Comparative Evaluation of Grapevine Root Development in Different Media and Ex Vitro Screening of Plantlets for Drought Tolerance

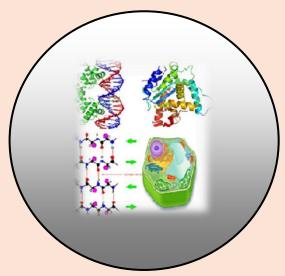
By

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RESEARCH PAPER

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Comparative Evaluation of Grapevine Root Development in Different Media and Ex Vitro Screening of Plantlets for Drought Tolerance Hamid Kheyrodin

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ABSTRACT

In this experimental research, in order to investigate the asexual reproduction (stem cuttings) of the grape variety by implementing the split plot statistical design or the split plot design in a completely random format in 7 replications in 10 cultivation beds in the greenhouse of Semnan University's Department of Desertology in pots Plasticization was done and some of the cuttings were directly planted in the form of a completely random design in greenhouse conditions. Cultivated cuttings were treated with indole butyric acid hormone with concentrations of zero, 1000, 5000 and 10000 parts per million (PPM). The data were analyzed using Duncan's test (5% level). The results obtained during a whole growing season show that multiplication by both methods is possible. Appropriate distribution of water is necessary in critical stages. These stages include: the stage of blooming and growth of buds, the stage of flowering and fruit formation, the stage of coloring until harvesting, the stage after harvesting. The method used in this research can be used to study the relationship between soil and plants in natural environments as an alternative to common methods that examine soil independently of plants. Also, based on the model of this research, it is possible to establish a systematic method for land classification with the purpose of afforestation in dry areas.

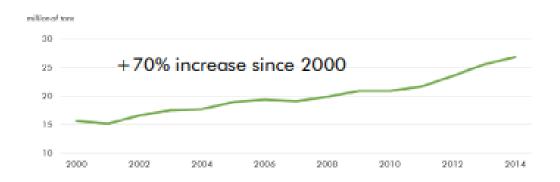
Keywords: Grapes, Ex Vitro Screening, Drought Stress, Afforestation and Hormones.

INTRODUCTION

Vine tree, which is also called hair tree, is a climbing shrub that climbs the wall and nearby trees. The stem of that node is in and has intermittent bugs. Grape leaves have five toothed lobes and dark green leaves. Its flowers are small, complex and appear as clusters. The fruit of the vine is fleshy and sweet and has different leaves.

According to Rostam al-Tawarikh book, the names of grape varieties in Iran at the time of Karim Khan Zand are: Shahani, Yaquti, Khalili, Askari, Rishbaba, Raisin Angoor, Fakhri, Sixty Arousan Siah Angoor, Alghi Siah, Mothghali, Razaqi, Shirazi, Nabati, Maderbeche. Grapes are heavenly fruit that contains vitamins A, B, and C. It also contains some magnesium, calcium, iron, phosphorus, potassium and albumin. Every person should eat grapes every day because it is an antiseptic fruit (Abera Jaleta and Mahbuba Sulaiman, 2019).

According to FAO statistics, in the mentioned period, France produced about 5 million 900 thousand tons, the United States with about 5 million 400 thousand tons, Turkey with 4 million 200 thousand tons, Turkey with 4 million 200 thousand tons, India with 3 million and 120 thousand tons, Chile with 2 million and 770 tons, Argentina with 2 million and 50 thousand tons and South Africa with 2 million tons of grapes are in the fourth to tenth place in the world. FAO also says: In the mentioned period, the Iranian country is in the 11th place in the world with a production figure (fig 1) of one million and 990 thousand tons. Also, the countries of "Uzbekistan, Egypt, Australia and Brazil" are in the next ranks (Table 1).



26.2 million tons: world consumption of table grapes

Figure 1. Show World Production of Grapes in 2000 – 2014.

Grapes are of great economic importance in the agricultural sector, and the products and products of this fruit, including raisins, vinegar, grape juice, grape juice, grape paste, currants, etc., speak for themselves. The vine or palm tree is one of the most sacred and oldest fruit trees known to man which can be propagated by two sexual methods (seeds) and asexual laying, grafting, leaf and stem cuttings, tissue culture. The capacity of a stem cutting for rooting by the reaction between the hereditary factors in the stem cells and factors such as plant growth regulators, the effect of buds and leaves, polarity, the amount of nutrients in the plant, the growth stage, the location of the stem on the plant, the type of tissue of the cutting and the time of taking the cutting are determined. The purpose of this project is to identify the best substrate for planting grape varieties and check the quantity, quality and shelf life of the rooted cuttings. It is intended to study the expansion properties of 90% perlite in comparison with cocopit and sand. Perlite is able to absorb 4 to 8 times its own weight in water and does not get flooded, as a result, it can store food inside itself after multiple irrigations and prevent its exit, which is important for the rooting and longevity of the cuttings.

