

# Detection of Sudan Dye in Red Chilli Powder

## Abstract

Sudan Dyes are used as colouring agents for chillies . They are food adulterants. Toxicity has been determined in various samples and analytically measurements have been carried out with the help of HPLC Graphical representation of the results have been made.

## Introduction

The spice chilli is widely distributed in India. It is an indispensable condiment of every Indian home. This spice stimulates saliva and gastric juice & helps in digestion. It has primary origin in Mexico & a secondary origin in Guatemala. India is the largest producer of chillies in the world, contributing 25% of the total world production. Around 97% of the total production of chillies is consumed within the country and only 3% is exported<sup>1</sup>. There are two common species of chillies in India viz *Capsicum annum* and *Capsicum frutescence* "the whole hot peppers". Almost all the varieties of chilli that are cultivated in India as a cash crop belongs to *Capsicum annum*.

Chilli [*Capsicum annum*] is one of the major crops of the night shade family having diversified use in household industrial purpose. One of the most important attribute of chillies is its attractive red color. Red color i.e. extracted from non pungent chilli fruit is extensively used in food and cosmetic industries. The major pigments responsible for red color in chilli are the carotenoids 'capsorubin' collectively known as 'oleoresin'<sup>2</sup>.

## Sudan Dyes Colouring

Sudan dyes are red dyes that are used for coloring solvent, oil, waxes, petrol and shoe and floor polishes. They have also been discovered in some important chilli powder from India. Sudan dyes (Sudan I,II,III,IV) are familiar of compounds in class of Azo dyes that are used for different scientific and industrial applications. They have been used in paints and cosmetic products. Sudan dyes are however illegally used as food adulterants particularly in red chilli powder & in chilli containing food such as curry, spice mix because of their intense red orange color & low price.

## Toxicity in Sudan Dye

Sudan Red compounds are hydrophobic azo dyes, still used as good additives in some countries. They have been shown to be unsafe, causing tumors in the liver and urinary bladder in rats; Sudan dyes have a carcinogenic effect a potential risk of gen-toxicity.

Sudan I was considered by the joint FAO/WHO expert committee on food additives (JECFA) IN 1973 to be unsafe for use in food.

Sudan dyes have reported as contact allergens and sensitizers. The greatest cancer has been on their possible carcinogenicity.

Sudan I is classified as a carcinogen by the International Agency for Research on Cancer (IARC)<sup>3</sup>.

Sudan I is not permitted food color additive under the color in Food Regulation(1995)<sup>4</sup>. Azo-colorants are biologically active through their metabolites<sup>5</sup> and have been associated with increased occurrences of bladder cancer in textile and bather, dyers, painters and hair-dressers<sup>6,7</sup>. Due to their potential carcinogenicity, many countries have banned the use of most azo dyes at any level in products for human consumption<sup>8</sup>.

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**Determination of Sudan in Chilli Powder****Collection of Sample**

The samples of chilli powder were collected from different markets of different areas in different forms i.e. in loose form & in sealed packets.

**Principle**

Sudan I,II,III,IV is extracted from the prepared sample with methanol filtered & concentrate at 60°C under vacuum & determined by HPLC with UV-VIS Detector.

**Reagents**

- GR, AR & HPLC grade reagents were suitable unless otherwise stated water should be ionized, distilled or of similar quality Sudan I,II,III & Sudan IV (a cross organic/fluca/sigma aldrich).
- Sudan I, II, III, IV Stock Solution:** weighed 0.1000g Sudan I,II,III & IV & transferred to a 100 ml volumetric flask with methanol & mix well. This solution has a concentration of 1000 mg/L.
- Working Solution:** Prepare the working solution as appropriate dilutions by diluting with methanol once prepared solutions should be protected from light as far as is practicable by wrapping with aluminium foil or stored amber glass bottle.
- Potassium dihydrogen phosphate.
- Tetrabutyl ammonium bromide.
- Methanol HPLC GRADE.
- Mobile phase for HPLC up as follows:
  - 0.6804 g potassium dihydrogen phosphate made up to 400 ml with water.
  - 2.0240 Tetrabutyl Ammonium bromide made up to 1600 ml with HPLC grade.
  - Methanol HPLC grade.

**A. Apparatus:**

- Normal laboratory glassware and apparatus 12ml. Screw top vials/stopper test tube for holding for sample extract.
- 0.45 micron x 13mm syringe filter.
- HPLC with UV/VIS detector.
- 50 ml measuring cylinder.

**B. Sample Preparation & Estimation Procedure:**

Preparation of test sample:

- Dried chilli products (e.g. chilli powder or crushed spices) need only to be mixed thoroughly.
- Wetted chilli containing food products should be homogenous as possible in a suitable laboratory homogenous.
- Weighed 10 gm sample into 50 ml measuring cylinder/volumetric flask.
- Added 50 ml methanol to the test portion.
- Shaked for 30 sec.
- Allowed to stand for 30 min shaking occasionally.
- Filtered the supernatant through a filter paper into a rotart evaporator flask.
- Concentrated under vacuum at 60°C to 5 ml.

- Filtered the sample through syringe filter 0.45µ & collected in vial.
- Protected all sample extract from light as far as possible by storing in the set up the HPLC system as follows-
- OD 54.6x250 mm/mobile phase: (A+B) : C(15:85)
- Flow rate-1.2 ml/min.
- Injection volume :20µl
- Detector :510 nm.
- Run time :30 min.
- Identification: Identified the peak of sudan I, II, III, & IV comparing with retention time of the corresponding standard material.

**Calculation**

$$\text{Concentration (mg/kg)} = \frac{\text{Area of Sample}}{\text{Area of Standard}} \times \frac{\text{Concentration of Standard}}{\text{Weight of Sample}}$$

- A- Area of identified peak in test portion.  
B- Area of identified peak in working standard.

NAME OF THE DYE	AREA OF SAMPLE (PER UNIT)	AREA OF STANDARD (PER UNIT)	WEIGHT (mg/kg)
Sudan I	7844208	37866791	20.7
Sudan II	5918393	54156010	10.92
Sudan III	1707756	13777009	12.39
Sudan IV	1622283	5943480	27.28

**Result & Discussion.**

The result of analysis of spice chilli powder collected from different markets of Delhi in different forms. Samples of chilli powder were analyzed quantitatively.

Out of 10 samples of chilli powder, 4 were found adulterated with non-permitted coal tar dyes. These 4 samples were found artificially coloured with Sudan-I, II, III & Sudan-IV.

The samples of chilli powder were found sold in loose form & also in sealed packets in the market of urban areas, rural areas & resettlement colonies.

**References**

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