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IRRIGATION OF MULBERRY *TREES* (*Morus nigra* and *Morus alba)* WITH PRETREATED SEWAGE EFFLUENT AND ITS EFFECT ON FRUIT PROPERTIES, ACCUMULATION OF SOME HEAVY METALS IN IRRIGATED WATERS, SOIL, RAW AND WASHED FRUITS

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ABSTRACT: *This trial was carried out to investigate the influence of using the collected sewage effluent of New Borg El-Arab Town after passing through three successive oxidation lagoons to rrigate using of two spices of an old mulberry trees and its effect on properties and content of some heavy metals (Cu, Zn, Mn, Pb, Cd, Cr and Ni)of fruits. Also the concentration of such metals in wastewater after passing through these lagoons and in the cultivated solls of these trees before and after irrigation was determined. The results showed that passing Borg El-Arab sewage effluent through three successive lagoons reduced its content of Zn, Nl and Pb while the others metals, Cu, Cr and Cd were slightly affected. Using pretreated sewage effluent in irrigation caused a noticeable rise in the heavy metals content of the mulberry trees, cultivated soil's and fruits, especially those of black mulberry trees than the control soil and fruits. Generally the determined heavy metals in mulberry fruits could be arranged according to their levels in the following descending order Zn, Cu, Cr, Pb and Cd, respectively. Except Pb, the other heavy metals in fruits of both mulberry trees species were found to be lower than the reported maximum permitted levels of such metals In Egyptian legislation. Also, using such water in irrigation reduced moisture content, T.S.S. and Total sugars of fruits. The ash level of the fruits of both mulberry trees was slightly affected. Soaking fruits in tap water for 15 min as a washlng method, led to slightly increase of their moisture content and a reduction of their T.S.S., ash and total anthocyanins. Washing led to reduce fruits more than 20% of Cu, 30% of Cr, 28% of Zn, 47% of Ni, 30% of Pb and 20% of Cd contents of fruits of both mulberry trees removed after washing process.*

Key Words: *Heavy-metals-Effluent- Washing-mulberry fruits-Irrigation water-Cultivated soils*

INTRODUCTION

Utilization of both pretreated an industrial wastewater and sewage effluent in plant irrigation is one of the promising economical waste disposal

solutions. The main constrains facing utilizing such irrigation water are its harmful effect on plants and human health. Tirmizi, *et al.* (1996) showed that uptake of inorganic elements by vegetables irrigated with the sewage effluent differed according to the vegetables species and their concentration in soil. Generally the dominant pathway for most trace elements to vegetables waste is the roots. According to Angelova *et al.* (1998) the majority of the absorbed heavy metals taken up by roots from soil were fixed and accumulated in the young feed rootlets with a 1 mm diameter. Voutsa *et al.* (1996) stated that trace elements and heavy metals in plant leaves appear to originate mostly as aerosols filling from the atmosphere. High accumulation ofheavy metals due to atmospheric deposition in leafy vegetables was Pb, Cr, and Cd (Zhelyazkov and Nielsen, 1996; Chizzola and Franz, 1996 .Angelova *et al.* (1998) found that the content of heavy metals in the crops grown in an industrial polluted region were greatly concentrated in the rachis, skin and present in low levels in the pulp. Results of Holubowicz (1999) indicated that heavy metals content in both soil leaf had no significant effect on the increase of these metals in apple and sour cherry fruits. Both fruits contained lower levels of such metals than in soil, leaves and the world acceptable standards. (Selem *et al.* 2000) . Holubowicz (1998) reported that heavy metals polluted soil did not affect plant growth and development. Generally washing the fruits with tape water reduce the heavy metals contents (El-Saeid and Shahat, 2000 and El-Sharnoubyand Abd EL-All, 2003). New Borg El-Arab city which lies at 60 Km south west Alexandria, Egypt, is one of the recent industrial areas in Egypt. One of the main problem, which faces extension of an industrial activity in this town is the sewage effluent disposal (Hassan *et al.*, 2002).This investigation aims to study the using collected wastewater of sewage effluent of this town after passing through three successive lagoons with a 5 ft depth to oxidize its organic mater as a source of wate rirrigation for two species of an old mulberry trees The influence of the irrigation with such water on the physicochemical properties and the accumulation of some heavy metals in raw and washed fruits of these two mulberry trees species were studied. Also, the concentration of such metals in wastewater after passing through lagoons and in the cultivated soils of these trees before and after irrigation was determined.

