molecular mass is 311.1 g/mol and its CAS Registry Number is 29193-02-0.

![Figure 4. Structure of manganese pidolate](image)

The synonyms proposed by the petitioner include the following: L-pyroglutamic acid manganese salt, manganese 5-oxoproline, manganese pyroglutame, manganese pyrrolidone carboxylate, and manganese PCA.

2.2. **Specifications**

The petitioners stated the following on the specifications:

**Manganese ascorbate**

Manganese ascorbate was described by one petitioner as light tan to light brown powder, with the mixture of manganese ascorbate being slightly soluble in water (1 g/L at 25 °C). The specifications proposed by the petitioners are summarised in Table 1.
Manganese ascorbate, manganese aspartate, manganese bisglycinate and manganese pidolate as sources of manganese added for nutritional purposes to food supplements

Table 1. **Chemical specifications proposed for manganese ascorbate**

<table>
<thead>
<tr>
<th>Technical dossier</th>
<th>Manganese content</th>
<th>Arsenic</th>
<th>Cadmium</th>
<th>Lead</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>% (w/w)</td>
<td>not more than mg/kg</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2005a</td>
<td>13.7</td>
<td>3</td>
<td>5</td>
<td>1</td>
</tr>
<tr>
<td>2005b</td>
<td>12-14</td>
<td>3</td>
<td>5</td>
<td>5</td>
</tr>
</tbody>
</table>

**Manganese bisglycinate**

Manganese bisglycinate was described as a white to tan powder that is slightly soluble in water, with its purity being not less than 98%. However, the other petitioner described manganese bisglycinate as soluble in water and practically insoluble in ethanol and acetone.

Table 2. **Chemical specifications proposed for manganese bisglycinate**

<table>
<thead>
<tr>
<th>Technical dossier</th>
<th>Assay</th>
<th>Manganese content</th>
<th>Arsenic</th>
<th>Cadmium</th>
<th>Lead</th>
<th>Mercury</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>% (w/w)</td>
<td>not more than mg/kg</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2006a</td>
<td>16</td>
<td>0.5</td>
<td>2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2005c</td>
<td>98%</td>
<td>3</td>
<td>5</td>
<td>1</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Manganese L-aspartate**

Manganese L-aspartate was described as a white to light tan coloured powder, soluble in water. Its purity is not less than 97%.

The reported limits for impurities are as follows: lead not more than 1 mg/kg and heavy metals not more than 10 mg/kg.

**Manganese pidolate**

Manganese pidolate was described as a slightly pink powder, readily soluble in water, specific optical rotation from $-25^0$ to $-22^0$, and its purity not less than 97.6%.

The reported limits for impurities are as follows: glutamic acid < 2%, sulphates < 2%, chlorides < 0.1%, iron < 200 mg/kg, heavy metals < 40 mg/kg, N-5-(oxo-2-pyrrolidinyl)carbonyl glutamic acid < 0.05%, and other secondary reaction products <0.05%.

The Panel notes that according to Commission Regulation (EC) No 629/2008 the maximum levels of lead, mercury and cadmium in food supplements as sold should be 3 mg/kg, 0.1 mg/kg and 1 mg/kg, respectively (EC, 2008).

2.3. **Manufacturing Process**

**Manganese ascorbate**
Manganese ascorbate is synthesised by the reaction of manganese carbonate and ascorbic acid.

**Manganese bisglycinate**

The manufacturing process for manganese bisglycinate was adequately described by one petitioner. The other petitioner stated that manganese bisglycinate is manufactured synthetically from a soluble manganese salt and glycine, and that it is possible that the glycine may have been produced using genetically modified microorganisms (GMMs). The Panel concludes that manganese bisglycinate produced from genetically modified micro-organisms is not part of the present opinion because it would require a separate submission under Regulation (EC) No 1829/2003 (EC, 2003).

**Manganese L-aspartate**

Manganese L-aspartate is synthesised by the reaction of a so-called “manganese base” (no further details have been provided) and L-aspartic acid.

**Manganese pidolate**

The manufacturing process was adequately described.

### 2.4. Methods of analysis in food

**Manganese ascorbate**

The petitioners did not provide a specific method for analysis of manganese ascorbate in foods. One of the petitioners indicated that the content of manganese was analysed by Inductively Coupled Plasma-Optical Emission Spectrometry (ICP-OES) and ascorbic acid by titration.

