



# Spectroscopic Methods for Screening Sudan dyes from Chili products

A thesis submitted to the Faculty of Science, University of  
Colombo for the Degree of Master of Analytical Chemistry in  
the Department of Chemistry

UCFS



P17168

E H M Ranasinghe  
March 2011

## ABSTRACT

Sudan dyes are synthetic azo dyes widely used in industrial and scientific applications. Although recognized as carcinogens, Sudan dyes are added to food such as chili powders to imitate, intensify and prolong the appearance of natural red color. Because of their carcinogenicity, Sudan dyes are illegal as food additives. Due to fraudulent use of Sudan dyes as food colorants, their presence in different food matrices, especially in different brands of chili powders, sauces and related products have drawn attention of chemist and analyst throughout the world.

A simple and fast analytical protocol was developed for detection of Sudan dyes (I, II, and IV) in commercially available chili powders and chili containing products based on characterizing samples through their UV-visible spectra and fluorescence emission spectra.

A reference chili extract sample was prepared by grinding the dried red chili pods using the standard procedure. The reference chili sample was spiked with a single type of Sudan dye (I, II and IV) and a mixture of Sudan dyes (I-II, I-IV and II-IV).

The UV-visible spectra due to different Sudan dyes with chili extracts are indistinguishable. Therefore, spectra were resolved mathematically through the derivartization of the absorbance signal. When chili is spiked with the single type of Sudan dye, the first-order derivative of absorbance spectra shows wavelength shift of maxima and significant differences in minima of Sudan I, Sudan II and Sudan IV for the wavelengths of 523 nm, 543 nm and 563 nm.

Therefore, Sudan dyes from chili powder can be quantified by preparing a calibration curve relating to the wavelength shift of absorbance maxima or relating  $dA/d\lambda$  at relative wavelength at minima of each Sudan dye to concentrations of Sudan dyes.

Quantification of Sudan dye content in chili powder can be done by above calibration plots for the concentration range of 1 ppm to 14 ppm and 1ppm to 20 ppm for the Sudan I and Sudan II or Sudan IV respectively.

When chili is spiked with the mixture of Sudan dyes, mixture of Sudan II and IV can be easily identified and quantified. However, the mixture of Sudan I with Sudan II or Sudan IV is difficult to identify separately. Quantitative analysis of mixture can be done in the concentration range of 2 ppm to 10 ppm.

Fluorimetry is highly sensitive and specific method compared with the UV-visible spectrophotometry. Therefore, another analytical protocol was developed for qualitative and quantitative analysis of Sudan dyes in chili powder using fluorimetry.

When chili is adulterated with a single type of Sudan dye, the spectra show significant difference. There were several prominent emission intensity peaks at different emission wavelengths. Sudan I and Sudan II show linear relationship between emission intensity ratios of 355 : 540 versus concentration. The best linear relationship was shown between 0-25 ppm. Sudan I and Sudan IV have shown linear relationship between emission intensity ratios of 390: 540 vs concentration. The best linear relationship was shown between 0-25 ppm. Quantitatively this protocol can be applied to determine concentration of Sudan dye in chili powder 1 ppm to 25 ppm by plotting calibration curve relating the ratio of fluorescence emission intensity at different wavelengths to concentrations of Sudan dyes.

Commercially available seven different samples were collected from different places in the suburbs of Colombo. The six out of seven samples shows similar shape of spectra, which have not shown significant deviation from the spectrum of reference chili sample. The UV-visible spectra of commercial samples have similar shapes which are identical to the spectrum of the reference chili extract. One commercial sample was deviates from the reference chili sample. The shape of spectra is similar to adulterated chili sample. The wavelength of  $\lambda_{max}$  of first order derivative curve of absorbance shows a red shift. According to that result, it can be predicted that chili has been adulterated with a single type of Sudan dye.