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FERROUS GLYCINATE (PROCESSED WITH CITRIC ACID)

Chemical and Technical Assessment (CTA)

First draft prepared by Inge Meyland

1 Summary

This substance has not previously been evaluated by JECFA. Ferrous glycinate is an iron (II) chelate of the amino acid glycine. It is manufactured by reaction of reduced iron with glycine in presence of citric acid. At chemical equilibrium over 97% of the ferrous ions are chelated. The resulting product is spray dried without previous removal of the citric acid to obtain a powder. The final product contains as the only significant impurities citric acid and ferric ions. The substance is highly hydroscopic and may contain water in variable amounts.

The intended use of ferrous glycinate is as a nutrient both as a direct food fortificant and as a food supplement. Overall results of various studies indicate ferrous glycinate to be well suited for use as an iron fortificant and beneficial in the treatment of iron deficient anaemia at lower doses than those associated with ferrous sulphate preparations. The stability of ferrous glycinate both what regards pH and temperature is acceptable for the intended food uses. No significant reaction with other nutrients in food has been reported. The most relevant reaction of ferrous glycinate with food to take note of is the lipid peroxidation, expected to occur in response to iron oxidation in instances of disintegration of the chelate. Compared to the control unfortified foods milk and milk products fortified with ferrous glycinate did not show significant differences in lipid peroxidation. In contrast the use of ferrous glycinate for fortification of maize and maize products lowered the sensory quality and storage compared to fortification with ferrous sulphate, ferrous triglycinate and sodium iron EDTA, ferrous glycinate of maize. In oily bases, specifically in margarine, ferrous glycinate was demonstrated to withstand process of oxidation for extended periods of time as compared to margarine fortified with typical iron salts.

2 Description

Ferrous glycinate [(process with citric acid)] is an iron (II) amino acid chelate. It contains also significant amounts of citric acid resulting from the manufacturing process. The substance is highly hydroscopic and may contain water in variable amounts.

Synonyms for ferrous glycinate include ferrous bisglycinate and bis-glycino iron (II). The chemical name for ferrous glycinate is iron, bis(glycinato-N,O). The Chemical Abstract Service (C.A.S.) number is 20150-34-9.

Ferrous glycinate is a grey-green, free-flowing, fine powder. It is soluble in water and practically insoluble in acetone and in ethanol.

Ferrous glycinate has not previously been evaluated by JECFA. It is intended for use in foods and beverages as a nutrient supplement. The formulation is aimed to provide a good bioavailablity allowing for its addition to food products without significant alteration of organoleptic properties.

3 Manufacturing

Ferrous glycine is produced by reaction of reduced iron with a mixture of glycine and citric acid an aqueous solution under controlled process conditions. The resulting product is spray dried without previous

removal of the citric acid to obtain a powder. (US Patent, 1989).See appendix 1 for the flow sheet of the process.

3.1 Raw materials

Reduced iron	FCC quality
Glycine	USP quality
Citric acid	USP quality
Water	Food quality

3.2 Method of manufacture

Reduced iron powder is reacted with a two-fold molar excess of glycine together with citric acid in an aqueous environment under controlled reaction conditions. At chemical equilibrium over 97% of the ferrous ions are chelated. The resulting product is dried to obtain a powder. All raw materials are of pharmacopoeia and/or food quality. Information from certificates of analysis for representative batches of raw materials is given in appendix 2. Ferrous glycinate [(process with citric acid)] is manufactured under current good manufacturing practices.

During the reaction the iron under neutral conditions forms a coordinate covalent complex with the ionised carboxylic acid moiety of glycine whereas under alkaline conditions, a further coordinate covalent bond is formed with the neutral amino group of glycine.

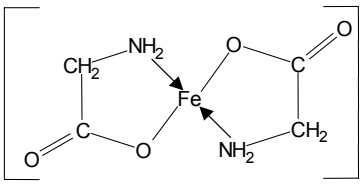
Commercial formulations contain in addition silicon dioxide (0.01%), maltodextrin (2%). These formulations were used in studies for stability and bioavailability.

Since ferrous glycinate is produced from reactions in an aqueous environment with subsequent drying to a powder, slight variations in product colour and particle size may be anticipated. Each manufactured lot is tested by infrared absorption measurements to control that chelation is obtained. A characteristic IR-spectrum of ferrous glycinate with a spectrum for glycine as comparison is shown in appendix 3.

4 Chemical characterization

In ferrous glycinate chemically, the carboxyl group and the α-amino group of glycine both donate electron pairs into the ferrous iron cation forming coordinate covalent bonds (McMurry & Fay, 1995; Atkins & Beran, 1992). Iron serves as the closing member in the formation of the two resulting 5-membered heterocyclic rings.

The chemical formula of anhydrous ferrous glycinate is Fe(COOCH2NH2)2, and its molecular weight is 203.96. The chemical structure is shown below.