**Manganese bisglycinate**

One of the petitioners listed Atomic Absorption Spectrometry (AAS) and Inductively Coupled Plasma Atomic Emission Spectrometry (ICP-AES) as instrumental techniques for the determination of the content of manganese after appropriate extraction and preparation of the samples. It has not been described how glycine was determined.

The other petitioner stated that InfraRed spectrophotometry may be used as a ‘fingerprint’ to confirm the identity of manganese glycinate.

**Manganese L-aspartate**

The petitioners listed AAS and ICP as instrumental techniques for the determination of the content of manganese after appropriate extraction and preparation of the samples.
Manganese pidolate
No method of analysis was provided.

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

Manganese ascorbate
It was indicated that degradation of manganese ascorbate is less than 2% and 3% after two and three years of storage, respectively. However, no specific information was provided. One petitioner stated that concomitant intake of manganese with foods rich in phytic acid (unleavened bread, raw beans, seed nuts and grains and soy isolates) or oxalic acid (spinach, sweet potatoes, rhubarb and bens) may reduce the absorption of manganese. In addition, it was stated by this petitioner that calcium, non-heme iron and magnesium supplements may reduce the absorption of manganese and that intake of magnesium-containing antacids and laxatives may decrease the absorption of manganese if taken concomitantly.

Manganese bisglycinate
It was stated by one of the petitioners that there is no evidence to indicate the occurrence of reactions with other food components following fortification of food substances with manganese bisglycinate. It was reported that in a 17-month assay, there was no interaction between manganese bisglycinate and other metal amino acid chelates of copper, zinc, iron and chromium present at levels of 0.45-0.55 mg, 1.37-1.67 mg, 1.8-2.2 mg, and 36-44 μg/per capsule, respectively. It was also reported that a multivitamin preparation (containing vitamins A, C, D, E and a number of the B vitamin series), in the presence of manganese bisglycinate, retained its levels of activity within the specifications of the United States Pharmacopeial (USP) throughout the duration of the study.

The other petitioner stated only that manganese bisglycinate was stable in foods, without further details.

Manganese L-aspartate,
Manganese L-aspartate was described by the petitioners as stable in foods. However, no specific information was provided.

Manganese pidolate
No specific information was provided.

2.6. Case of need
All petitioners proposed the use of manganese sources, considered in this opinion, in the manufacturing of food supplements including tablets, caplets, capsules, chewable tablets, effervescent powders and/or liquids or in all.
2.7. Proposed use levels and exposure

**Manganese L-aspartate**

According to the petitioner, manganese L-aspartate would be added to food supplements to supply up to 2 mg/day of manganese. This supplementation would correspond to approximately 11 mg/day of manganese L-aspartate, based on the reported manganese content, and to 9 mg/day of L-aspartate.

**Manganese L-Ascorbate**

No information was provided by one of the petitioners on the amounts of manganese ascorbate intended to be added to food supplements. The other petitioner proposed a daily intake of 2-5 mg manganese, which would correspond to an intake of approximately 14-35 mg of manganese ascorbate/day and approximately 12-30 mg ascorbic acid/day.

**Manganese bisglycinate**

No information was provided by one of the petitioners on the amounts of manganese bisglycinate intended to be added to food supplements. This petitioner only stated that the daily intake of manganese from manganese bisglycinate would not exceed those levels anticipated through existing manganese supplementation, adding that the levels of addition of manganese would be similar to levels used for other forms of manganese that are already approved for use in the EU.

The other petitioner stated that the amount to be added to food supplements will be determined by individual formulators but is normally the quantity necessary to supply adults with up to 2 mg manganese/day. This supplementation would correspond to approximately 7.4 mg of manganese glycinate/day (based on a theoretical content of manganese of 27%) and to 5.4 mg of glycinate/day.

**Manganese pidolate**

The petitioner on manganese pidolate did not define precisely the amounts that would be added to food supplements. However, he defined a range of 3.4-20.3 mg/day of manganese pidolate as dosages recommended by the manufacturers of food supplements. Therefore in the absence of more precise data, these dosages would be considered in this opinion as use levels. Accordingly, these supplementations would correspond to approximately 0.6-3.8 mg of manganese/day (based on the highest percentage of manganese content reported) and to 2.8-16.5 mg of pidolate/day.

2.8. Information on existing authorisations and evaluations

No information was provided on previous authorisations or evaluations of the manganese sources considered in this opinion.

