IOSR Journal Of Pharmacy And Biological Sciences (IOSR-JPBS)
e-ISSN:2278-3008, p-ISSN:2319-7676. Volume 15, Issue 3 Ser. IV (May –June 2020), PP 28-30
www.Iosrjournals.Org

Chlorophyll content in pearl millet (*Pennisetum glaucum* L.) genotypes

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Abstract: *The pigment contents in leaves, including chlorophyll (a), (b) and carotene vary considerably with plant genotype and could increase or decrease with plant age.Chlorophylls are the essential catalysts of photosynthesis and occur universally as green pigments in all photosynthetic plant tissues; they occur in the chloroplasts in relatively large amounts, often bound loosely to protein. In this study the pigments concentration in leaves of the four pearl millet genotypes was assessed. This includes chlorophyll (a), chlorophyll (b), ratio of a/b, total chlorophyll content and carotene content. The results reflected that the highest chlorophyll content was found in leaves of the genotype Bauda (47.00mg/g), followed by Darmasa (43.60mg/g), Ugandi (41.13mg/g) and finally Madlkawia with the lowest value (38.56mg/g). The ratio of a/b was high in Bauda and Darmasa which were 0.65 and 0.72 respectively; while it was low in Ugandi and Madlkawia, these were 0.48 and 0.51 respectively.Carotene content was very high in Bauda leaves (9.6 x 10^6 mg/g), far exceeding the results of the other three genotypes which ranged almost around 1.0 x 10^6mg/g.*

Date of Submission: 04-06-2020 Date of Acceptance: 20-06-2020

I. Introduction and Literature Review

P. glaucum is the third in importance after sorghum and wheat in the Sudan. The grain is particularly devoid of paste during storage and has long storage and keeping quality (Chandra and Matta, 1990).Pearl millet is drought tolerant and cannot withstand saturated soils. In addition, naturally it will do better in rich soils; the plant is also capable of growing and yielding a crop on very poor soil.Most species of *Pennisetum* are protogynous, but pearl millet is more conspicuous in this regard. This facilitates the introgression of characters from other annual penicillaries into pearl millet and hence has helped in the genetic enrichment of this species (Jauhur 1981). It responds very well to heterosis breeding (Hazza, 1994) that is why pearl millet has high variability (Salih 2006).

The visible color of plant pigments are the result of absorption of light energy and the reflection of specific wavelength of light. Theproperty of light absorbance in pigments provide a base for quantitative analysis (Stryer, 1988).The coloration seen in leaves, stems, flowers and fruits is due to several pigments that play a role in photosynthesis and plant protection. While most plants appear green, they contain several other pigments of different colours that are masked by the dominant chlorophyll molecules. The secondary pigments include the lipid soluble orange carotene and yellow xanthophylls.Chlorophylls are the essential catalysts of photosynthesis and occur universally as green pigments in all photosynthetic plant tissues; they occur in the chloroplasts in relatively large amounts, often bound loosely to protein but are readily extracted into lipid solvents such as acetone or ether.Chlorophylls (a) and (b) occur in higher plants, ferns and mosses; chlorophylls (c) to (e) are only found in algae while other chlorophylls are confined special to certain bacteria (Harborne, 1988).Chlorophylls are very effective photoreceptors because they contain net-works of alternating single and double bonds: such compounds are called polyenes; they have very strong absorption bands in the visible region of the spectrum, where the solar output reaching the earth is maximal.Also absorption spectra of chlorophyll (a) and (b) are different. Waves which are not captured by chlorophyll (a) at 460nm for example are readily captured by chlorophyll (b), which has intense absorption at that wavelength. Thus the two kinds of chlorophylls complement each other in absorbing the incident sunlight (Stryer, 1988).

Carotenoids which are C40 tetraterpenoids are an extremely widely distributed group of lipid soluble pigments found in all kinds of plants from simple bacteria to yellow flowered composite. In animals, one particular carotenoid; B-carotene is an essential dietary requirement, since it provides a source of vitamin A, a C20 Isoprenoid alcohol. There are now over 600 known carotenoids only few are common in higher plants (Harborne, 1988).Carotenoids serve as light-harvesting molecules in photosynthetic assemblies and also play a role in protecting prokaryotes from the deleterious effect of light.The visible colour of plant pigments are the result of absorption of light energy and the reflection of specific wavelength of light, the property of light absorbance of pigments provide a basis of quantitative analysis (Stryer, 1988).The pigment contents in leaves,

including chlorophyll (a), (b) and carotene vary considerably with plant genotype (Jauhur, 1981) and could increase or decrease with plant age (Mukminah, 2001).

