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Effect of Chemical and Organic Fertilizers on Some Chemical Components of Calyces and Seed of Roselle (*Hibscus sabdriffa* L)

Abdelbagi Ahamed Abdelbagi, Elkheir Hassaballah Abdallah Ahamed, Elbager Idris Mohammed Lbrahim

Department of Agronomy, Faculty of Agriculture, Omdurman Islamic University, Sudan
Corresponding author: E-mail: ahamedelkheir@yahoo.com

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Abstract

The nutritional importance of roselle calyces depends upon their chemical composition. Afield experiment was conducted for two consecutive summer seasons (2011 and 2012) at the Top Farm of the Faculty of Agriculture, Omdurman Islamic University (*Fitaihab*). The work was conducted to investigate the effect of chemical and organic fertilizers on some chemical components of calyces and seeds of two roselle (*Hibscus sabdriffa* L.) varieties. The experiment was set in a split- plot design with four replications. The treatments were two varieties namely, Bulk (V_1 – local) and Omshiback (V_2- improved). Chemical fertilizers involved 0, 40, 80 kg N/fed and 40 N + 40 kg P_2O_5/fed, beside 2 tons chicken manure/fed and 3 tons cattle manure/fed (denoted as T_0, T_1, T_2, T_3, T_4 and T_5, respectively). Calyx chemical components taken were calyx anthocyanin content (%), calyx protein content (%) and calyx phosphorous content, in addition to seed oil content (%). The findings of the study revealed that all studied calyx chemical components (anthocyanin, protein, phosphorous) and oil content in both seasons were not significantly affected by varieties, fertilizers and their interaction, except P content, which was significantly affected by fertilizers in both seasons. In the first season, T_5 treatment significantly increased calyx phosphorous content relative to T_0, T_1, T_2, T_3 and T_4 treatments by about 21.9%, 25.0%, 16.0% and 14.2%, respectively, whereas T_2 treatment in the second season significantly increased it as compared to T_0, T_1, T_3, T_4 and T_5 by about 31.8%, 16.0%, 41.5%, 18.4% and 11.5%, respectively.

Keyword: Roselle, anthocyanin, protein, calyces, phosphorous

Introduction

Roselle (*Hibscus sabdriffa* L) belongs to the family Malvaceae and it's an important annual crops, which grows successfully in the tropical and sub tropical regions for its popular edible calyces, stem fibers, leaves and seeds (Cobley, 1975; Babanjide *et al* ., 2004; Mahadevan, 2009). The main producing countries include India, China, Jamaica, Nigeria, Cuba, Philippine, Indonessia, Somalia, Sudan, while the main importing are USA, France Italy, Germany and Arab countries. The crop is imported by these countries for its economics importance which lies on several uses of its parts. The fresh and dry calyx, which stands for the

main economic parts, is used as cold and hot drink. Calyx extraction is also used for coloring of food and for pharmaceutical and cosmetic purposes. The seed contain 17-23% oil, which is rich in oleic, linoleic, epoxyoleic and cyclopropenoids acids (khidir, 1997). In addition the drink has a laxative effect due to organic acids present in the calyxes and the calyx extraction has a great therapeutic action for curing heart and nerve diseases and high blood pleasure. It presents antimicrobial activities due to phenolic compound and also it contains fibers, calcium, iron, carotens, high amount of ascorbic acids, rich in riboflavin, niacin and vitamin C whereas, seeds of this plant have found to be a good source of protein (Babanjide *et al*., 2004; FAO, 2004; QI et al., 2005; Fasoyiro *et al.*, 2005; Halimatul et al., 2007; Mukhtar, 2007; Hassan, 2009; Abo-Bakar and Gehan 2011; Anokwuru *et al*., 2011).

