

Effect of Irrigation Regime on the Cultivation of Five Wild Colocynth [*Citrullus colocynthis* (L.) Schrad.] Ecotypes in Terms of Their Growth, Seed Yield and Oil content¹.

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Abstract: The objective of this research was to evaluate the response of five wild colocynth (an important medicinal plant) ecotypes to irrigation regime as a basic input of cultural practices. The research was laid out in a split plot design with three replications. Treatments were two irrigation regimes (Q1= 100 % Etc and Q2= 50 % Etc) and five wild colocynth ecotypes (G, SF, SD, GD and NK). The experiment was run for two seasons (2014 and 2015). Parameters measured were days to 50 % flowering, number of fruits per plant, number of seeds per fruit weight of 1000 seeds, seed yield and oil content of seeds. Days to 50 % flowering were not significantly affected by watering regimes for the two seasons; however, ecotypes were significantly effective in the first season only. Number of fruits per plant was not significantly affected by watering regimes in both seasons; yet, ecotypes were significantly

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affected in the first season only. The highest number of fruits/plant was produced by ecotype G in the first season and by NK in the second season. The lowest number was produced by ecotype SD in both seasons. Number of seeds per fruit was not significantly different between the watering regimes in both seasons; it was, however, significantly different among ecotypes in both seasons. There was no significant difference in seed yields between watering regimes in both seasons. Ecotype GD produced the highest seed yield in both seasons. The two watering regimes and ecotypes were significantly different regarding weight of 1000 seeds in the first but not in the second season. Mean oil percent of seeds resulting from irrigated ecotypes (15.2%) was higher than that of seeds collected from the wild (12.1%). Moreover, mean oil percent from seeds produced by high and low irrigation was individually higher than that of seeds from the wild habitat. In the wild habitat, SD produced the highest (16.0%) oil percent and NK the lowest (6.4%). When irrigated, ecotypes showed differential positive response. In conclusion, wild colocynth ecotypes responded positively to irrigation regarding oil content of seeds, which is the most important product, indicating possibility of inclusion of colocynth in the existing cropping system thereby ensuring its conservation and economic use. More research, however, is needed to determine, precisely, the irrigation water requirement for colocynth seed production.

Key words: ecotype, colocynth, irrigation regime, *Citrullus colocynthis*, seed oil, bitter apple

INTRODUCTION

Medicinal and aromatic plants (MAPs) have been an important resource for human healthcare from prehistoric times to the present days. Between 50000 and 70000 plant species are known to be used in traditional and modern medicinal systems throughout the world (Lange and Schippman, 1997). Nearly 25 % of modern medicines were produced from plants that had been first used traditionally (Hudson

1989). Relatively few species are cultivated, but the great majority is still being provided by collection from the wild vegetation (Srivastava *et al.*, 1996). Unfortunately, however, over exploitation as a result of industrialization, land conversion due to urbanization, inappropriate agricultural policies and habitat loss due to climate change increasingly jeopardized the existence of a considerable portion of world's MAPs species. The rational exploitation of MAPs can depend on two different conservation strategies: *in situ* preservation through development of improved forms of naturally growing plants, and *in domo* conservation through development of cultivation practices as means to conserve the species within the human domain (Wiersum, 2003). Both strategies reflect two degrees of semi-domestication of wild plants (Wiersum and Shackleton, 2005). That is, the process of stimulating domestication is normally considered a two-phased process in which, first proper crop cultivation techniques are developed by scientists, followed by subsequent transfer of the scientifically developed domestication technologies to potential growers. Hence, in response to the combined impacts of dwindling supplies resulting from over-exploitation of the natural resources and the increasing demand due to population growth and the growing global markets, MAPs have been increasingly introduced into the farming system.

