

## THE EFFECT OF SALINITY ON RUBINIA TINCTORUM ROOT PIGMENTS AND THEIR RELATIONSHIP WITH SOME SOIL CHEMICAL CHARACTERISTICS

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### Abstract:-

*Rubinia tinctorum is a perennial herb plant. It is resistant to salinity and it has one of the strangest plant red colors which are used in the dye industry especially in the carpet industry. The purpose of this study is to investigate the effect of soil salinity on Rubinia tinctorum pigments, in the areas under cultivation of it. So, an experiment was done on the basis of a completely randomized design with three replications. Soil samples were taken from 6 regions under cultivation of Rubinia tinctorum in Yazd province. After determining the electrical conductivity of soil samples, they were classified into three groups of 4, 4-8, and 8-16 ds/m and the effect of salinity on desire parameters was investigated. Data analysis was done using SPSS and Excel software. The results showed that the amount of wool brightness ( $L^*$ ) decreased from 40 to 32.38 following soil salinity increase from the level of less than 4 to the level of 8-16 ds/m, but the amount of  $A^*$  (redness and greenness) and  $B^*$  (yellowness and blueness) parameters was increased with increasing soil salinity ( $P < 0.01$ ). The results indicated that there is a positive correlation between  $B^*$  parameter and soil acidity so that  $B^*$  parameter increases after increasing acidity ( $p < 0.01$ ). There is a positive correlation between electrical conductivity and  $A^*$  parameter while there is a negative correlation between  $L^*$  parameter and EC, Calcium and Magnesium, SAR ( $p < 0.01$ ). Totally, the quality of Rubinia tinctorum color was increased with increasing soil salinity. So, progressing toward economic production of this industrial and medicinal plant is possible by planting Rubinia tinctorum in saline soils which are inappropriate for most plants.*

**Keywords:-** Pigment, Rubinia tinctorum, Salinity, Soil chemical characteristics

## INTRODUCTION

A wide range of salinity is occurring in the soils of the world and every year this amount will increase (Koochaki and Mahalati, 1994). After drought stress, the salinity is the most important environmental stress that affects plants and greatly reduces the development of salt-sensitive plants (Homaeia, 2002). However, it is impossible that the problems of plants growing can be completely solved through proper irrigation management. Therefore, research on plants that are able to grow on saline soils is tangible.

*Rubia tinctorum* belongs to the herbaceous and perennial madder family, which grows as a self-pollinated plant in the Mediterranean from Spain to Asia Minor and also, North Africa and some other parts of Asia. It is extremely salt resistant plant by tolerance method and the soil salinity is well tolerated up to 0.3 percent (Dashtakian and Bahrani, 2007). Madder (*Rubia tinctorum*) is one of those plants that have multiple uses such as economic, medical, industrial and forage uses (Tavakkoli Saberi, 1989; Niebuhr, 1970; Chiej, 1984; Grieve, 1984; Bown, 1995).

The plant's roots produce Anthraquinone pigments (Baghestani Meybodi, 1990). Its color has been used for dyeing from 2000 BC and it is one of the most stable natural reddish-purple colors in the carpet industry (Javid Tabesh, 2000).

Some experts in the field of carpet industry believe that the natural vegetable dyes compared to the chemical dyes have a good fastness to light and washing and scrubbing (Mardaani Nezhad et al., 2002). The use of chemical dyes, in addition to adverse effects on humans, caused severe environmental pollution. While a lot of wild plants which are capable of coloring can be used and never faced with the waste and pollution risks. Persian carpets and other handicrafts, after oil is the most important exports of Iran. Thus, on one hand the production of their raw materials in the country due to the high quality of natural colors can greatly reduce the outflow of currency for purchasing the chemical colors, and on the other hand the Persian rug market will be thriving again (Jahanshahi Afshar, 1996). Several studies have been done in relation to that of the madder plant, including investigation of Abbasi et al., (2009). They showed that by increasing salinity the resistance of madder was increased and specific leaf weight and the number of branching stems, leaf area, root to shoot ratio, the central cylinder diameter to the root diameter, leaf weight of each plant, stem and root weight were decreased. Sepaskhah and Beirouti (2009) studied the effect of irrigation intervals and water salinity on growth of madder and concluded that the growth reduction per unit increase in soil salinity and irrigation water salinity for top growth are 2.0, 3.7 % per dS m<sup>-1</sup>, respectively. These values are 1.9, 3.1 % per dS m<sup>-1</sup>, respectively for root growth. Baghalian and Maghsodi (2010) have studied the Genetic diversity of Iranian madder (*Rubia tinctorum*) population based on agro- morphological traits, phytochemical content and RAPD markers. They have stated that the population had a significant effect on all evaluated traits. Also, correlation analysis showed that shoot weight had a significant positive correlation with root weight and dye content. Banakar and Khorsandi (2012) have stated that the madder plant is tolerant to salt in the vegetative stage and irrigation water with minimum salinity of 10 ds / m can be used for its cultivation. Madder plant is adapted to arid and desert climates and it is able to tolerate salinity. It is cultivated in some areas such as Yazd province using saline waters (Zargari, 1988).