**Manganese**
Manganese ascorbate, manganese aspartate, manganese bisglycinate and manganese pidolate as sources of manganese added for nutritional purposes to food supplements

The SCF (1993) estimated that 1-10 mg/day was an acceptable range of manganese intake based on intake from food (SCF, 1993). In 1999, the SCF estimated that the use of manganese carbonate, chloride, citrate, gluconate, glycerophosphate and sulphate were acceptable sources of manganese for use in the manufacture of foods for particular nutritional purposes (PARNUTS) (SCF, 1999).

The SCF considered that an upper level of 0.5 mg manganese/L in natural mineral waters appeared to be acceptable for human consumption (SCF, 1996). This amount would be equivalent to 1 mg manganese/day assuming an intake of 2 L of water/day.

In its latest evaluation of manganese, the SCF could not set an Upper Tolerable Intake Level (UL) (SCF, 2000). The SCF considered that limitations in the human and animal data did not allow the identification of a No-Observed-Adverse-Effect-Level (NOAEL) for critical endpoints for manganese, and stressed that the margin, between oral effect levels in humans as well as in animals and the estimated intake from food, was very low (SCF, 2000). It was thus concluded that given the neurotoxicity findings and the potential higher susceptibility of some subgroups in the general population, oral exposure to manganese beyond the levels normally present in food and beverages could represent a risk of adverse health effects without evidence of any health benefit.

Other populations potentially at an increased susceptibility risk to manganese toxicity have been identified, including patients with liver disease and long-term users of anti-psychotic drugs as well as older people and ethanol consumers (EVM, 2003; SCF, 2000).

Like the SCF, the Expert Group on Vitamins and Minerals (EVM) could not establish a Safe Upper Level for manganese (EVM, 2003). The EVM considered for guidance purposes that for the general population supplemental intake of up to 4 mg manganese/day, in addition to manganese from the diet, would be unlikely to produce adverse effects. In older people, the EVM assumed that up to 0.5 mg supplemental manganese/day, in addition to manganese from the diet, would not result in adverse effects. Assuming dietary intakes of 8.2 mg manganese, acceptable total intakes were estimated by the EVM to be 12.2 mg manganese/day in the general population (equivalent to 0.2 mg/kg bw for a 60-kg adult) and to 8.7 mg manganese/day (equivalent to 0.15 mg/kg bw) for older people.

Manganese dichloride, gluconate, sulfate and citrate salts are affirmed as Generally Recognized as Safe (GRAS) in the United States and can be used as an ingredient in certain foods with no limitation other than current good manufacturing practices and in nutrient supplements (Code of Federal Regulations, 2006).

The World Health Organization (WHO) considered that 2-3 mg of manganese/day was adequate for adults and that consumption of up to 8–9 mg/day would be safe (WHO, 1973).

The US Environmental Protection Agency (EPA) has established an oral Reference Dose (RfD) for manganese of 0.14 mg/kg bw/day (EPA, 1996). This would be equivalent to 8.4 mg/day for a 60 kg individual.

The Food Nutrition Board (FNB) of the US and the Institute of Medicine (IOM) have set adequate intake levels for manganese in humans (Table 3).
Table 3. Adequate intake levels for manganese derived by ATSDR (ATSDR, 2008)

<table>
<thead>
<tr>
<th>Population</th>
<th>Age</th>
<th>Males (mg/day)</th>
<th>Females (mg/day)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Infants</td>
<td>0-6 months</td>
<td>0.003</td>
<td>0.003</td>
</tr>
<tr>
<td>Infants</td>
<td>7-12 months</td>
<td>0.6</td>
<td>0.6</td>
</tr>
<tr>
<td>Children</td>
<td>1-3 years old</td>
<td>1.2</td>
<td>1.2</td>
</tr>
<tr>
<td>Children</td>
<td>4-8 years old</td>
<td>1.5</td>
<td>1.5</td>
</tr>
<tr>
<td>Children</td>
<td>9-13 years old</td>
<td>1.9</td>
<td>1.6</td>
</tr>
<tr>
<td>Adolescents</td>
<td>14-18 years old</td>
<td>2.2</td>
<td>1.6</td>
</tr>
<tr>
<td>Adults</td>
<td>&gt; 19 years old</td>
<td>2.3</td>
<td>1.8</td>
</tr>
<tr>
<td>Pregnancy</td>
<td></td>
<td>-</td>
<td>2.0</td>
</tr>
<tr>
<td>Lactation</td>
<td></td>
<td>-</td>
<td>2.6</td>
</tr>
</tbody>
</table>

The Agency for Toxic Substances and Disease Registry of the United States (ATSDR) proposed recently an interim guidance value for oral exposure to inorganic manganese of 0.16 mg manganese/kg bw/day, based on FNB’s UL of 11 mg manganese/day (ATSDR, 2008).