Anthocyanin is a water-soluble pigment responsible for the orangey-red color of calyces and their juice (Tsai and Ou, 1996). When the concentration of anthocyanin is high, roselle calyces are good colorant and also a potential of good source of anti-oxidant component. The percentage of anthocyanin based upon varieties. Eltinay and Ismail (1985) reported 1.4% anthocyanin, whereas Hassan (1988) reported 2.6% and 0.66% anthocyanin for two varieties. In another investigation, Eltinay and Ismail (1985), farid *et al*, (1994), El Tinay and Cair, (1993), Okosun *et al*., (2006), Egharevba and Law-Ogbomo (2007), and Alshoosh (1997) stated that anthocyanin content was significantly increased by application of both nitrogen and phosphorus fertilization. Osman (1998) found that application of both N and P significantly increased anthocyanin content in roselle calyces, with high value obtain by application of 80 Kg N/ha and 50 Kg P_2O_5/ha. Abdelbagi (2001) also observed that anthocyanin content was significantly affected by both genotype and phosphorus application, whereas nitrogen did not significantly affect this character. Ahmed *et al*. (1998) showed that application of 14, 18, and 27 m^3/cattle manure /fed to roselle plants improved the quality of calyces compared to untreated one. Moreover, Nabil and Aly (2002) and Gad (2011) found that application of both chicken and cattle manures significantly increased anthocyanin content.

Eltinay and Ismail (1985), farid *et al*, (1994), Alshoosh (1997), Osman (1998), Abdelbagi (2001), Ahmed *et al*. (1998), Nabil and Aly (2002) and Gad (2011) found that calyx chemical contents of roselle (anthocyanin, protein, P and oil) significantly affected by application of chemical fertilizers. In fact, in Sudan roselle, which is called *karkade* is grown mainly by the traditional farmers in the rainfed areas at Kordofan and Darfur, beside that it is successfully grown as cash crops under irrigation system (N.B.A.P, 1999). The crop is not fertilized although many investigations recorded that it responds very well to chemical and organic Fertilizers. Babanjide *et al*. (2004) showed that application of NPK to roselle significantly improved calyx quality. Similar results were also reported by Abdelbagi (2001), when roselle plants were fertilized by nitrogen and phosphorus element. On the other hand, Gad (2011) mentioned that Roselle calyx quality was significantly influenced by application of different sources and rates of organic fertilizers. Also Ahmed *et al*. (1998) pointed out that both chicken and cow manures improves chemical constitution of calyces and seed of roselle**.** The objective of this study was to investigate the effect of chemical and organic fertilizers on some chemical components of calyces and seed of

roselle, and determine the best type of fertilizer.

Materials and methods:

Location and description of experimental layout:

Afield experiment was conducted at two consecutive summer seasons during 2011 and 2012 in the Demonstration Farm of the Faculty of Agriculture, Omdurman Islamic University, (*Alfitaihab*), latitude 15º 34 N, longitude 32º 34 E and about 393 m above the sea level. The soil pH character ranging between 7.5 – 7 which suffer from low depth and fertility (Amin and Fadual 2007).

Two varieties of Roselle (*Hibscus sabdriffa* L) namely Bulk and Omshiback were used. The Bulk variety described as local variety and characterized by red stem and small seeds which referred as V_1 and Omshiback described as improved variety and characterized by dark red stem and large seeds, referred as V_2. The seed of both varieties were obtained from Elobaied Research Station, Agriculture Research Station – Sudan (production of 2009/2010 season).

The fertilizer treatments include two chemical fertilizers (N and P) and two organic fertilizers (Cow and Chicken manure), in addition to check treatment (T_0). Nitrogen treatments were 40 kg N/fed. (referred to as T_1), 80 kg N/fed. (referred to as T_2), and a combination of 40 kg N/fed. + 40 kg P_2O_5/fed. (referred to as T_3). Fermented organic fertilizers (Cow and Chicken) were: 3 tons/fed, cow manure (referred to as T_4) and 2 tons/fed, chicken manure (referred to as T_5). The process of fermentation of both cow and chicken manures as compost was done according to Billington (1943). The treatments were applied at the 3rd week of July for both 2011 and 2012 seasons. The experiment was a signed in a split–plot design with four replications according to Gomez and Gomez (1984). The varieties were allotted to the main plots, whereas the fertilizer treatments were designated to the sub-plots.