On the other hand, the storage capacity of cocopit soil is very high compared to other types of cultivation bed; therefore, saving water consumption and also helps in the optimal growth of cuttings, and this water saving is important in the protection of new cuttings and roots against decay and soil fungal diseases, and its use is very economical. Wood mulch can also play an important role in soil improvement and rooting development of grape cuttings. In addition to enriching the soil, this useful substance loosens it, which can be used as beautiful mulch. The study of different types of cultivation bed and the effect of environmental conditions and stress is one of the necessities of the plan (Alizadeh Amin, 1357).

Water plays an important role in regulating different stages of plant growth and development. With regard to the problem of lack of water and fertilizer, which leads to the increase in the cost of production and rooting of grape cuttings, and the use of chemical fertilizers and hormones to improve soil conditions and increase the function of roots (and increase the function of genes in the plant) and increase the percentage of roots, this research aims to investigate and also evaluate the effect of substrate values and identify suitable substrate and identify the mutual effect of drought stress and substrate moisture, rooting hormone and identify the effects of quartz, sand and compost in Semnan city in order to reduce water consumption and increase product efficiency and identify important pests and diseases of different grape cultivars will be done. By carrying out the above plan, it is possible to recommend the chemical hormone fertilizer in the greenhouse and determine the residual performance of the roots in the substrate and identify the effects of drought and water stress (Basiri Abdullah, 1370).

MATERIAL AND METHODS

The chemical properties of culture medium and urease enzyme are determined and a number of other enzymes such as polyphenol oxidase are measured. Duncan's test will be used under SPSS conditions to compare the means and relationships between growth components and effects of drought stress and its effects (Khairaldin, 2013).

Examination of Hypotheses and Study Topics

Hypothesis: Do drought stress (*Asterus hydric*) and different cultivation medium have a significant effect on the production of grape cuttings and are they effective on the shelf life of the cuttings of grape cultivars (Haddadian Mahbubeh and Ismail Fallahi, 1400).

Cuttings test beds, cocopit, wood cover, cod, sand wind, sand and manure, windy sand, fertilizer and coarse sand, sand, clay, fertilizer (1:1:1), agricultural soil and wood pulp (1:1) after keeping the cuttings for several months in the greenhouse, the traits evaluated in this study include: the time of callus formation, the percentage of callus formation, the time of root formation, root length, the number of aberrant roots and the percentage of cuttings, and the characteristics and performance of photosynthesis such as leaf area. Production of cuttings and chlorophyll size of leaves, weight of wet cuttings and morphological traits are significantly affected by the type of substrate. Identification of grape cultivars tolerant to drought stress (Mahmoodzadeh 2019). It is one of the most important things in breeding and production programs, so the test was done in winter and its follow-up in summer and drought stress is also estimated separately in the second stage. In the second part, it is better to perform screening for drought stress only in a specific substrate.

That is, the rooted seedlings, for example, are taken out of the perlite bed and planted in the bed containing agricultural soil, and after the establishment of drought stress, they are applied to examine and compare their durability (Hosseini Abid Seyed Mohammad and Bijan Kaousi (2013).

In this experiment, we have several main goals

The study of the survival rate and residual weight of grape cuttings and the comparison of the longest length and vegetative volume of the root in 3 different cultivars after transfer to a new substrate of agricultural soil and the identification of the best substrate for root production in terms of fresh weight and dry weight of the root in the greenhouse.

Also, the measurement of the balance of all types of micronutrients and divalent metals in the rhizosphere of grape leaves and roots (Haidari Mohammad (2018).

Study and identification of genetic, physiological, phenotypic, morphological and biochemical characteristics of suitable grape cultivars in Semnan city.

Identifying the chemical and physical characteristics such as measuring the acidity and moisture percentage by weight of the growing media for grape cuttings and rooting conditions and the interactions between them.

Studying the cellular division of leaves by measuring leaf structure and chlorophyll a and b is another goal of the plan, knowing the carbon biomass of the substrates and the physiology of microorganisms (CO_2) and the best stress and irrigation treatment to root these cuttings also, the study of enzymology of the culture medium of grape cuttings and the appearance characteristics of long-lasting cuttings the study of root cytology and the study of factors stimulating root cells in grapes (Kheyrodin Hamid (2013) (fig 2).

