SYNONYMS
Sodium copper chlorophyllin, potassium copper chlorophyllin, C.I. (1975) No. 75810, INS No. 141(ii)

DEFINITION
The alkali salts of Copper Chlorophyllins are obtained by the addition copper to the product obtained by the saponification of a solvent extraction of grass, lucerne, nettle and other plant material; the saponification removes the methyl and cyclophytol ester groups and may partially cleave the pentenyl ring; after addition of copper to the purified chlorophyllins, the acid groups are neutralised to form the salts of potassium and/or sodium; the commercial products may be presented as aqueous solutions or dried powders. Only the following solvents may be used for the extraction: acetone, dichloromethane, methanol, ethanol, propan-2-ol and hexane.

Chemical names
The major colouring principles in their acid forms are
- 3-(10-Carboxylato-4-ethyl-1,3,5,8-tetramethyl-9-oxo-2-vinylphorbin-7-y1)propionate, copper complex (Copper chlorophyllin a)
- 3-(10-carboxylato-4-ethyl-3-formyl-1,5,8-trimethyl-9-oxo-2-vinylphorbin-7-y1)propionato, copper complex (Copper chlorophyllin b)
Depending on the degree of hydrolysis the cyclopentenyl ring may be cleaved with the resultant production of a third carboxyl function.

Chemical formula
Copper chlorophyllin a (acid form): C_{34}H_{32}CuN_{4}O_{5}
Copper chlorophyllin b (acid form): C_{34}H_{30}CuN_{4}O_{6}

Structural formula

where
X = CH₃ for the "a" compound  
X = CHO for the "b" compound  
M = Potassium and/or sodium

**Formula weight**  
Copper chlorophyllin a: 640.20  
Copper chlorophyllin b: 654.18  
Each may be increased by 18 Daltons if the cyclopentenyl ring is cleaved.

**Assay**  
Not less than 95% total copper chlorophyllins after drying (100º, 1 h).

**DESCRIPTION**  
Dark green to blue/black powder or dark green solution.

**FUNCTIONAL USES**  
Colour

**CHARACTERISTICS**

**IDENTIFICATION**

**Solubility (Vol. 4)**  
Soluble in water; very slightly soluble in lower alcohols and ketones and diethyl ether; insoluble in chloroalkanes, hydrocarbons and fixed oils

**Spectrophotometry (Vol. 4)**  
A (1%, 1 cm) of the sample, dried at 100º for 1 h, at 405 nm in pH 7.5 phosphate buffer is not less than 540.

**Test for copper**  
Dissolve the sulfated ash of the sample (using 1 g of sample, Method I) in 10 ml of dilute hydrochloric acid TS by heating on a water bath. Filter if the solution is not clear, and dilute to 10 ml with water. Use this solution as the test preparation for the following tests.

To 5 ml of the test preparation add ammonia TS to make the solution alkaline. Blue colour should appear.

To 5 ml of the above test preparation add 0.5 ml of a 1 in 1000 sodium diethyldithiocarbamate solution. A brown precipitate should be formed.

**Test for sodium (Vol. 4)**  
Passes test  
Test the solution described under TESTS, IDENTIFICATION TESTS Test for copper

**Test for potassium (Vol. 4)**  
Passes test  
Test the solution described under TESTS, IDENTIFICATION TESTS Test for copper

**PURITY**

**Basic dyes**  
To 5 ml of a 0.5% aqueous solution of the sample in a test-tube add 1 ml of 1 N hydrochloric acid and add 5 ml of diethyl ether. Mix thoroughly and allow to separate. The ether layer should be no darker than pale green.

**Residual solvents (Vol. 4)**  
Acetone, methanol, ethanol, propan-2-ol, hexane: Not more than 50 mg/kg, singly or in combination  
Dichloromethane: Not more than 10 mg/kg  
Determine gas chromatographically using either the method of entrainment
distillation (*Determination of Residual Solvents*) or headspace analysis (*Limit Test for Solvent Residues*).

**Free ionizable copper**

Not more than 200 mg/kg

Accurately weigh about 1 g of the sample and dissolve in 20 ml of arachid oil, with the aid of gentle heat. Add exactly 200 ml of water, stir mechanically, and adjust to pH 3.0 by careful addition of 0.5 N hydrochloric acid (avoid overshooting). Allow the mixture to stand for 10 min. If necessary readjust to pH 3.0 by careful addition of 0.5 N hydrochloric acid. Transfer to a separating funnel and allow to stand for about 20 min. Filter the aqueous phase through a No. 50 Whatman filter paper, rejecting the first 10 ml. Subject this solution to analysis for copper by *atomic absorption spectrometry* (see Volume 4).

**Total copper**

Not more than 8% of the total copper phaeophytins

Ignite about 0.1 g, accurately weighed, of the sample contained in a silica dish, at a temperature not exceeding 500º, until all carbon is removed; moisten with one or two drops of concentrated sulphuric acid and re-ash. Dissolve the ash by boiling with 3 portions (each of 5 ml) of 10% (w/w) hydrochloric acid, filtering each addition through the same small filter paper into a 100 ml volumetric flask. Cool, and make up to volume with purified water. Subject this solution to analysis for copper by *atomic absorption spectrometry* (see Volume 4).

**Arsenic (Vol.4)**

Not more than 3 mg/kg (Method II)

**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."

**METHOD OF ASSAY**

Weigh accurately about 1 g of the sample, dried previously at 100º for 1 h, then dissolve in 20 ml Phosphate Buffer Solution (pH 7.5) and dilute to 1000 ml with distilled water.

Dilute 10 ml of this solution to 100 ml with Phosphate buffer Solution (pH 7.5). Measure the optical density of the final solution (0.001% w/v) in a suitable spectrophotometer, using a 1 cm cell and slit width of 0.10 mm at 403-406 nm, recording the maximum within this range.

The percentage of sodium copper chlorophyllin is given by the expression:

\[
\frac{\text{optical density} \times 10^4}{565 \times \text{weight of sample (g)}}
\]

This formula was derived on the assumption that 100% pure sodium copper chlorophyllin has a specific absorbance of 565.