Husbandry

The land was disc-ploughed, leveled and then ridged at 70 cm spacing. The experimental unit was a plot of 12.5 m^2 contained 6 ridges. The sowing date was at 20th July for the two seasons. Five seeds per hole were sown on the top side of the ridge at 50 cm spacing between holes. Phosphorus, cow and chicken fertilizers were then applied on the bands of one side of the ridge at the depth of 3 inches, while nitrogen fertilizer was broad casted. The crop was immediately irrigated after sowing and then irrigated consequently every 7 days; accordingly, the crop received 14 irrigations during the season, which extended to about 4 months. Thinning to 2–3 plants/hole and re-sowing were carried-out after the 3rd irrigation for both seasons. Three manual weeding were done after the 3rd, 5th and 6th irrigations. No pesticides or fungicides were used in both seasons.

Chemical analysis of calyces and seed

Chemical analysis was done for only three replications for each treatment in both seasons.

Calyx anthocyanin contents

This parameter was determined by using extraction method according to Allen Quarm (1989) by using a sample of 2.5 g from each treatment was used for this purpose**.**

Calyx protein contents

Nitrogen content of calyces was determined by using Micro- kjeldahl method and then protein content was estimated according to the formula

Protein Content (%) =

$$\frac{\text{total } f \times 0.1 \times 0.014 \times 100}{0.2}$$

Calyx phosphorous contents

Phosphorous content of calyces was determined by chloride reaction method described by Grimshaw *et al*. (1989). A sample of 1.5 g from each treatment was used for this purpose.

Seed Oil Contents (%)

Oil content of seed was determined according to A.O.A.C. system (1979). A Sample of 2.0 g from each treatment was used to estimate this attribute.

Statistical analysis

Data were analyzed using ANOVA of split plot design according to Gomez and Gomez (1984), to detect any effect on calyx and seed chemical compositions due to application of the above mentioned treatments. Means separation was done by using Duncan's multiple range test (DMRT).

Results and Discussion

Calyx anthocyanin content (%)

Table1 shows that in both seasons anthocyanin content of calyces was not significantly affected by Varieties, but it was slightly higher in V_1 (1.40%) than in V_2 (1.21) in the 1st season, whereas the reverse was true in the 2nd season, when V_2 reported 1.09% corresponding to 1.03% for V_1. This result was in contrast to that found by Abdelbagi (2001), who observed that calyx anthocynmin was significantly different between Varieties. Sanyal *et al*. (1961) stated that the production of anthocyanin in roselle is controlled by multiple alleles as well as dominant pigment intensifier genes and hence it shows variation among varieties.

Furthermore, fertilizer treatments also did not significantly affect this parameter in both seasons, but as shown from table1 that T_5 treatment in the 1st season had insignificantly higher percentage of anthocyanin content (1.62%), followed by T_1 (1.34) and T_3 (1.30%), whereas T_0 (control) recorded the lower value in this season (1.05%). In the 2nd season, the insignificantly higher value of anthocyanin (1.27%) was associated with T_0, followed by T_1 (1.23%) and T_4 (1.22%), while T_3 registered the lower value (1.00%). Previous studies (Eltinay and Ismail, 1985; Farid *et al*, 1994; Alshoosh, 1997 and Osman (1998)) indicated that anthocyanin content was significantly increased by application of N and P fertilizers. Also Ahmed *et al*. (1998); Nabil and Aly (2002) and Gad (2011) mentioned that this parameter increased with application of cattle and chicken manures.

Also Hassan (2009) found that application of bio-fertilizers alone or in a combination with chemical fertilizers to roselle cultivars significantly increased total anthocyanin content. The author attributed this insignificant increment to the positive effect of these treatments on growth characters. However, Abdelbagi (2001) observed that application of 0, 50, and 100 Kg N/ha did not significantly affect anthocyanin content of roselle calyx.

Calyx protein Content (%)

Neither varieties nor fertilizers and their interaction significantly influenced protein content of roselle calyces in both seasons, but as shown in table1 that V_1 in both seasons slightly increased this parameter as compared to V_2 by about 1.4% and 1.3% in first and second seasons respectively. Meanwhile, the control treatment (T_0) resulted in insignificantly higher mean of protein content of roselle calyx as compared to all other fertilizers in both seasons (table1), with an increasing as compared to T_1, T_2, T_3, T_4 and T_5 estimated by about 3.4%, 2.4%, 3.6%,

8.4% and 6.8% in the first season and 7.0%, 3.9%, 5.5%, 11.1% and 6.5% in the second season respectively. In contrast, Abdelbagi (2001) found that application of 0, 50, and 100 Kg N/ha significantly affected the protein content.