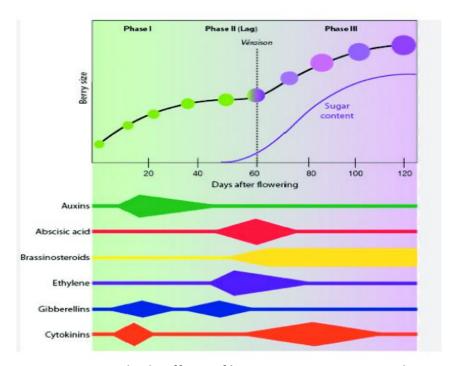


Figure 2. Multiple effects of hormones on grape growth.

RESULTS

The results presented in table 1. The traits of number and length of stem, number of leaves, number and length of roots, number and length of secondary roots were measured. Table 2 Variance analysis table to investigate the effects of cuttings and substrate on Grapes (Kheyrodin Hamid (2023).

Table 1. Shows differents Chemical and Physical Properties.

| 72 0.43 0.20 0.4 0.75 458 6.82 50 57.5 1.1 7 6 Coarse sand manure 44.5 0.33 0.23 0.3 0.7 2.1 6.7 7 23.5 0.5 2 1.5 A mixture of manure and we pulp 40 0.37 0.2 0.31 1 786 8.76 11 54 3.1 4 1 A mixture of sa and manure 5.5 0.3 0.2 0.27 1.72 300 6 100 72 2 4 9 A mixture of sa manure, clay a wood pulp | % water | <u>Carot</u> enoid | Chlo b narrometer | Chlo. A | %С | EC mho/cm | Ph 1/5 | Fresh weight | Dry weight | The length | The number | lleaf area | cutting bed |
|--|------------|-----------------------|----------------------|------------|------|--------------|-----------|-----------------|---------------|---------------|---------------|------------------|---------------------|
| 94 0.48 0.58 0.25 0.2 7.2 7 40 38 1 4 6 6 6 6 7 7 7 7 7 7 | | Nanometer | | nanometer | | | | | | | | Cm2 ⁾ | |
| 94 0.48 | | | | | | | | plant | plant | | branches | | |
| 94 0.48 0.58 0.25 0.2 7.2 7 40 38 1 4 6 clay 90 0.42 0.14 0.32 0.2 56.3 6.8 80 27 1.2 6 8 sand 57 0.40 0.23 0.4 1.96 546 7.3 76 41 3 7 9 manure 81 0.47 0.23 0.54 1.58 399 6.6 20 38 1.8 4 6 Wood cover 72 0.43 0.20 0.4 0.75 458 6.82 50 57.5 1.1 7 6 Coarse sand manure 44.5 0.33 0.23 0.3 0.7 2.1 6.7 7 23.5 0.5 2 1.5 A mixture of sand manure 5.5 0.3 0.2 0.31 1 786 8.76 11 54 3.1 4 1 A mixture of sand manure 5.5 0.3 0.2 0.27 1.72 300 6 100 72 2 4 9 A mixture of sand manure, clay a wood pulp | | | | | | | | | gr | petiole | | | |
| 90 0.42 0.14 0.32 0.2 56.3 6.8 80 27 1.2 6 8 sand 57 0.40 0.23 0.4 1.96 546 7.3 76 41 3 7 9 manure 81 0.47 0.23 0.54 1.58 399 6.6 20 38 1.8 4 6 Wood cover 72 0.43 0.20 0.4 0.75 458 6.82 50 57.5 1.1 7 6 Coarse sand manure 44.5 0.33 0.23 0.3 0.7 2.1 6.7 7 23.5 0.5 2 1.5 A mixture of manure and we pulp 40 0.37 0.2 0.31 1 786 8.76 11 54 3.1 4 1 A mixture of sa and manure 5.5 0.3 0.2 0.27 1.72 300 6 100 72 2 4 9 A mixture of sa manure, clay a wood pulp | | | | | | | | gr | | cm | | | |
| 57 0.40 0.23 0.4 1.96 546 7.3 76 41 3 7 9 manure 81 0.47 0.23 0.54 1.58 399 6.6 20 38 1.8 4 6 Wood cover 72 0.43 0.20 0.4 0.75 458 6.82 50 57.5 1.1 7 6 Coarse sand | 94 | 0.48 | 0.58 | 0.25 | 0.2 | 7.2 | 7 | 40 | 38 | 1 | 4 | 6 | clay |