In the European Community some manganese salts are authorised sources of minerals that may be added for specific nutritional purposes in foods for particular nutritional uses (EC, 2001) and in foods for special medical purposes (EC, 2006).

**Aspartate**

L-aspartic acid is an acceptable amino acid source for use in foods for particular nutritional uses in the same foods as described before (EC, 2001).

The ANS Panel identified a NOAEL for aspartate of 700 mg/kg bw/day from a 90-day rat study based on renal toxicity findings in males (EFSA, 2008a).

The SCF and the FNB have stressed that caution should be warranted when using any single amino acid at levels significantly above those normally found in food, since it could lead to nutritional imbalances (SCF, 1990; IOM, 2005).

There are neither upper tolerable intake level nor required intakes set for aspartic acid (IOM, 2005; WHO, 2002). In 2002, the IOM noted that supplementation of up to 8 g/day of aspartic acid, in addition to approximately 3 g/day from food, did not result in any documented adverse effects (IOM, 2002).

The WHO expert consultation on protein and amino acid requirements in human nutrition considered that for dispensable amino acids (which include aspartic acid) 0.48 g/kg bw/day should be sufficient to maintain body nitrogen homeostasis in healthy adults (WHO, 2002). For a 60 kg individual, this amount would be equivalent to 29 g/day.

**Ascorbate**

The Scientific Panel on Dietetic Products, Nutrition and Allergies (NDA) could not establish a tolerable upper intake level for vitamin C. However, the NDA Panel noted that supplemental daily doses of vitamin C up to about 1 g/person/day, in addition to normal dietary intakes, are not associated with adverse gastrointestinal effects, but that acute gastrointestinal effects may occur at higher intakes (3-4 g/person/day) (EFSA, 2004).
The ANS Panel considered that an additional exposure to ascorbate, in the form of food supplements up to a level of 1232 mg vitamin C/day, is of no safety concern (EFSA, 2009).

**Bisglycinate**

The Scientific Panel on Food Additives, Flavourings, Processing Aids and Materials in Contact with Food (AFC) considered that the similarity in chemical structure between metal glycimates and ferrous bisglycinate anticipated that the glycine part of glycimates will exhibit similar toxicological characteristics as their ferrous bisglycinate counterpart. The Panel agreed that the subchronic studies on ferrous bisglycinate could be used to assess the subchronic toxicity of the glycimates. From these studies a NOAEL of 500 mg/kg bw/day for ferrous bisglycinate in rats (the highest dose tested) was derived corresponding to approximately 400 mg glycinate/kg bw/day (EFSA, 2008b).

Glycine (synthetic or natural) is already permitted in the EU for use in foods, under Directive 2001/15/EC on substances that may be added for specific nutritional purposes in foods for particular nutritional uses (EC, 2001). Glycinate (E 640) and its sodium salt have a not-specified ADI (SCF, 1991) and are permitted as food additives in the EU, under Directive 95/2/EC on food additives other than colours and sweeteners (EC, 1995).

**Pidolate**

The AFC Panel concluded that consumption of up to 3 g/day of L-pidolic acid in food supplements was of no safety concern, based on the natural occurrence of L-pidolic acid in foods and its endogenous formation (EFSA, 2007).

### 2.9. Available information on dietary exposure to manganese, aspartate, ascorbate, glycine and pidolate

**Manganese**

The concentrations of manganese in foodstuffs vary considerably, but most are below 5 mg/kg such as in grain, rice and nuts. However, manganese levels exceeding 10 or 30 mg/kg have been reported in some cases (SCF, 2000).

In adults, the daily average manganese intake in European adults is on average between 1.4 and 4.9 mg/person/day, and the 97.5th percentile of manganese intake varies from 4.8 to 8.2 mg/person/day (EVM, 2003; Turconi et al., 2009; Leblanc et al., 2005). For children, the daily average manganese intake varies from 1.8 to 2.2 mg/person/day, and the 97.5th percentile from 3.3 to 4.2 mg/person/day (AFSSA, 2009; EVM, 2003; Turconi et al., 2009).

