# Isolation and Composition of a Natural Dye from the Stems of Sorghum bicolor (L.) Moench subsp. americanum caudatum

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#### ABSTRACT

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A simple process has been developed for the isolation of a natural dye from the stems of *Sorghum bicolor* (L.) Moench subsp. *americanum caudatum*. The process, with a yield of 20% dry material, can be extrapolated to an industrial scale for use in the food and cosmetic industries. The composition of the dye was elucidated by high-performance liquid

chromatography, which identified a major anthocyanidin (apigeninidin, 17% of the extract) and two main flavonoids (luteolin and apigenin, 9 and 4%, respectively). The absence of the corresponding flavonoid glucosides was confirmed.

Sorghum, a typical African cereal, is a nutrient widely consumed in the Sahel region. Its stems are also used as fodder and building material. Sorghum varieties with colored seeds are used chiefly to provide malt for brewing a sorghum beer called *Ndolo*.

The chemical components of the sorghum grain cuticle are flavonoids, anthocyanidins, and tannins (Gupta and Haslam 1978, Bullard et al 1981, Glennie 1983, Mc Grath et al 1983, Hahn et al 1984, Doherty et al 1987, Kaluza et al 1988). Pigments reported within sorghum species and varieties include apigenin, quercimeritrin, kaempferol glucosides, and apigenidin glucosides (Nip and Burns 1969, Tomita and Kimura 1982). Moreover, apigeninidin, luteolinidin, and 7-O-methyl luteolin glucoside have been found in the cuticle of Sorghum durra glumes (Misra and Seshadri 1967). So far, no investigation has been devoted to the pigmentation of the stems. Therefore, the isolation of the dye from stems of Sorghum bicolor (L.) Moench subsp. americanum caudatum from the northern part of the Ivory Coast is reported here.

## **MATERIALS AND METHODS**

## Material

Stems of Sorghum bicolor (L.) Moench subsp. americanum caudatum from Korhogo, Ivory Coast, were supplied by Ake Assi, Department of Botany, University of Abidjan, Ivory Coast.

## Dye Isolation

Extraction was performed according to Tomita and Kimura (1982). Stems (100 g) were dried at 20°C, powdered (IKA-Labotechnik grinder model A 10), bolted (particle size <0.20 mm), and mixed with 0.25N NaOH (2,000 ml) at 60° for 3 hr. The solution was then filtered and cooled; 10N HCl was added to bring it to pH 3. The precipitate was filtered, washed with distilled water, dissolved in 0.01N Na<sub>2</sub>CO<sub>3</sub> (200 ml), and mechanically stirred at 80°C for 1 hr. After cooling, the pH was readjusted with 10N HCl to pH 3. The precipitate was filtered, washed with distilled water, and dried at 60°C for 1 hr to yield a red dye. The whole process was repeated five times on different samples to check reproducibility. From 100 g of stems, 19–21 g of pigment was obtained.

#### **Dve Analysis**

High-performance liquid chromatography (HPLC) analyses were performed on a chromatograph (Varian model 5000) equipped with an injector (Rheodyne model 7125) and a photodiode-

array detector (Merck L3000) under computer control (Merck HPLC Manager). Compounds were separated at 20°C on a reversed-phase column (Lichrospher 100 RP-18, 125-  $\times$  4-mm i.d., 5-mm particle size). The mobile phase was water, tetrahydrofuran, phosphoric acid (85%), and isopropanol (58:40:1:1, v/v) at a flow rate of 1 ml/mn. The injection volume was 10  $\mu$ l. UV detection was monitored at 270 nm. Pure apigenin, luteolin, and apigeninidin chloride were purchased from Extrasynthese S.A. (Genay, France).

#### RESULTS

The precipitate obtained by extraction of sorghum stems was  $20 \pm 1\%$  of previously dried stems.

According to HPLC data, the constituents of the red dye are: an anthocyanidin, apigeninidin ( $17 \pm 1\%$  of the extract); and two flavonoids, luteolin and apigenin ( $9 \pm 1\%$  and  $4 \pm 1\%$ , respectively) (Fig. 1). Neither cyanidin nor luteolinidin were encountered. Moreover, only aglycones appear to be present, not the corresponding glucosides.

## **DISCUSSION**

In the literature, the pigment yields reported from grains were only in the range of 0.8 to 1% (Misra and Seshadri 1967, Doherty et al 1987). Isolating the dyes from stems in our procedure strongly improved the yields. We obtained a yield of  $\sim 20\%$  dry matter. The red dye from sorghum stems is highly soluble in weak alkaline solution or in propylene glycol.

Utilization of the pigment in the food and cosmetic industries may be considered. Of particular advantage are the high yields and low cost, as well as valorization of sorghum in developing countries, especially those in Africa.

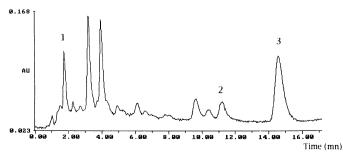


Fig. 1. High-performance liquid chromatography profile of the natural dye extracted from sorghum stems. 1, apigeninidin; 2, luteolin; 3, apigenin.

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