Colocynth [*Citrullus colocynthis* (L.) Schrad.], an important medicinal plant, belonging to the Cucurbitaceae family is a desert viny plant that grows mostly in sandy soils. It has a wide and diverse use as anti-cancer, anti-microbial, anti-oxidant, anti-diabetic, anti-inflammatory, analgesic and affects hair growth (Marzouk *et al.*, 2009; Alkan and Sandeep, 2017). So, the plant is a great promise for development of novel drugs with wide range of pharmacological activities which could be utilized for dreadful human diseases because of its effectiveness and safety. Rational exploitation of colocynth as a medicinal plant, therefore, necessitates its cultivation as a crop, which requires determination of its optimum cultural practices. The objective of this research was, therefore, to study the response of five wild colocynth

ecotypes to watering regimes as a basic input in cultural practices for their cultivation/conservation as field crops.

MATERIALS AND METHODS

Experiment site

The experiment was carried out in the experimental field of the Department of Horticulture, Faculty of Agriculture-University of Khartoum at Shambat (15°40'N and 32°32'E). The climate is semi- arid, tropical. Mean maximum and minimum temperatures are as high as 41.6°C in summer and as low as 14.1°C in winter (Adam, 1996).

Ecotypes' seed source

Seeds were collected from wildy growing colocynth plants in the following five diverse ecologies:

G, from Gadarif State, with heavy clay soil.

SF, from Acacia forest on the eastern bank of the White Nile, Khartoum State.

SD, from South Darfur State, with clay loam soil in rich savannah ecology.

GD, from Gash River delta, with alluvial deposit soil, Kassala State.

NK, from North Kordofan State desert with sandy soil.

Treatments and experimental design

Treatments comprised two factors *i.e.* two irrigation regimes (Q1= 100 %Etc and Q2 = 50% ETc) and five wild colocynth (G, SF, SD, GD, NK) ecotypes. The two factors were arranged in a split plot design with

three replications such that the irrigation regimes were randomized in the main plots and the ecotypes in the sub-plots.

Seed treatment ,planting and data collection

Colocynth seeds are known to have seed dormancy. To ensure uniform germination and establishment of plants in the field before commencement of irrigation treatments, seeds of the five ecotypes were scarified by shaking with coarse sand for 10 minutes, then soaked in water overnight before planting, (Baskin, 2014; Menon *et al.*, 2014). Treated seeds were planted in mid- March for the two seasons (2014 and 2015) at 40 cm between plants on beds 1.5 m wide and 4 m long. Plots were irrigated immediately after sowing. Irrigation treatments started six weeks after planting. Number of days to 50 % flowering was recorded for each plot; five plants were randomly selected, one week before harvesting, to record number of fruits per plant and number of seeds per fruit; weight of 1000 seeds, seed yield and seed oil content were recorded after harvesting.

Determination of irrigation regime treatments

Reference crop evapotranspiration ET_0 was calculated by Penman-Monoleith equation as reported by Smith *et al.* (1989) as follows:

$$ET_0 = \left[\frac{0.408 \Delta (R_n - G) + \gamma \frac{900}{T + 273} U_2 (e_s - e_a)}{\Delta + \gamma (1 + 0.34 U_2)} \right]$$

Where

ET_0 = Reference crop evapotranspiration (mm day^{-1})

R_n = Net radiation at crop surface ($\text{Mjm}^{-2} \text{ day}^{-1}$)

T = average temperature at 2 m height ($^{\circ}\text{C}$).

$e_s = S_{vp}, k P_a e_a$ = Actual cp (kPa).

$e_s - e_a$ = Saturation pressure deficit for measurement at 2m height (kPa)

U_2 = Wind speed at 2m height (ms^{-1})

Δ = Slope of vapour pressure curve (kPa °C)

γ = Psychometric constant (kPa °C).

900 = Coefficient for reference crop ($kJ\ kg\ day^{-1}$).

0.34 = Wind coefficient for the reference crop (Sm^{-1}).

G = Soil heat flux ($MJ\ m^{-2}\ day^{-1}$).

The amount of irrigation water was applied to each plot as a portion of crop evapotranspiration (ETc) using CROPWAT model as follows:

1. Applying 100% of crop evapotranspiration.
2. Applying 50% of crop evapotranspiration.

Due to lack of information about colocynth plant, sweet melon was taken as a reference crop. For applying water rates a Maddalena Udina Italia 20, 11/2 flow meter was used. The water quantities applied were determined using the following equation described by Makki and Mohamed (2005):

$$V = \frac{ETc \times I \times A \times 100}{Ea\%}$$

Where

V= volume of water to be applied to each plot (litres).