Using the highest proposed use level of 5 mg/day of manganese from the manganese sources considered in this opinion the Panel noted that the total anticipated exposure to manganese from food intake and from food supplements would be 6.4-9.9 mg/day at the mean and 9.8 - 13.2 mg/day at the 97.5th percentile for adults. For children 3-14 years old, the total anticipated exposure to manganese from food intake and from food supplements would be 6.8 - 7.2 mg/day at the mean and 8.9-9.2 mg/day at the 97.5th percentile (see Table 4).
Manganese ascorbate, manganese aspartate, manganese bisglycinate and manganese pidolate as sources of manganese added for nutritional purposes to food supplements

Table 4. **Summary information on manganese intake and anticipated exposure to manganese from manganese sources considered in this opinion**

<table>
<thead>
<tr>
<th>Manganese</th>
<th>Average Intake range (mg/day)</th>
<th>High intake (mg/day)</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intake range from food in Europe for adults</td>
<td>1.4 – 4.9</td>
<td>4.8 – 8.2</td>
<td>Leblanc <em>et al.</em>, 2005; EVM, 2003; Turconi <em>et al.</em>, 2009</td>
</tr>
<tr>
<td>Intake range from food in Europe for children (3-17 years old)</td>
<td>1.8 – 2.2</td>
<td>3.3 – 4.2</td>
<td>AFSSA, 2009; EVM, 2003; Turconi <em>et al.</em>, 2009</td>
</tr>
<tr>
<td>Highest amount (range of proposed supplementation) of manganese supplementation as indicated by one petitioner</td>
<td>5 (0.6-5)</td>
<td></td>
<td>Technical dossier</td>
</tr>
<tr>
<td>Total anticipated exposure to manganese from food supplements and food intake$^1$ for adults</td>
<td>6.4 – 9.9</td>
<td>9.8-13.2</td>
<td>Calculation by the Panel</td>
</tr>
<tr>
<td>Total anticipated exposure to manganese from food supplements and food intake$^2$ for children (3-17 years old)</td>
<td>6.8 – 7.2</td>
<td>8.9-9.2</td>
<td>Calculation by the Panel</td>
</tr>
</tbody>
</table>

$^1$calculation based on highest proposed use level of 5 mg/day plus average dietary intake of 1.4-4.9 mg/day and high dietary intake of 4.8-8.2 mg/day for adults

$^2$calculation based on highest proposed use level of 5 mg/day plus average dietary intake of 1.8–2.2 mg/day and high dietary intake of 3.3–4.2 mg/day for children

**Aspartate**

The mean intake of aspartic acid has been reported to range from 4.1 g/day (children 1-3 years old) to 9.3 g/day (males 19-30 years old). At the 95th percentile, aspartic acid intakes ranged from 6.6 g/day (children 4-8 years old) to 12.9 g/day (males 19-50 years old) (EFSA, 2008a).

**Glycine**

The normal (mean) intake of glycine in proteins, from both food of animal origin and vegetable origin, was calculated to be about 26 mg/kg bw/day for adults (>15 years old) and to about 43 mg/kg bw/day for children (< 15 years old) (EFSA, 2008b).

**Pidolate**

L-pidolate acid occurs naturally in numerous plants and the highest reported concentrations found in products were 0.61 g/L in wine and 1.55 g/L in tomato juice (EFSA, 2007). Estimations of exposure from these sources were not available.
3. Biological and toxicological data

3.1. Bioavailability and biological data

No specific studies on bioavailability were made available on any of the manganese sources considered in this opinion. However, it can be assumed that these sources will dissociate in the stomach and/or in the gastrointestinal fluids into their constituents, and that bioavailability of manganese from these sources would be at least similar to that from other dissociable sources of manganese.

The absorption and metabolic fate of manganese have been thoroughly described previously (SCF, 2000; EFSA, 2006; EVM, 2003; ATSDR, 2008). In summary, results from animal studies indicate that the gastrointestinal absorption of manganese is rapid and it has been reported to take place in the small intestine via a carrier-mediated mechanism. The amount of manganese absorbed across the gastrointestinal tract in human adults is reported to be variable, typically averaging at about 3–8%. However, this absorption appears to be higher in young infants, without specific absorption ranges being proposed. The relative manganese absorption for different manganese compounds is unknown. The retention of manganese from food was 5% at day 10, and from water was 2.9% at day 10. Once absorbed, manganese enters the systemic circulation and accumulates in the liver, kidney, pancreas and the brain. In humans, manganese absorption tends to be greater from manganese chloride (in demineralized water) than from foods labelled intrinsically or extrinsically with $^{54}$Mn. However, the biological half-life of manganese from either manganese chloride or food is the same. Manganese is excreted essentially in the faeces showing biphasic elimination in humans, with half-lives of 13 and 34 days. Iron, calcium, phosphorus and magnesium as well as other food components (e.g. phytate, fibres) are reported to interfere with manganese absorption.