MATERIALS AND METHODS

Materials:-

This experiment was carried out during the period extended from April 2001 to April 2002, in four experimental orchards each of 250 acre area in New Borg El-Arab town.containing 9 or 10 years old mulberry trees. Two of the four orchards were planted with white mulberry trees (*Morus alba*) and the other's two with black mulberry trees (*Morus nigra*). One of the two orchards of white and black mulberry trees was irrigated with Nubaria channel water, the

common irrigated water in this area, while the other two orchards trees irrigated with pretreated wastewater of Borg El-Arab town. During the period of the experiment, the weather was very closed to the mean fluctuation recorded in this area through many years ago, moderately rainy, windy and cold (12-14°C) in winter dry hot (35-38°C) in summer nearly moderate with (25-28°C) temperature in both spring and an autumn. The nature of the soil in this area is calcareous. It consists of 70% sand, 11% clay, and 19% silt with a texture of sandy loam. Samples of soil, irrigation water (normal and wastewater) and mulberry fruits at technological ripeness stage were collected to estimate their levels of heavy metal as following:-

Surface soil sample at 0-20 cm depth were collected according to scheme of Magathaes *et al.* (1985) at April, 2001 before starting irrigation and in October 2001, after 6 months of irrigation from the four mulberry trees orchards. Generally, the mulberry orchards were irrigated twice monthly through the year of experiment.

Irrigation water samples, either from Nubaria canals or pretreated wastewater sewage effluent which derived from an industrial and municipal sources, before and after passing in a stored lagoons prior to use in irrigation were collected from the center of the main irrigation canal of the four orchards.. The water samples were collected 3 times at April, October of year 2001, and March 2002.

Samples of new crops of white and black mulberry fruits which were irrigated either with Nubaria canal or pretreated wastewater were collected through their technological ripening stage in March and April 2002.

Methods:-

The soil, irrigated water, and fruit samples were wet-digested using 1:3:5 v/v mixture of $HClO_4$, HNO_3 and H_2SO_4 then Zn, Cu, Ni, Cd, Pb and Cr were determined using Hitachi Model A- 1800 Atomic Absorption Spectrophotometery. Chemicals of an analytical grade, pyrix glass and deionized water used during determination of heavy metals (Chapman and Pratt, 1961).

Different fruits characters were estimated just after collection or after soaking in tap water for 15 min. Total soluble solids (T.S.S) of white and black mulberry fruit were measured with refractometer (Model-ZWAJ, China), moisture, total sugars and ash of the two mulberry fruits were estimated as mentioned in A.O.A.C (1990). The total anthocyanins of black mulberry fruits was determined as described by Fuleki and Frances (1968).

RESULTS AND DISCUSSION

Heavy metals content in irrigation water: Heavy metals content in irrigated water presented in Table (1) indicated that concentration of the determined heavy metals were generally lower in Nubaria canal irrigation water than the sewage effluent irrigation water. The analysis of heavy metals of El-Nubaria canal water after 6 months showed slight changed in their concentrations.

This means that the sources of these heavy metals were stable. They mostly come from fertilizers and pesticides used to control pests during plants development. Untreated sewage effluent of Borg El-Arab town contained high levels of the determined heavy metals particularly Cu, Zn, Ni and Pb. The main sources of such water drived from the industrial activity of various manufactures in this town.

Pretreated Borg El-Arab sewage effluent by passing through three successive lagoons reduced some heavy metals and did not affect the other's. Zn, Ni and Pb contents were reduced after such pretreatments. Meanwhile, the others Cu, Cr and Cd, were nearly stable. This mean that the unaffected heavy metals are found in water soluble salt forms in such irrigation water.

Table (1) Heavy metals concentration in irrigation water).

Heavy metals (ppm)	Source of water irrigation					
	Nubaria Canal		Sewage effluent	Pretreated stored sewage effluent		
	A_1	B_1		A_2	B_2	C
Cu	0.09	0.12	0.33	0.31	0.30	0.30
Zn	0.20	0.23	0.31	0.20	0.20	0.22
Ni	0.13	0.11	0.24	0.17	0.16	0.16
Cr	0.10	0.10	0.15	0.13	0.14	0.15
Cd	0.06	0.06	0.09	0.08	0.08	0.09
Pb	0.10	0.12	0.80	0.67	0.61	0.63

A_1 and B_1 Nubaria canal water used for irrigation mulberry trees in May and October 2001.
A_2, B_2 and C pretreated stored sewage effluent used for irrigation mulberry trees in May and October 2001, and March 2002.