Ferrous glycinate [(process with citric acid)] consists predominantly of ferrous glycinate, citric acid and water. The only impurity present in significant is ferric ions. Contents of lead in the final products are low. On the basis of the manufacturing process including raw materials there is no need for criteria for other inorganic impurities in the specifications. See appendix 4 for composition of final products.

5 Functional uses

In the studies referred to below ferrous glycinate has been administered as the commercial formulation of ferrous glycinate [(process with citric acid)] containing maltodextrin and silicon oxide.

Ferrous glycinate may be used as a direct food fortificant or as a food supplement. As a food supplement, it may be formulated into tablets, capsules, solutions or suspensions, or alternatively appear as an ingredient

of other products used for purposes of dietary supplementation. Currently, commercially available foods fortified with ferrous glycinate include milk, fruit juices and fruit flavoured powdered drinks, yoghurt, soft cheese products, cookies, bread, granola and snack bars, all of which are primarily restricted to Latin America with the exception of milk and yoghurt, which also are available to consumers in Italy and Spain. Properties of stability and high bioavailability characterize ferrous glycinate make it suitable as an iron supplement option for use in feeding intervention programs, particularly those involving the fortification of milk with iron. As discussed by Name (1996), primary attributes of ferrous glycinate, with respect to its use as an iron milk fortificant, include no alteration of milk's properties, such as taste and colour, and a lack of oxidative process involving components of the milk upon the addition of ferrous glycinate.

Table 1. Levels of Fe as Ferrous Glycinate in Commercial Food Products

GSFA Food Category	Food Product	Fe Level as Ferrous Glycinate
1.1 Milk and dairy-based drinks	Milk	2.5-3 mg/L
1.6.4 Processed cheese	Soft cheese	2.7 mg/100g
1.7 Dairy-based desserts	Yoghurt	2.7 mg/100g
7.1.1 Breads and rolls	Bread	2.6/100g
7.2.1 Cakes and cookies	Cookies	6 mg/100g
13.6 Food supplements	Vitamin and mineral supplements	8-32 mg/tablet
14.1.2.1 Canned or bottled (pasteurized) fruit juice	Fruit juice	2.5-3 mg/L
14.1.4.3 Concentrates (liquid and solid) for drinks	Fruit flavoured powdered drinks	5.4 mg/25 g powder
15.1 Snacks	Granola and snack bars	1 mg/28g

As of 1994, the Brazilian State of São Paulo has established regulations requiring the fortification of liquid milk with ferrous glycinate, and two years later it issued adjoining requirements for the addition of ferrous glycinate or alternatively ferrous sulphate to powdered milk. The levels of iron from ferrous glycinate were restricted to between 2-3 mg/100 g (São Paulo, 1996).

In a study conducted by Latham et al., (2001), haemoglobin values were designated to monitor the iron status in children and pregnant women receiving orange flavoured powdered drinks fortified with ferrous glycinate in addition to a number of other vitamins and minerals for a period of six months. The powder contained 5.4 mg of iron per 25 g sachet that was re-constituted into 200 ml of water and consumed once per day. Following the study period, haemoglobin levels were reported to be statistically higher in the treatment groups as compared to the baseline haemoglobin levels obtained prior to treatment commencement.

Iost et al. (1998) reported similar results in a study involving normal, anaemic and severely anaemic children (diagnosed based on haemoglobin levels), estimated to consume 2.1 mg of iron/day from milk fortified with 3 mg iron/l derived from ferrous glycinate. Haemoglobin levels were reported to normalize 51% of the number of children with haemoglobin values below normal levels as determined at the onset of the study period.

Levels of haemoglobin were noted to increase significantly from initial values in anaemic study groups receiving iron as either ferrous sulphate or ferrous glycinate as a food supplement. Furthermore, equivalent effects were attained with 23 mg iron/day as ferrous glycinate as compared to doses of 45 mg iron/day as ferrous sulphate (Gualandro & Name, 1996). Administered concurrently with 250 µg folic acid, ferrous glycinate as a food supplement was demonstrated to be effective at doses of 30 mg of iron/day for the treatment of iron deficiency anaemia in adolescents based on haemoglobin values, demonstrating treatment results statistically equivalent to those obtained with 120 mg iron/day from ferrous sulphate in conjunction with folic acid (Pineda et al., 1994). Furthermore, twice as many individuals receiving 120 mg iron/day from ferrous sulfate reported gastric problems as compared to patients treated at the same dose level with

the chelated compound. At dose levels of 30 and 60 mg iron/day from ferrous glycinate adverse gastric effects were reported for 0 and 9.5% of individuals, respectively. Overall results of various studies indicate ferrous glycinate to be well suited for use as an iron fortificant and beneficial in the treatment of iron deficient anaemia at lower doses than those associated with ferrous sulphate preparations.

6 Reactions and Fate in Food

In the studies referred to below ferrous glycinate has been administered as the commercial formulation of ferrous glycinate [(process with citric acid)] containing maltodextrin and silicon oxide.