3.2. Toxicological data

No specific toxicity data were made available on most of the sources concerned by this opinion. Applications refer mainly to manganese toxicity and its neurotoxic effects upon inhalation. The Panel noted the lack of toxicological and absorption, distribution, metabolism, and excretion (ADME) data on the sources concerned by this opinion, but given the essentiality of manganese and its ubiquity in foods as well as the large number of manganese compounds already permitted in food, the Panel considered that evaluation of these sources can be based on toxicity data for manganese.

3.2.1. Manganese pidolate

3.2.1.1. Acute toxicity

The median lethal dose (LD$_{50}$) of manganese pidolate in a group of 10 fasting Sprague-Dawley rats was determined to be $>2000$ mg/kg bw after a single dose administration in aqueous solution according to the OECD Test Guideline No. 401.
3.2.1.2. Genotoxicity

A reverse mutation assay carried out according to OECD Test Guideline 471 in Salmonella typhimurium strains TA98 and TA100 reported no mutagenic effect for manganese pidolate with and without metabolic activation at dose levels of 648, 1080, 1800, 3000 and 5000 µg/plate.

3.2.2. Manganese toxicity

The weight of evidence from a number of studies in rodents and primates support the notion that neurobehavioral end-points are the most relevant for evaluation of manganese toxicity from inorganic sources. Analysis of manganese feeding studies in rats [in most cases as manganese sulphate (MnSO₄) or manganese chloride (MnCl₂)], carried out during different exposure lengths (13 weeks to 8 months), showed neurochemical alterations in the brains and neuromotor and behavioural changes in exposed animals, as summarized below. In humans, examination of available data from epidemiology studies or intoxication reports, suggest that neurological effects can occur in children and adults upon ingestion of manganese by the oral route (SCF, 2000; ATSDR, 2008). A characteristic syndrome known as “manganism” has been reported among workers chronically exposed to manganese by inhalation, and is characterised by irreversible effects such as weakness, anorexia, muscle pain, slow speech, emotionless “mask-like” facial expression and a slow movement of limbs.

In summary, the Lowest Adverse Effect Level (LOAEL) identified by the SCF in 2000 was 0.28 mg manganese/kg bw/day in a rat study (SCF, 2000). Oral administration of 25 mg MnCl₂·4H₂O (6.9 mg/kg bw/day) for 18 months to four male rhesus monkeys was reported to induce muscular weakness, rigidity of lower limbs and degeneration of neurons in one brain region as well as testicular changes. In four rhesus monkeys muscular weakness, rigidity and other effects were observed at doses of approximately 7 mg manganese/kg bw/day. In human cohort studies assessing manganese exposure from water in relation to neurological effects (impaired learning, decrease of muscle tone, mental disturbances, etc), doses down to 0.48 mg manganese/day have been related to some of these effects (SCF, 2000). However, given the paucity of the data and uncertainty of the contribution of food to manganese exposure from food it has not been possible to establish ULs for manganese.

According to the ATSDR evaluation (2008), daily oral intakes of 328 mg manganese/kg bw/day during neonatal periods caused subtle neurobehavioral changes in primates (but not 107.5 mg manganese/kg bw/day). In rodents, exposure to doses ≥ 10 mg manganese/kg bw/day can cause the same subtle changes. The LOAELs for manganese, reported as being associated with severe neurobehavioral effects in adult rats following a 30-day exposure, were 5.6 mg /kg bw/day with decreased open-field locomotor activity and impaired cognitive performance in male rats, 6.5 mg /kg bw/day following a 10-week exposure by gavage and 11 mg /kg bw/day for increased pulse-initiated acoustic startle response in rats exposed orally on postnatal days 1–21. Based on the NOAEL of 107 mg manganese/kg bw/day established in monkeys, the ATSDR estimated that a value of 1 mg manganese/kg bw/day could be derived (applying a 100 uncertainty factor). However, this monkey-based value would be about 6-fold higher than the FNB’s UL value of 11 mg/day for adults, and the ASTDR considered the FNB’s value as adequate to derive an Interim Guidance Value for oral exposure to inorganic manganese of 0.16 mg manganese/kg bw/day for a 70-kg individual (ATSDR, 2008).
According to the EVM evaluation, in laboratory animals adverse neurological effects have been reported following long term exposure to manganese at levels higher than 50–200 mg manganese/kg bw/day. Detailed neurological examinations performed in a mice study showed effects at doses ≥ 130 mg manganese/kg bw/day. In humans, results from two retrospective studies showed conflicting neurotoxic effects in cohorts exposed to water containing manganese at levels of up to 1.8–2.3 mg manganese/L (positive results, > 10 years exposure duration) and 0.3–2.1 mg manganese/L (negative results, 10–40 years exposure duration). Assuming a water consumption of 2 L/day allowed the EVM to estimate manganese intakes in these studies to be approximately 3.6–4.6 and 0.6–4.4 mg manganese/day, respectively. Based on the results from the retrospective study showing negative results, the EVM estimated that it was reasonable to assume that in the general population, a supplemental intake of up to 4 mg manganese/day in addition to the diet would be unlikely to produce adverse effects. Based on the findings from the retrospective study showing positive results, the EVM estimated that it could be assumed that up to 0.5 mg manganese/day, in addition to the diet, would not result in adverse effects in older people (EVM, 2003).