Storing of the pretreated Borg El-Arab sewage effluent for one year caused slight fluctuation in its content of heavy metals. Again this is an indication that, these heavy metals are presented in water soluble form in such water. According to Abd El-Aal *et al* (1991) and El-Hassanin *et al* (1993) irrigation with sewage water increases of total and available forms of Pb, Ni, Cd, Cu,and Cr in the surface layers of cultivated soil.Same Finding were also reported by Shalaby *et al* (1996)and Hassan *et al* (2002).

Heavy metals content in cultivated soils:- Results of heavy metals in Table (2) declared that Pb found in the highest concentration followed descendingly by Ni, Cu, Zn, Cd and Cr in the mulberry trees cultivated soils irrigated with Nubaria canal, water slight changes in the levels of such heavy metalsin soils were noticed after 6 months of irrigation. Meanwhile using pretreated stored sewage effluent in irrigating these soils caused a noticeable rise in its content of Zn, Cu, Ni and slight increase in the others determined heavy metals. These finding agree with those reported by Selem *et al.* (2000) and Minaisy (2003). They found that irrigation with sewage water

increased the available Fe, Zn, Cu, Mn, Pb and Cd, fine materials and lowered the pH of soils.

Table (2) Available heavy metals in the soil mulberry trees.

Heavy metals (ppm)	Mulberry trees cultivated soil				
	A	B_1	B_2	C_1	C_2
Cu	1.12	1.11	1.12	1.26	1.29
Zn	1.08	1.10	1.13	1.40	1.48
Ni	1.52	1.52	1.50	1.63	1.65
Cr	0.65	0.65	0.69	0.75	0.80
Cd	0.69	0.69	0.71	0.74	0.73
Pb	6.60	6.60	6.40	6.94	7.12

A= Soil before irrigation in May 2001.
B_1, B_2= Soil of white and black mulberry trees after 6 months of irrigation with Nubaria canal water.
C_1, C_2= Soil of white and black mulberry trees with pretreated stored sewage effluent.

Heavy metals in mulberry fruits :-Data in Table (3) showed that black mulberry fruits had less values of heavy metals than white mulberry one. Fruits of both white and black mulberry trees irrigated with Nubaria canal water contained lower levels of the determined heavy metals than that irrigated with pretreated stored sewage effluent.

Table (3) Heavy metals content of mulberry fruits

Heavy metals (ppm)	White mulberry fruits		Black mulberry fruits		Egyptian Standarization for heavy metals
	A	B	A	B	
Cu	1.19	2.10	1.08	2.42	5.00
Zn	1.29	3.00	1.19	3.40	5.00
Cr	0.38	0.74	0.36	0.85	5.00
Cd	0.01	0.04	0.01	0.02	0.10
Pb	0.40	0.59	0.39	0.51	0.30
Ni	N.D	N.D	N.D	N.D	-

A- Irrigated with Nubaria canal water.
B- Irrigated with pretreated stored sewage effluent.
N.D = Not determined.

Generally, the determined heavy metals in mulberry fruits could be arranged according to their levels in the following descending order Zn, Cu, Cr, Pb and Cd,respectively. Except Pb, the others determined heavy metals in both mulberry fruits were very low than the maximum permitted concentration of such metals in Egyptian legislation (Anonymous, 1993). The presence of Pb in high level in fruits may be due to the surrounding air which laden with industrial vapors, cars exhausts and insecticides containing Pb.

Holubowicz (1999) found that fruits of apple and sour cherry trees in Grodziec Maly, Poland, where high heavy metals content in the soil was recorded contained less levels of heavy metals than the acceptable standard valuesp

Some phsicochemical properties of mulberry fruits: Thedetermined physicochemical properties of mulberry fruits in Table (4) showed that, black mulberry fruits contained lowermoisture content, higher T.S.S. and ash values than white fruits. On the other hand, total sugars were nearly similar in both types of fruits. Irrigation with pretreated sewage effluent reduced moisture content, TSS, total sugars and increased the ash of both white and black mulberry fruits. However the anthocyanins content of black mulberry fruits was not affected with source of irrigation water. These results are in partial harmony with that of Amin and Attia (2003) who found that fruits of *Morus albe* species were white in colour, larger in size lower in total sugars than those of *Morus nigra*. .Generally the fruits of *Morus nigra* consideres a good source of anthocyanins (201 mg/100g).