The solubility and stability of ferrous glycinate is pH-dependent. Ferrous glycinate is unstable at pHs less than 3 or greater than 10 due to weakening of chelate bonds. Studies in stimulated gastric juice indicate that ferrous glycinate is stable in the stomach as a result of the buffering capacity of ferrous glycinate and other food components. Although the chelate ferrous glycinate is heat stable to temperatures higher than 220ºC, ferrous glycinate [(process with citric acid)] due to its content of citric acid is not suited to be used at food processing temperatures for above 153ºC. Most processed foods do not reach internal temperatures of greater than 153ºC. This has been verified for processes including pasteurisation, frying and baking techniques (Name, 1996).

Similarly, there is no evidence to indicate the occurrence of reactions following fortification of food substances with ferrous glycinate [(process with citric acid)]. Product stability and the potential for interaction with food at various temperatures were investigated in a study by the addition of ferrous glycinate to milk, yoghurt and corn flour over a period of 1 month, during which products were tested on Days 3, 10, and 18 (Hendricks & Ashmead, 1995). Lipid peroxidation in food, expected to occur in response to iron oxidation in instances of disintegration of the chelate, as evidenced by the presence of thiobarbituric acid reactive substances (TBARS), was used as an indicator of reactive processes. Ferrous glycinate was added at levels equivalent to 3 mg of elemental iron/litre of raw milk or yoghurt, subsequently packaged in sterile containers and stored in a freezer, a refrigerator, and at room temperature (22ºC). Prior to packaging the fortified milk was subjected to homogenisation and pasteurisation procedures. Alternatively, 30 mg elemental iron as ferrous glycinate was added per kilogram of corn flour, which was subsequently stored under the same conditions. Compared to the control unfortified foods, no significant differences in lipid peroxidation were recorded in the foods at any of the temperature examined. In contrast, both whole maize meal and a porridge made from whole maize meal that was fortified with 30 mg or 60 mg/kg of iron from ferrous glycinate and stored at 30, 40 and 50oC for 20 days resulted in significant production of hexanal, a lipid peroxidation marker. A dose response increase in the production of rancidity (taste panel) paralleled the production of hexanal. Compared to fortification with ferrous sulphate, ferrous triglycinate and sodium iron EDTA, ferrous glycinate lowered the sensory quality and storage of maize (Bovell-Benjamin et al., 1999).

In oily bases, specifically in margarine, ferrous glycinate was demonstrated to withstand process of oxidation for extended periods of time as compared to margarine fortified with typical iron salts (Name, 1996). In particular, as assayed using the RANCIMAT test, which simulates conditions of storage in an accelerated process, administered for 120 minutes equivalent to 6 months of storage, shelf lives of 3 and 6 months before the onset of oxidation were reported for the margarine fortified with ferrous glycinate and the control, respectively. Conversely, margarine fortified with the typical iron salt was subject to immediate oxidation.

Latham et al. (2001) formulated a fruit-flavoured drink powder containing 5.4 mg of iron from ferrous glycinate and several vitamins including vitamin A, C, E, B12, B6, folic acid and riboflavin, and minerals including zinc and iodine. The product, including all nutrients, was stable for up to one year.

Several studies have excluded the possibility of interaction between ferrous glycinate and vitamins. An animal feed supplement of iron amino acid chelates containing ferrous glycinate, added at levels of 5 mg iron/ml to a solution of Vitamin A (20,000 UI/ml), demonstrated no effect on the level of Vitamin A as observed over a period of 324 days. Alternatively, an accelerated degradation of Vitamin A was observed in the presence of 5 mg/ml of ferrous chloride (Marchetti et al., 2000).

Vitamins and minerals were assayed in a 17-month study integrating various metal amino acid chelates together with a multi-vitamin preparation including Vitamins A, D, C and E as well as the Vitamin B series, into a powdered blend, which was subsequently encapsulated for the purposes of the trial (Ashmead and

Ashmead, 1995). Capsules were formulated to contain copper, manganese and zinc as amino acid chelates, at levels of 0.450-0.550, 0.86-1.05 and 1.37-1.67 mg metal/capsule, respectively. Additionally, 36.0-44.0 µg chromium as amino acid chelate, and 1.80-2.20 mg iron present as ferrous glycinate, were incorporated into each capsule. Stability of the chelate products was demonstrated by a lack of interactions between any of the components of the capsules, including the vitamins, which were reported to retain levels within the specifications of the USP for the duration of the study period.

The impact of iron fortification on ascorbic acid stability and availability of iron, intrinsic zinc and calcium in yoghurt and milk was studied by Drago and Valencia (2002) in an in vitro digestion model. Yoghurt was prepared from commercial pasteurised fluid skim milk fortified with either 2.5 mg of iron as ferrous sulfate or ferrous glycinate with or without the addition to L-ascorbic acid at a 4:1 molar ratio of ascorbic acid to iron. Iron from both sources was significantly more available from yoghurt than milk, and was increased equally by the presence of ascorbic acid. Iron fortification had no effect on the availability of intrinsic zinc or calcium. The level of ascorbic acid in iron fortified yoghurt stored at 4o C for up to 14 days decreased by 25-30% with either iron source.

7 References

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