4. Discussion

Exposure calculations to the sources (anions) based on proposed use levels show that for aspartate the exposure would be 9 mg/day, corresponding to 0.15 mg/kg bw/day for a 60-kg individual. This amount of aspartate would be more than 4000 times lower than the NOAEL for aspartate identified in a previous evaluation (700 mg/kg bw/day) and would thus be of no safety concern. For ascorbate the exposure would be up to 30 mg/day. This amount of ascorbate would be acceptable considering that supplementation with up to 1232 mg of ascorbic acid/day has been considered previously as of no safety concern. For bisglycinate, taking into consideration the only use level proposed by the petitioners, the exposure to glycinate would be 5.4 mg/day corresponding to 0.09 mg/kg bw/day. This amount would be of no safety concern remaining between 200 to 400 times less than the estimated glycine content in the diet for adults and children, respectively. For pidolate, the calculated exposure based on available use levels would be between 3 and 16 mg/day. These amounts of pidolate would be acceptable considering that supplementation with up to 3000 mg/day of L-pidolic acid has been considered previously as of no safety concern.

The Panel considered that the bioavailability of manganese from these sources would be at least similar to that from other dissociable sources of manganese in the gastrointestinal tract. Overall, manganese bioavailability has been estimated to range between 3–8% in adults, whereas this absorption appears to be higher in young infants.

Manganese dietary intake from food in Europe has been reported to be between 1.4 and 4.9 mg manganese/day on average and between 4.8 and 8.2 mg manganese/day at the high percentile intake for adults. For children, the average dietary intake from food was reported to be between 1.8 and 2.2 mg manganese/day and between 3.3 and 4.2 for the high percentile intake. The total anticipated exposure to manganese from the diet, and from the highest manganese supplementation level considered in this opinion, was estimated by the Panel to be on average between 6.4–9.9 and 6.8–7.2 mg manganese/day for adults and children, respectively, assuming a supplementation of 5 mg manganese/day. The highest percentile intakes estimated for adults and children would be between 9.8-13.2 and 8.9-9.2 mg manganese/day, respectively.
The Panel concurs with the SCF considerations that exposure to manganese should remain low and should not exceed that found in the diet, taking into consideration that due to its ubiquity, the evidence for manganese deficiency in humans is limited, that manganese is an essential element found naturally in foods and that the margins of exposure between oral manganese levels showing neurotoxic effects and the estimated levels of manganese from the diet remain low.

The Panel estimates that the EVM considerations on supplemental intake of manganese in the general population and older people can be used as the basis for evaluating the safety of manganese sources considered in this opinion. The Panel considers in agreement with the EVM, that the supplemental intakes of 4 mg manganese/day for the general population and 0.5 mg manganese/day for older people, respectively, are unlikely to produce adverse effects. This supplementation would result in total intakes of 12.2 manganese/day for the general population and 8.7 mg manganese/day for older people, respectively, taking into consideration a level of dietary manganese intake of 8.2 mg/day.

The Panel notes that use levels proposed by the petitioners are not all in line with these guidance levels for manganese supplementation.

CONCLUSIONS

The present opinion deals only with the safety of manganese ascorbate, manganese aspartate, manganese bisglycinate and manganese pidolate, as sources of manganese, added for nutritional purposes in food supplements and with the bioavailability of manganese from these sources. The safety of manganese itself, in terms of amounts that may be consumed, is outside the remit of this Panel.