ETc=Crop water requirement (mm/day).

I = Irrigation frequency (days).

Response of wild colocynth to irrigation

A = Area of the plot.

Ea = Application efficiency (%).

Application of water quantities started in May 2014 after six weeks from planting. Quantities were calculated for each month at seven days interval and the same treatments were repeated for the second season 2015. A total of 2652 and 1326 litres, respectively for Q1 and Q2 were used in eleven irrigations.

Extraction of oil

Extraction of seed oil was carried out at the chemistry laboratory of the Medicinal and Aromatic Plants Research Institute, National Centre for Research, according to the method described by Harborne (1984).

Plants were fertilized by NPK at the rate of 240 kg/ha. One month after planting. Plants were protectively sprayed against pests when needed.

All data for the two seasons were statistically analyzed using Statistical Analysis System, version 9.00 (SAS, 2002).

RESULTS AND DISCUSSION

Irrigation regimes did not significantly affect number of days to 50% flowering in both seasons; yet, ecotypes reflected significant differences among them. The earliest ecotype to flower was SD in both seasons (Table 1). Interaction between irrigation regimes and ecotypes were not significant indicating that variability among the genotypes of the ecotypes was not affected by irrigation. Number of fruits per plant was not significantly affected by irrigation regimes in both seasons; however, ecotypes were significantly different in the first season only. Number of fruits per plant was lower in the first season than the second. It can be attributed to the high rainfall of the first (53.8mm) than the second (7.2mm), which could have interfered with fruit setting (washing off pollen grains from male flowers).

Table 1. Effect of irrigation regimes on number of days to 50 % flowering and number of fruits per plant for five wild ecotypes in 2014 and 2015

Treatments	<u>Seasons</u>	
	2014	2015
<u>Irrigation Regimes(I):</u>	<u>Number of days to 50% flowering</u>	
100% Etc	46.1 ^a	41.3 ^a
50% Etc	46.0 ^a	41.2 ^a
Means	46.1	41.3
<u>Ecotypes (E):</u>		
G	45.7 ^{abc}	42.5 ^a
SF	47.3 ^{ab}	42.7 ^a
SD	44.2 ^c	35.5 ^{ab}
GD	48.2 ^a	43.2 ^a
NK	45.0 ^{bc}	42.5 ^a
Means	46.1	41.3
IxE	NS	NS
	<u>Number of fruits per plant</u>	
<u>Irrigation Regimes (I):</u>		
100% Etc	2.6 ^a	5.3 ^a
50% Etc	2.8 ^a	5.2 ^a
Means	2.7	5.3
<u>Ecotypes (E):</u>		
G	3.3 ^a	4.8 ^a
SF	2.6 ^a	5.3 ^a
SD	1.4 ^b	4.4 ^a
GD	3.2 ^a	6.1 ^a
NK	2.9 ^a	5.7 ^a
Means	2.7	5.3
IxE	NS	NS

Means within column(s) followed by the same letter(s) are not significantly different at $P \leq 0.05$

NS= Not significant

Number of seeds per fruit, weight of 1000 seeds as well as seed yield were not significantly affected by the irrigation regimes in both seasons; yet, ecotypes were significantly different in both seasons (Table 2). Ecotype GD produced the highest and ecotype SD the lowest number of seeds per fruit in the first season, while ecotype SF gave the

highest value and SD the lowest in the second season. Weight of 1000 seeds was significantly different among ecotypes in both seasons. The highest value was produced by ectype SD in both seasons and the lowest resulted from NK in the first and SF in the second season. This result is not surprising as SD gave the lowest number of seed per fruit in both seasons, meaning larger seeds. Seed yield was not significantly affected by irrigation regimes in both seasons; however ecotypes were significantly different and interaction between irrigation regimes and ecotypes was not significant. The highest seed producer was ecotype GD and the lowest was SD in both seasons. Ellidir (1999) showed that yield and yield components of visnaga were not affected by watering regimes. The insignificant effect between irrigation regimes regarding measured parameters, can be explained by the fact that colocynth plant's habitat is generally rather dry and plants are adapted to dryer growing conditions which might be much lower even than the lowest irrigation treatment.