Table (1): Effect of varieties and chemical and organic fertilizers on anthocyanin and Protein Content of roselle calyx

Treatments	Anthocyanin content (%)		Protein Content (%)	
	2011	2012	2011	2011
Varieties			**Varieties**	
V_1	1.40[a]	103[a]	3.75[a]	6.40[a]
V_2	1.21[a]	1.09[a]	3.70[a]	6.32[a]
S.E±	0.06	0.02	0.10	0.10
CVa (%)	19.33	21.47	8.29	6.48
Fertilizers			**Fertilizers**	
T_0	1.05[a]	1.27[a]	5.17[a]	6.71[a]
T_1	1.44[a]	1.23[a]	5.00[a]	6.27[a]
T_2	1.33[a]	1.09[a]	5.05[a]	6.46[a]
T_3	1.30[a]	1.00[a]	4.99[a]	6.36[a]
T_4	1.10[a]	1.22[a]	4.77[a]	6.04[a]
T_5	1.62[a]	1.21[a]	4.84[a]	6.30[a]
S.E±	0.17	0.10	0.13	0.20
CV b (%)	30.84	19.88	6.59	7.61

Means within columns which having similar letters are not significantly different at 0.05 level of probability according to DMRT

Calyx phosphorous contents (%)

Application of fertilizers significantly affected Phosphorous content of calyces, in both seasons, whereas varieties and the interaction between varieties and fertilizers in both seasons did not significantly affect this character. Abdelbagi (2001) showed no significant difference in calyx phosphorous content of two roselle varieties (0.14% for each), however, Alshoosh (1997) obtained 0.19% and 0.33% phosphorous content for the same varieties.

In the 1st season, the treatment T_5 as shown in table2 resulted in a significantly higher mean of phosphorous content (1.45%) as compared to all other treatments. In this season, T_5 increased phosphorous content of calyces relative to T_0, T_1, T_2, T_3 and T_4 treatments by about 21.9%, 25.0%, 16.0% and 14.2%, respectively. Moreover, in the 2nd season T_2 had a significantly higher percentage (0.58%) of this parameter as compared to all other treatments except T_5. In this season the percentage of increment in this character for T_2 as compared to T_0, T_1, T_3, and T_4 by about 31.8%, 16.0%, 41.5% and 18.4%, respectively. The significant improvement of roselle calyx quality by application of chemical and organic fertilizers was also reported by Ahmed *et al*. (1998); Abdelbagi (2001); Babanjide *et al*. (2004) and Gad (2011). The authors believed that the effect of these sources of fertilizers on plant growth may be behind the improvement of calyx quality. Abdelbagi (2001) showed that application of 100 Kg N/ha to roselle plants significantly increased calyx phosphorous content.

Seed oil content (%)

Oil content of roselle seed was not significantly influenced by varieties, fertilizers and their interaction in both seasons as shown in table2. As shown from the table that the two varieties to some extend had similar values of oil content in their seeds, but they differed between seasons, when the second season exceeded the first season in this parameter by about 50.6% and 45.7% for V_1 and V_2 respectively. Ahamed *et al* (1998) stated that the Sudanese local varieties have oil content ranged between 17.8 – 19.4% however, Bakheet (1989) reported 21.7%, 20.9% and 19.4% oil content for Elrahad, El fashir and Kadogli varieties, respectively.

As for fertilizers, the organic manure treatments (cattle and chicken) obtained slightly increase of this character in comparison to all other treatments, but with no significant differences (Table2). Abdelbagi (2001) showed no significant differences in seed oil content when 0, 50, and 100 Kg N/ha applied to roselle plants in the second season, whereas these treatments in the first season significantly increased this character as compared to control.