It can be assumed that manganese sources will dissociate in the stomach and/or in the gastrointestinal fluids into their constituents and that bioavailability would be at least similar to that from other dissociable sources of manganese.

The Panel estimates that exposure from these sources to aspartate, ascorbate, pidolate and glycinate at the levels considered in this opinion would be of no safety concern.

The Panel considers that supplemental intakes of 4 mg manganese/day for the general population and 0.5 mg manganese/day for older people, respectively, are unlikely to produce adverse effects. This supplementation would result in total intakes of 12.2 manganese/day in the general population and 8.7 mg manganese/day for older people, respectively taking into consideration a level of dietary manganese intake of 8.2 mg/day.

The Panel concludes that the use of manganese aspartate, manganese L-ascorbate, manganese pidolate and manganese bisglycinate as sources of manganese in food supplements are not of safety concern provided that the guidance levels for manganese supplementation set by the EVM are not exceeded, 4 mg manganese/day for the general population and 0.5 mg manganese/day for older people.

The Panel concurs with the SCF considerations that exposure to manganese should remain low and should not exceed that found in the diet, taking into consideration that due to its ubiquity, the evidence for manganese deficiency in humans is limited, that manganese is an essential element found naturally in foods and that the margins of exposure between oral manganese levels showing neurotoxic effects and the estimated levels of manganese from the diet remain low.
Manganese ascorbate, manganese aspartate, manganese bisglycinate and manganese pidolate as sources of manganese added for nutritional purposes to food supplements

**DOCUMENTATION PROVIDED TO EFSA**


**REFERENCES**


The EFSA Journal (2009) 1114, 19-23
Manganese ascorbate, manganese aspartate, manganese bisglycinate and manganese pidolate as sources of manganese added for nutritional purposes to food supplements


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### GLOSSARY / ABBREVIATIONS

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Description</th>
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<tbody>
<tr>
<td>AAS</td>
<td>Atomic Absorption Spectrometry</td>
</tr>
<tr>
<td>ADME</td>
<td>Absorption, Distribution, Metabolism, and Excretion</td>
</tr>
<tr>
<td>AFC</td>
<td>Scientific Panel on Food Additives, Flavourings, Processing Aids and Materials in Contact with Food</td>
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<tr>
<td>ANS</td>
<td>Scientific Panel on Food Additives and Nutrient Sources added to Food</td>
</tr>
<tr>
<td>ATSDR</td>
<td>Agency for Toxic Substances and Disease Registry</td>
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<tr>
<td>CAS</td>
<td>Chemical Abstracts Service</td>
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<tr>
<td>EC</td>
<td>European Commission</td>
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<tr>
<td>OECD</td>
<td>Organisation for Economic Co-operation and Development</td>
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<tr>
<td>EFSA</td>
<td>European Food Safety Authority</td>
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<tr>
<td>EPA</td>
<td>Environmental Protection Agency</td>
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<tr>
<td>EVM</td>
<td>Expert Group on Vitamins and Minerals</td>
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<tr>
<td>FNB</td>
<td>Food Nutrition Board</td>
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<tr>
<td>GMMs</td>
<td>Genetically Modified Microorganisms</td>
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<tr>
<td>GRAS</td>
<td>Generally Recognized as Safe</td>
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<tr>
<td>ICP</td>
<td>Inductively Coupled Plasma</td>
</tr>
<tr>
<td>ICP-AES</td>
<td>Inductively Coupled Plasma-Atomic Emission Spectrometry</td>
</tr>
<tr>
<td>ICP-OES</td>
<td>Inductively Coupled Plasma-Optical Emission Spectrometry</td>
</tr>
<tr>
<td>IOM</td>
<td>Institute of Medicine</td>
</tr>
<tr>
<td>LOAEL</td>
<td>Lowest Observed Adverse Effect Level</td>
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<tr>
<td>NDA</td>
<td>Scientific Panel on Dietetic products, nutrition and allergies</td>
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<tr>
<td>NOAEL</td>
<td>No Observed Adverse Effect Level</td>
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<tr>
<td>PARNUTS</td>
<td>Foods for particular nutritional uses</td>
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<tr>
<td>RfD</td>
<td>Reference Dose</td>
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<td>SCF</td>
<td>Scientific Committee on Food</td>
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<tr>
<td>UL</td>
<td>Upper Tolerable Intake Level</td>
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<tr>
<td>USP</td>
<td>United States Pharmacopeial</td>
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<td>WHO</td>
<td>World Health Organization</td>
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