Table 3 shows that mean oil % of seeds resulting from the irrigated ecotypes (15.2%) was higher than that of the seeds collected from the wild habitat (12.4%). Means of seed oil % resulting from high and low irrigation regimes were also individually higher than that of wild habitat. This clearly indicates that ecotypes positively responded to irrigation regimes. In the wild, SD produced the highest oil % (16.0%) and NK produced the lowest (6.4%). When irrigated at 100% ET_c, the highest oil % was produced by SF (18.1 %) and when irrigated at 50% ET_c, SD produced the highest oil % (23.1%). Mean oil % of ecotypes irrigated at low regime (15.6%) was higher than those irrigated at high regime (14.8 %). Previous reports on colocynth's seed oil % ranges were 15-27% (Abdel Gadir, 1995), 20-26 % (Abdalla, 1997) and 14.5-22.2% (El Nour, 2004). Such inconsistency could be attributed to the ecology of plants, season of collection of seeds and oil extraction methods.

Table 2. Effect of irrigation regime on number of seeds/fruit, weight of 1000 seeds (g) and seed yield (t/ha.) of five wild colocynth ecotypes in 2014 and 2015 seasons

Treatments	<u>Seasons</u>	
	2014	2015
<u>Irrigation Regimes(I):</u>	<u>Number of seeds per fruit</u>	
100% Etc	153.3 ^a	272.4 ^a
50% Etc	177.7 ^a	285.9 ^a
Means	165.5	279.2
<u>Ecotypes (E):</u>		
G	160.9 ^b	311.4 ^a
SF	167.9 ^b	341.4 ^a
SD	24.5 ^c	112.7 ^b
GD	281.1 ^a	332.5 ^a
NK	193.1 ^b	297.9 ^a
Means	165.5	279.2
IxE	NS	NS
<u>Irrigation Regimes (I):</u>	<u>Weight of 1000 seeds</u>	
100% Etc	25.6 ^a	26.6 ^a
50% Etc	23.4 ^b	25.6 ^a
Means	24.5	26.1
<u>Ecotypes (E):</u>		
G	21.5 ^c	22.4 ^c
SF	17.6 ^d	16.5 ^c
SD	41.0 ^a	44.1 ^a
GD	26.6 ^b	28.1 ^b
NK	15.9 ^d	19.3 ^d
Means	24.5	26.1
IxE	*	NS
<u>Irrigation Regimes(I):</u>	<u>Seed yield</u>	
100% Etc	0.5 ^a	1.9 ^a
50% Etc	0.6 ^a	1.7 ^a
Means	0.5	1.8
<u>Ecotypes (E):</u>		
G	0.6 ^b	1.7 ^b
SF	0.4 ^b	1.5 ^b
SD	0.1 ^c	1.1 ^b
GD	1.2 ^a	2.9 ^a
NK	0.5 ^b	1.7 ^b
Means	0.5	1.8
IxE	NS	NS

Means within columns followed by the same letter(s) are not significantly different at $P \leq 0.05$ according to Duncan's Multiply Range Test. NS= not significant.

Response of wild colocynth to irrigation

Table 3. Seed oil % from wild and cultivated five colocynth ecotypes

Ecotype	Wild seed	100% Etc (Q1)	50 % Etc(Q2)	Mean(Q1&Q2)
G	11.5	13.8	10.9	12.4
SF	15.2	18.1	18.3	18.2
SD	16.0	14.9	23.1	19.0
GD	11.2	15.1	12.7	13.9
NK	6.4	12.1	12.9	12.5
Means	12.4	14.8	15.6	15.2

CONCLUSIONS

- Neither irrigation regime nor ecotypes significantly affect days to 50 % flowering.
- Number of fruits/plant, number of seeds/fruit and seed yield are significantly affected by irrigation regime; yet ecotypes showed differential response.
- Seed oil percent is higher in seeds from irrigation than seeds collected from the wild; yet, variation among ecotypes in seed oil content is noted