Table(2): Effect of genotype and chemical and organic fertilizers on calyx phosphorous and seed oil content of roselle

Treatments	Phosphorous Content (%)		Oil Content (%)	
	2011	2012	2011	2011
Varieties			**Varieties**	
V_1	1.22[a]	0.49[a]	14.22[a]	21.42[a]
V_2	1.30[a]	0.48[a]	14.21[a]	20.70[a]
S.E±	0.02	0.01	0.03	0.37
CV a (%)	7.53	11.41	0.75	7.45
Fertilizers			**Fertilizers**	
T_0	1.19[b]	0.44[cd]	14.29[a]	21.16[a]
T_1	1.16[b]	0.50[bc]	14.05[a]	20.90[a]
T_2	1.25[b]	0.58[a]	13.61[a]	20.93[a]
T_3	1.25[b]	0.41[d]	13.90[a]	21.00[a]
T_4	1.27[b]	0.49[bc]	14.82[a]	21.16[a]
T_5	1.45[a]	0.52[ab]	14.63[a]	21.23[a]
S.E±	0.06	0.02	0.04	0.36
CV b (%)	11.22	14.97	5.22	4.14

Means within columns which having similar letters are not significantly different at 0.05 level of probability according to DMRT

Conclusion and Recommendation

Based on the findings of the present study it could be concluded that calyx anthocyanin and protein as well as seed oil contents of roselle were not significantly affected by the studied varieties and application of organic and inorganic fertilizers, whereas calyx phosphorous content was significantly affected by these fertilizers. It is recommended that more studies in roselle associated with application of organic and inorganic fertilizers (types and levels) should be carried out.

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أثر إستخدام السماد الكيميائي والعضوي علي بعض المحتويات الكيميائية لسبلات وبذور الكركدي

(*Hibscus sabdriffa* L., var. sabdriffa)

عبدالباقي احمد عبد الباقي ،الخير حسب الله عبد الله* والباقر ادريس محمد ابراهيم

قسم انتاج المحاصيل الحقلية– كلية الزراعة – جامعة امدرمان الاسلامية

ahamedelkheir@yahoo.com

المستخلص

الأهمية الغذائية لسبلات الكركدي تعتمد علي تركيبها الكيميائي. أجريت تجربة حقلية لموسمين صيفيين متتاليين (2011 و2012) بالمزرعة التجريبية لكلية الزراعة–جامعة أم درمان الإسلامية (الفتيحاب). أجري البحث بغرض التحقق من أثر إستخدام السماد الكيميائي والعضوي علي بعض المحتويات الكيميائية لسبلات وبذور الكركدي (*Hibscus sabdriffa* L., var. sabdriffa). صممت التجربة بتصميم القطع المنشقة بأربعة مكررات. شملت المعاملات صنفين هما – V_1 Bulk (local) والأخر (V_2– improved) Omshiback. السماد الكيميائي عبارة عن 0, 40 80, و40 كجم نيتروجين /الفدان+,40 كجم خامس أكسيد الفسفور/الفدان بجانب 2 طن ذرق دجاج/الفدان و3 طن روث أبقار/الفدان. المحتوي الكيميائي للسبلات شمل محتوي الأنثوثينين (%), محتوي البروتين (%) ومحتوي الفسفور (%) إضافة لمحتوي البذور من الزيت (%). أظهرت النتائج أن كل معايير المحتوي الكيميائي للسبلات التي تمت دراستها (الأنثوثينين, البروتين والفسفور) ومحتوي الزيت بالبذرة للموسمين لم تتأثر معنويا بالأصناف وكذلك إستخدام الأسمدة الكيميائية والعضوية ما عدا محتوي الفسفور الذي تأثر معنويا بالسماد للموسمين. المعاملة T5 في الموسم الأول زادت معنويا محتوي الفسفور مقارنة بالمعاملات T1, T2, T3 وT4 بمقدار 21.9%, 25.0%, 16.0%و14.2% علي التوالي, بينما المعاملة T2 في الموسم الثاني زادت هذا المعيار معنويا مقارنة بالمعاملات T1, T3, T4 وT5 بمقدار 31.8%, 16.0%, 41.5%, 18.4% و11.5% علي التوالي.