Materials and Methods Four pearl millet genotypes collected from Western Sudan (table 1) were used in this study.

Table 1: Four genotypes of pearl millet used in the study and their origins

No.	Accession	Origin	Notes
1	Bauda	Darfur/ Western Sudan	Check
2	Darmasa	Darfur/ Western Sudan	Collection
3	Ugandi	Uganda/adopted	Check
4	Madlkawia	Kordofan/ Western Sudan	Collection

1. Pigments extraction

Chlorophyll a, chlorophyll b and carotene pigments were extracted following the method adopted by Harborne (1988). Ten grams of fresh green leaves from 3 week old seedlings were collected from each genotype.Leaves were macerated in methanol by means of pestle and mortar. The macerated solutions were filtered twice, once with a piece of clean cloth and refiltered again by means of filter papers through glass funnels.

2. Determination of Pigments Concentration

The absorbance of the 4 solutions was read spectrophotometerically at the following wave length: 470, 649, 653, 665 and 666nm.The concentrations of chlorophyll a, chlorophyll b and carotene were calculated using the formula adopted by Wellburn (1994):

Chlorophyll a (Ca) = 15.65 A666 – 7.34 A653

Chlorophyll b (Cb) = 27.05 A649 – 5.32 A665

Carotene = (1000 Ax¬470-2.86 Ca – 129.2 Cb) 221.

Where Ax is the absorbance at x wave length.

II. Results and Discussion

The concentrations of chlorophyll (a), chlorophyll (b) and carotene were calculated for the four pearl millet genotypes, using the equation adopted by Wellburn (1994). The results were presented in table (2) including total chlorophyll content (mg/g) and the ratio of chlorophyll (a) to chlorophyll (b) (a/b).

Table 2: Chlorophyll (a), Chlorophyll (b), total chlorophyll, ratio of a/b and carotene content in leaves of four pearl millet genotypes

Genotype	Chlorophyll (a) mg/g ± S.D.	chlorophyll (b) mg/g ± S.D.	Total chlorophyll mg/g ± S.D.	Ratio of a/b ± S.D.	Carotene mg/g ± S.D.
Bauda	18.57 ± 1.78	28.41 ± 1.05	47.00 ± 1.96	0.65 ± 0.04	9.6×10^6 $\pm 5.97 \times 10^4$
Darmasa	18.26 ± 1.89	25.32 ± 1.19	43.60 ± 2.08	0.72 ± 0.07	0.90×10^6 $\pm 2.50 \times 10^4$
Ugandi	13.74 ± 2.87	28.75 ± 3.37	41.13 ± 3.65	0.48 ± 0.03	1.3×10^6 $\pm 3.4 \times 10^4$
Madlkawia	13.10 ± 2.49	25.67 ± 2.03	38.56 ± 2.85	0.51 ± 0.03	1.0×10^6 $\pm 2.90 \times 10^4$

Total chlorophyll content ranged between 47.0mg/g for Bauda and 38.6mg/g for Madlkawia as the minimum total chlorophyll content. Darmasa and Ugandi total chlorophyll content were 43.6 and 41.1mg/g respectively. a/b ratios were: 0.72, 0.65, 0.51 and 0.48 recorded for Darmasa, Bauda, Madlkawia and Ugandi respectively (table 2).These results agree with the visual observation of the leaves color. Leaves of Bauda and Darmasa had blue green color while those of Ugandi and Madlkawia were of yellow green color. Blue green is indication of high concentration of chlorophyll a, while yellow green is an indication of high concentration of chlorophyll b.Jauhur (1981) who stated that pigment content vary considerably with the plant genotype.Carotene content was very high in Bauda leaves (9.6×10^6 mg/g), far exceeding the results of the other three genotypes which ranged almost around 1.0×10^6mg/g (table 2). Concerning the carotene content, the genotype Bauda had a very high value far exceeding the other three genotypes, similar to the results obtained by the maximum velocity of the enzyme peroxidase (Salih, 2006).

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Manal A. M. Salih, et. al. "Chlorophyll content in pearl millet (Pennisetum glaucum L.) genotypes." *IOSR Journal of Pharmacy and Biological Sciences (IOSR-JPBS)*, 15(3), (2020): pp. 28-30.