Table (4) Some physicochemical properties of mulberry fruits .

Property	White mulberry fruits		Black mulberry fruits	
	A	B	A	B
Moisture (%)	80.27	78.79	79.82	76.33
Total soluble solid (%)	17.93	16.90	20.61	18.91
Total sugar (%)	68.59	66.92	68.29	66.83
Ash (%)	4.89	5.07	5.75	5.89
Anthocyanin (mg/100g)	-	-	200.4	199.82

A- Irrigated with Nubaria canal water.
B- Irrigated with pretreated stored sewage effluent.

Effect of washing on heavy metals concentration and some physico-chemical properties of mulberry fruits:- Figs (1) illustrates the influence of soaking white and black mulberry fruits in water for 15 min as a washing treatment on some of their physicochemical and content of heavy metals. Soaking in water led to slight increase in moisture content of fruits due to water imbibition through soaking. A reduction in TSS, total sugars, ash and total anthocyanins as a results of leaching out of such components in water. The changes in the previous properties were an obvious in black mulberry fruits than white ones. Fig (2) also showed that more than 20% of Cu, 30% of Cr, 28% of Zn, 47% of Ni, 30% of Pb and 20% of Cd contents of both white and black mulberry fruits were removed through soaking in water. According to Motyleva and Sosmina (1996), Holubowicz (1998), Angelova *et al.* (1998) and El-Saeid and Shahat (2000) nearly 25-30% of heavy metals content of fruits and vegetables were reduced after washing with tap water. El-Sharnouby (2003) found that blanching of vegetables in hot water (90°C) increased from reduction of their heavy metal content.

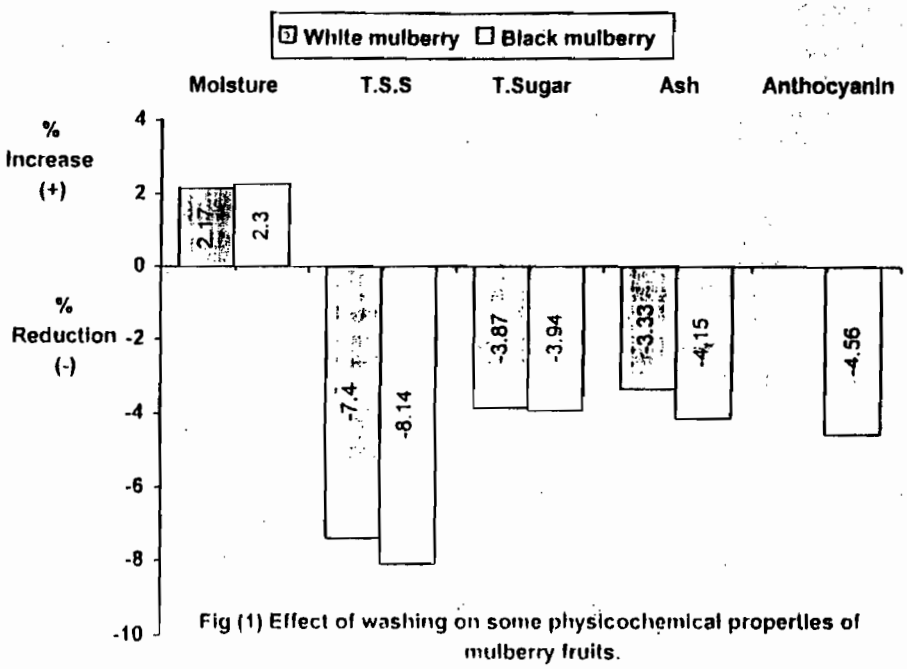

Fig (1) Effect of washing on some physicochemical properties of mulberry fruits.