In order to achieve sustainable development in the water, soil and Medicinal plant sectors, and with regard to different uses of the madder plant, especially in the dye industry and the importance of salinity, the objective of this study was set to investigate Pigments changes of *Rubia tinctorum* Root under different levels of soil salinity in the areas under cultivation of this plant.

## Materials and Methods

In order to investigate the effect of salinity on *Rubia tinctorum* pigments, the experiment was conducted in the form of complete randomized block design with three replicates in 6 areas under cultivation of this plant in Ardakan, Yazd. After sampling and determining the electrical conductivity of soils, the soil samples were classified into three groups of 4, 4-8, and 8-16 ds/m. After transferring of soil samples to the laboratory, parameters such as soil texture, electrical conductivity (EC), acidity (pH), sodium adsorption ratio (SAR) and ions of calcium, magnesium, sodium was measured in saturated extract. Also, to determine the pigment components of madder, the roots of the plants were harvested in autumn and after washing they have been dried and made into powder. 10 grams of powder was poured into 100 cc of distilled water and after boiling for an hour it was filtered. At this stage, 1 gr of wool in 30 cc of the solution was boiled at 70° C within an hour and it was washed and dried to remove excess dye. After preparation of the samples, the values of dye components including, L \* (indicates the brightness so that the greater amount, the brighter and less color absorption of wool), A \* (indicated the redness and the greenness of wool so that in the more positive value, the wool is more red, and in the more negative value, the wool is more green) and B \* (indicates the yellowness and the blueness of wool (it is not very effective in this test because the color is red)) were determined in the laboratory using spectrophotometer (model X-RITE).

The ANOVA method was used to analyze the data, and Duncan test was used to compare means. Statistical analysis of data was performed using SPSS and the graphs was drawn in EXCEL.

## Results

### Effect of different salinity treatments on wool color components

The results of the variance analysis of data related to the color component of wool dyed with madder root color showed that the salinity treatments had a significant effect on all three color components ( $p < 0.01$ ) (table 1).

**Table1. Variance analysis of the effect of salinity treatment on the color component of wool dyed with madder root color**

Sources of changes	Degrees of freedom	mean squares		
		L*	A*	B*
Salinity	2	53/15**	15/71*	18/10**
Error	6	10/1	0/811	1/67

**Significance at the level of 0.05: \* significance at the level of 0.01: \*\***

Mean comparison of data using Duncan's test showed that with increasing soil salinity, wool brightness (L \*) has decreased. The most amount of the L\* value was equal to 40 and it was observed in the salinity of less than 4 ds/m. There was no significant difference between it and the measured amount of 34.1 in the salinity of 4-8 ds/m. While there was a significant difference between it and the measured amount of 32.38 in the salinity of 8-16 ds/m.

The effect of salinity treatment on the brightness (L \*) of wool dyed with madder root. Different letters indicate the presence of a significant difference at the level of five percent in Duncan's test.

In the case of the redness and the greenness of wool, with increasing soil salinity, the component (A \*) was increased and there was a significant difference between treatments. The component in the salinity of less than ds/m is equal to 24.33 and it has its maximum value of 39 in the salinity of 8-16 ds/m. In other words, the red and green color components by increasing soil salinities to over 8 ds/m will be increased to 62.5 percent.

The effect of salinity treatment on the redness and the greenness of wool (○ A \*) dyed with madder root. Different letters indicate the presence of a significant difference at the level of five percent in Duncan's test.

