

CARMINES

Prepared at the 55th JECFA (2000) and published in FNP 52 Add 8 (2000), superseding specifications prepared at the 44th JECFA (1995) and published in FNP 52 Add 3 (1995). Metals and arsenic specifications revised at the 59th JECFA (2002). A group ADI of 0-5 mg/kg bw for carmines, as ammonium carmine or the equivalent of Ca, K and Na salts was established at the 26th JECFA (1982) and maintained at the 55th JECFA (2000).

SYNONYMS

Cochineal carmine, Carmine, CI Natural Red 4, CI (1975) No. 75470; INS No. 120

DEFINITION

Obtained by aqueous extraction of cochineal, which consists of the dried bodies of the female insect *Dactylopius coccus* Costa; the colouring principle is a hydrated aluminium chelate of carminic acid in which aluminium and carminic acid are thought to be present in the molar ratio 1:2.

In commercial products the colouring principle is present in association with ammonium, calcium, potassium or sodium cations, singly or in combination, and these cations may also be present in excess. Products may also contain proteinaceous material derived from the source insect, and may also contain free carminate or a small excess of aluminium cations.

Chemical names

Hydrated aluminium chelate of carminic acid (7-beta-D-glucopyranosyl-3,5,6,8-tetrahydroxy-1-methyl-9,10-dioxo-anthracene-2-carboxylic acid)

C.A.S. number

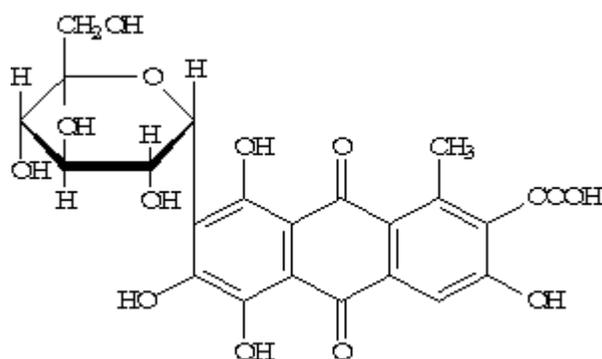
1390-65-4 (carmine)
1260-17-9 (carminic acid)

Chemical formula

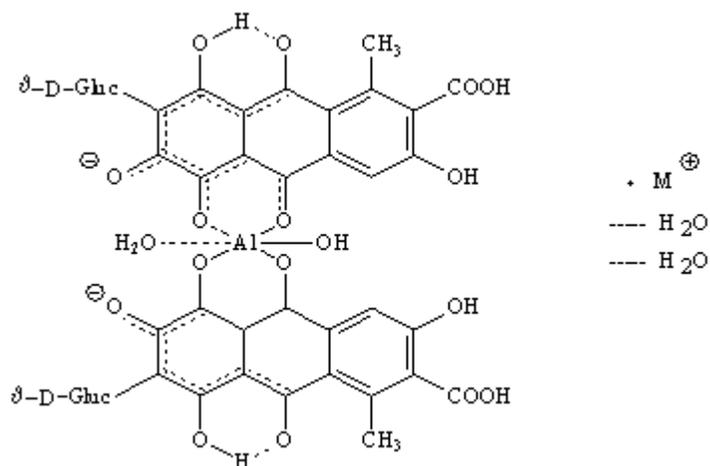
Carminic acid: $C_{22}H_{20}O_{13}$

Structural formula

Carminic acid:



Possible structural formula for the aluminium complex of carminic acid:



M^+ : cation $\frac{1}{2} Ca^{++}$, Na^+ , K^+ , NH_4^+

Formula weight

Carminic acid: 492.39

Assay

Not less than 50% of $C_{22}H_{20}O_{13}$ on the dry basis

DESCRIPTION

Red to dark red, crumbly solid or powder

FUNCTIONAL USES

Colour

CHARACTERISTICS

IDENTIFICATION

Solubility (Vol. 4)

The solubility of carmine preparations varies depending on the nature of the cations present. Products where the major cation is ammonium (ammonium carminate) are freely soluble in water at pH 3.0 and pH 8.5. Products where the major cation is calcium (calcium carminate) are very slightly soluble in water at pH 3.0 but freely soluble at pH 8.5.

Colour reactions

Make a solution of the sample slightly alkaline by adding 1 drop of 10% sodium hydroxide or potassium hydroxide solution. A violet colour is produced.

Add a small sodium dithionite ($Na_2S_2O_4$) crystal to acid, neutral or alkaline solutions of the sample. The solutions are not decolourized.

Dry a small quantity of the sample in a porcelain dish. Cool thoroughly and treat the dry residue with 1 or 2 drops of cold sulfuric acid TS. No colour change occurs.

Acidify a dispersion of the sample in water with 1/3 volume of hydrochloric acid TS and shake it with amyl alcohol. Wash the amyl alcohol solution 2-4 times with an equal volume of water to remove hydrochloric acid. Dilute the amyl alcohol solution with 1-2 volumes of petroleum ether (40-60°) and

shake with a few small portions of water to remove colour. Add, dropwise, 5% uranium acetate, shaking thoroughly after each addition. A characteristic emerald-green colour is produced.

PURITY

Loss on drying (Vol. 4) Not more than 20% (135 °, 3h)

Total ash (Vol. 4)) Not more than 12%
Test 1 g of the sample as directed in the test for Ash (Total Ash)

Protein (Vol. 4) Not more than 25%
Proceed as directed under Nitrogen Determination (non-ammonia N x 6.25)

Matter insoluble in dilute ammonia (Vol. 4) Not more than 1%
Dissolve about 0.25 g of the sample, previously dried and accurately weighed, in 2.5 ml of dilute ammonia solution (160 ml of strong ammonia TS, made up to 500 ml) and dilute to 100 ml with water: the solution is clear. Filter through a sintered glass filter (British Standard Grade No. 3). Wash with a 0.1% ammonia solution and dry to constant weight at 105°.

Lead (Vol. 4) Not more than 5 mg/kg
Determine using an atomic absorption technique appropriate to the specified level. The selection of sample size and method of sample preparation may be based on the principles of the method described in Volume 4, "Instrumental Methods."

Microbiological criteria (Vol. 4) *Salmonella*: Negative per test

METHOD OF ASSAY Weigh accurately about 100 mg of the sample, dissolve in 30 ml of boiling 2N hydrochloric acid and cool. Transfer quantitatively to a 1000-ml volumetric flask, dilute to volume with water, and mix. Determine the absorbance of the solution in a 1 cm cell at the wavelength of maximum absorbance (about 494 nm) using water as the blank. Calculate the percentage of carminic acid in the sample analysis using the formula:

$$\frac{100 \times A \times 100}{1.39 \times W}$$

where

A = absorbance of the sample solution;

W = weight, in mg, of the sample taken; and

1.39 = absorbance of a solution of carminic acid having a concentration of 100 mg per 1000 ml

If the measured absorbance of the solution is not within the range 0.650 to 0.750, prepare another sample and adjust the weight accordingly.