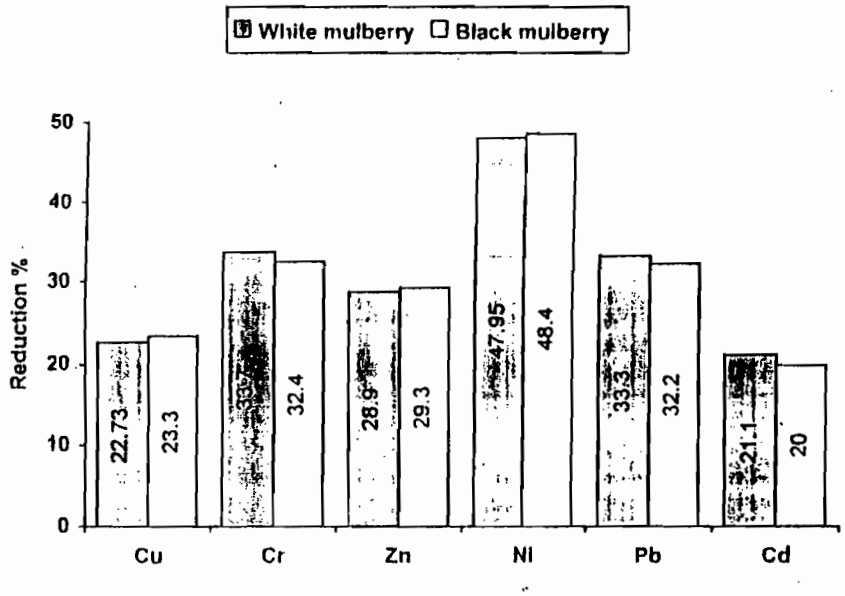

Fig (2) Effect of washing on heavy metal content of mulberry fruits.

CONCLUSION

The above mentioned results suggested the possibility of using sewage effluent in irrigating mulberry trees in sandy loam soils. Also soaking the fruits of these trees in water for 15 min.as washing method before consuming reduced more than 20% of their heavy metals contents.

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رى اشجار التوت بمياة الصرف المعالجة اوليا وتآثيرها على خواص الثمار وتراكم بعض المعادن الثقيلة فى مياة الرى والارض والثمار الخام والمغسولة

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الملخص العربى

أجريت هذه الدراسة على استخدام مياه الصرف المجمعة من مدينة برج العرب الجديدة بعد مـــرورها خلال ثلاث قنوات متتالية لأكسدة ما بها من مواد عضويه فى رى نوعين من أشجار الـــتوت القديمة (الأبيض والأسود) وتأثير ذلك على تراكم بعض المعادن الثقيلة (نحاس، زنك، منجنـــيز، رصاص، كادميوم وكروم) فى ثمار التوت الخام والمغسولة بالإضافه إلى تقدير تركيز هذه المعادن فى مياه الصرف قبل وبعد المعاملة وكذلك فى الأرض المزروعة فيها هذه الأشجار قبل وبعد الرى. وقد أظهرت النتائج أن مرور مياه الصرف لمدينه برج العرب خلال ثلاث قنوات متتالـــية أدى إلـــى خفـــض محتواها من الزنك والرصاص والنيكل ونسب بسيطة من النحاس والكـــروم والكادميوم. كما أدى استخدام هذه المياه فى الرى إلى رفع محتوى التربة المزروعة فـــيها هـــذه الأشجار وثمارها من المعادن الثقيلة خاصة ثمار شجر التوت الأسودوامكن ترتيب تركـــيز المعادن الثقيلة فى ثمار شجر التوت ترتيبا تنازليا كالآتى الزنك ثم النحاس ثم الكروم ثم الرصـــاص ثم الكادميوم. فيما عدا الرصاص فإن تركيز المعادن الثقيلة المقدرة فى ثمار أشجار التوت كان منخفضاً عن التركيزات المسموح بها من هذه المعادن فى المواصفات المصرية.كما أدى اســـتخدام مـــياه الصرف المعالجة فى الرى إلى خفض محتوى الثمار من الرطوبة والمواد الذائـــبة الكلـــية والســـكريات الكلـــية وزيـــادة فى محتوى الرماد وتأثير بسيط جداً على نسبه الانثوســـيانين. وأدى نقـــع الـــثمار لمدة ١٥ دقيقة على درجة حرارة الغرفة كطريقة من طرق الغســـيل إلى زيادة بسيطة فى محتواها من الرطوبة وخفض نسبه كل من المواد الذائبة الكلية والســـكريات الكلية والرماد والانثوسيانين. كما أدت عملية الغسيل أيضاً إلى خفض أكثر مـــن ٢٠% مـــن الـــنحاس، ٣٠% مـــن الكـــروم، ٢٨% من الزنك، ٤٧% من النيكل، ٣٠% من الرصاص، ٢٠% من الكادميوم الموجود فى الثمار.