The results obtained by mean comparison also showed that with increasing soil salinity in the 8-16 ds/m treatments, the yellowness and the blueness of wool (B \*) has increased significantly compared to treatments with electrical conductivity of less than 4 ds/m and 4-8 ds/m. The component in the salinity of 8-16 ds/m is equal to 26.17 and it shows an increase of 23% compared to the other two treatments.

The effect of salinity treatment on the yellowness and the blueness of wool (○ B \*) dyed with madder root. Different letters indicate the presence of a significant difference at the level of five percent in Duncan's test

**The correlation between some of the soil chemical characteristics of the areas under cultivation of madder and dye components of wool**

By examining correlations between some of the soil chemical characteristics of the area under cultivation of madder with each other and with dye components of wool such as L \*, A \* and B \*, it has been found that there is a correlation between A \* and EC, SAR ( $p < 0.01$ ), calcium, magnesium, sodium and soil pH ( $p < 0.05$ ). B \* is correlated with soil pH ( $p < 0.01$ ) and L \* is correlated with soil electrical conductivity (EC), SAR, calcium and magnesium ( $p < 0.01$ ). In Table 2, the correlation between these factors have been shown.

**Table 2. Correlation between some of the soil chemical characteristics of the areas under cultivation of madder with each other and with the amount of wool dye components**

	B*	A*	L*	EC	pH	Ca+Mg	Na	SAR
B*	1	0/825**	-0/303	0/562	-0/786**	0/464	0/494	0/519
A*		1	-0/707*	0/800**	0/768*	0/676*	0/786*	0/850**
L*			1	-0/920**	0/664	-0/864**	-0/945	-0/948**
EC				1	-0/503	0/330	0/494	0/854**
pH					1	-0/949**	-0/763*	-0/480
Ca+Mg						1	0/817**	0/352
Na							1	0/637
SAR								1

Shows the correlation between electrical conductivity, pH, sodium absorption ratio, calcium, magnesium and sodium content of the soil and color components of wool. Results show that there is a positive correlation between B\* and soil acidity (pH) ( $p < 0.01$ ) so that the B\* value has been increased as pH value increases. Also, there is a positive correlation between A\* and as electrical conductivity increases, the A\* value also increases ( $p < 0.01$ ), by increasing the electrical conductivity, sodium adsorption ratio, calcium, magnesium and sodium content, the wool brightness is reduced, it means that there is a negative correlation between these parameters and components L\* ( $p < 0.01$ ).

### Discussion and conclusion

Study results showed that with increasing salinity, the wool brightness (L\*) was decreased, in other words, it causes the increase in the pigments of the madder roots. The components have the maximum value in the EC of less than 4 ds/m. It had no significant difference with the measured amount in the EC of 4-8 ds/m while it had a significant difference with the measured amount in the EC of 8-16 ds/m. Decrease in the brightness level of wool indicates that the absorbed amount is more and the dyed wool seems to be darker. High negative correlation of L\* with soil chemical characteristics is a clear proof of this claim. Changes in the compounds which added to the dye, can cause the changes in the molecular structure of madder pigments. Montazer and Parvinzade (2002) have investigated the effect of ammonia on color change in wool dyed with natural colored materials such as madder and pointed to the effects of ammonia on the molecular structure of madder pigments.

Also, the results of the present study about the rate of the redness and the greenness of wool (A\*) showed that with increase in soil salinity to over 8 ds/m, A\* will be increased to 62.5 percent. The rate of the yellowness and the blueness of wool (B\*) also increased with increasing in soil salinity. Moreover, the results showed a high positive correlation between soil characteristics and A\* and B\* parameters.

Against all agricultural and horticultural crops which damaged by environmental stress, the active ingredients of medicinal and industrial plants will increase greatly in stress conditions (Rashidi, 2011). The general effect of salinity on the plants is reduction in the amount of their pigments, but depending on the species and the additive effects can be seen (Parida and Das, 2005). In some plants, the pigment amount increases with increasing in salinity. Destruction of the fine structure of chloroplasts and instability of pigment-protein complexes, chlorophyll degradation and changes in the content and composition of pigments such as carotenoids are the results of salinity. In general, according to the results increase in soil salinity can lead to improve the color quality of madder. Therefore, the use of appropriate salinity treatments could be a step towards economic production of industrial and medicinal plant and ultimately sustainable development could be achieved.

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