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تأثير كميات مياه الري على إستزراع خمسة أنواع إيكولوجية برية من نبات الحنظل [*Citrullus colocynthis* (L.) Schrad.] من حيث النمو وإنتاج البذور ومحتواها من الزيت

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المستخلص: يتم الحصول على النباتات الطبية والعطرية في معظم الأحيان من خلال الجمع من النباتات البرية في البيئات المختلفة، مما يعرضها لخطورة الإنقراض الناتج عن الجمع الجائر، التوسع الزراعي وتلف البيئة نتيجة للتغير المناخي. عليه فإن الإستغلال العقلاني لهذه المجموعة المهمة من النباتات يستوجب، ضمن طرائق أخرى، أستزراعها كمحاصيل. وهذا يستوجب تحديد المعاملات الفلاحية المناسبة. هدفت هذه التجربة لتقييم إستجابة خمسة أنواع إيكولوجية برية من نبات الحنظل لكميات مياه الري كأحد مدخلات الإنتاج الرئيسية للمحاصيل. تصميم التجربة كان القطاعات العشوائية المنشطرة بثلاثة مكررات. كانت المعاملات كميتان من مياه الري (Etc 100 % و Etc 50 %) وضعتا عشوائيا في القطع الرئيسية و خمسة أنواع برية للحنظل وزعت عشوائيا في القطع المنشطرة. أجريت التجربة لموسمى 2014 و 2015. البيانات التي جمعت وحللت إحصائيا كانت عدد الأيام لبلوغ 50 % من الإزهار، متوسط عدد الثمار في النبات، متوسط عدد البذور في الثمرة، وزن الـ 1000 بذرة، إنتاج البذور ومحتوى الزيت في البذور. لم يتأثر عدد الأيام لبلوغ 50 % من

الإزهار تأثيراً معنوياً ثابتاً (للموسمين) بكميات الري ولا بالأنواع البرية. متوسط عدد الثمار في النبات لم يتأثر معنوياً بنظام الري في الموسمين بكميات مياه الري في الموسمين ولكن الأنواع البرية اختلفت معنوياً فيما بينها في الموسم الأول فقط. أعلى قيمة سجلت للنوع G في الموسم الأول وللنوع NK في الموسم الثاني وكانت أدنى قيمة للنوع SD في الموسمين. فيما لم يتأثر متوسط عدد البذور في الثمرة بكميات مياه الري في الموسمين، اختلفت الأنواع فيما بينها اختلفاً معنوياً في الموسمين. لم تؤثر كميات مياه الري تأثيراً معنوياً على إنتاج البذور في الموسمين ولكن تأثرت الأنواع معنوياً فيما بينها؛ أعطى النوع GD أعلى إنتاجاً في الموسمين. متوسط محتوى الزيت في البذور الناتجة من زراعة الأنواع البرية (15.2 %) كان أعلى قليلاً من البذور التي جمعت برياً (12.5 %). متوسط نسبة الزيت في البذور الناتجة من كمية ماء الري الأعلى والدنيا كانت أعلى، كلا على حدة، من نسبة الزيت في البذور البرية. أنتج النوع SD أعلى نسبة زيت (16 %) في البيئة البرية وأنتج النوع NK أدناها (6.4 %)، ولكن عند الري كانت استجابة الأنواع مختلفة إيجاباً عن بعضها البعض. في الخلاصة، لم تؤثر كميات مياه الري كثيراً أو بصفة ثابتة على نمو النبات وإنتاج البذور، ولكن بالمقابل تباينت الأنواع إيجاباً في استجابتها للري فيما يختص بمحتوى الزيت في البذور؛ وهذا هو الأهم. وعليه يمكن شمل نبات الحنظل في التركيبة المحصولية بالسودان لقيمة الزيت الإقتصادية والطبية العالية، ويتطلب ذلك المزيد من البحث لتحديد المقننات المائية الدقيقة لمحصول الحنظل.