| 81 0.47 0.23 0.54 1.58 399 6.6 20 38 1.8 4 6 Wood cover 72 0.43 0.20 0.4 0.75 458 6.82 50 57.5 1.1 7 6 Coarse sand manure 44.5 0.33 0.23 0.3 0.7 2.1 6.7 7 23.5 0.5 2 1.5 A mixture of manure and we pulp 40 0.37 0.2 0.31 1 786 8.76 11 54 3.1 4 1 A mixture of sa and manure 5.5 0.3 0.2 0.27 1.72 300 6 100 72 2 4 9 A mixture of sa manure, clay a wood pulp | 90 | 0.42 | 0.14 | 0.32 | 0.2 | 56.3 | 6.8 | 80 | 27 | 1.2 | 6 | 8 | sand |
| 72 0.43 0.20 0.4 0.75 458 6.82 50 57.5 1.1 7 6 Coarse sand manure 44.5 0.33 0.23 0.3 0.7 2.1 6.7 7 23.5 0.5 2 1.5 A mixture of manure and wo pulp 40 0.37 0.2 0.31 1 786 8.76 11 54 3.1 4 1 A mixture of sa and manure 5.5 0.3 0.2 0.27 1.72 300 6 100 72 2 4 9 A mixture of sa manure, clay a wood pulp | 57 | 0.40 | 0.23 | 0.4 | 1.96 | 546 | 7.3 | 76 | 41 | 3 | 7 | ٩ | manure |
| 44.5 0.33 0.23 0.3 0.7 2.1 6.7 7 23.5 0.5 2 1.5 A mixture of manure and wo pulp 40 0.37 0.2 0.31 1 786 8.76 11 54 3.1 4 1 A mixture of sa and manure 5.5 0.3 0.2 0.27 1.72 300 6 100 72 2 4 9 A mixture of sa manure, clay a wood pulp | 81 | 0.47 | 0.23 | 0.54 | 1.58 | 399 | 6.6 | 20 | 38 | 1.8 | 4 | 6 | Wood cover |
| 44.5 0.33 0.23 0.3 0.7 2.1 6.7 7 23.5 0.5 2 1.5 A mixture of manure and working pulp 40 0.37 0.2 0.31 1 786 8.76 11 54 3.1 4 1 A mixture of sa and manure of s | 72 | 0.43 | 0.20 | 0.4 | 0.75 | 458 | 6.82 | 50 | 57.5 | 1.1 | 7 | 6 | Coarse sand |
| manure and wo pulp 40 0.37 0.2 0.31 1 786 8.76 11 54 3.1 4 1 A mixture of sa and manure 5.5 0.3 0.2 0.27 1.72 300 6 100 72 2 4 9 A mixture off sa manure, clay a wood pulp | | | | | | | | | | | | | manure |
| pulp 40 0.37 0.2 0.31 1 786 8.76 11 54 3.1 4 1 A mixture of sa and man ure 5.5 0.3 0.2 0.27 1.72 300 6 100 72 2 4 9 A mixture off sa manure, clay a wood pulp | 44.5 | 0.33 | 0.23 | 0.3 | 0.7 | 2.1 | 6.7 | 7 | 23.5 | 0.5 | 2 | 1.5 | A mixture of |
| 40 0.37 0.2 0.31 1 786 8.76 11 54 3.1 4 1 A mixture of sa and man ure 5.5 0.3 0.2 0.27 1.72 300 6 100 72 2 4 9 A mixture off sa manure, clay a wood pulp | | | | | | | | | | | | | manure and wood |
| and manure 5.5 0.3 0.2 0.27 1.72 300 6 100 72 2 4 9 A mixture off sa manure, clay a wood pulp | | | | | | | | | | | | | |
| 5.5 0.3 0.2 0.27 1.72 300 6 100 72 2 4 9 A mixture off sa manure, clay a wood pulp | 40 | 0.37 | 0.2 | 0.31 | 1 | 786 | 8.76 | 11 | 54 | 3.1 | 4 | 1 | A mixture of sand |
| manure, clay a wood pulp | | | | | | | _ | | | _ | | _ | |
| wood pulp | 5.5 | 0.3 | 0.2 | 0.27 | 1.72 | 300 | 6 | 100 | 72 | 2 | 4 | 9 | |
| | | | | | | | | | | | | | |
| | 22 | 0.6 | 0.33 | 0.8 | 0.45 | 86 | 6.4 | 52 | 33 | 3 | 5 | 8 | A mixture of clay, |
| | 22 | 0.0 | 0.33 | 0.8 | 0.45 | 00 | 0.4 | 32 | 33 | 3 | 3 | ٥ | sand and fertilizer |
| 14.6 0.1 0.46 0.05 0.04 100 5.7 2 12.5 0.3 1 0.5 sand wind | 1/16 | 0.1 | 0.46 | 0.05 | 0.04 | 100 | 5.7 | 2 | 12.5 | 0.3 | 1 | ın 5 | |
| 14.0 0.1 0.40 0.00 0.04 100 3.7 2 12.3 0.3 1 0.3 said will | 14.0 | 0.1 | 0.40 | 0.03 | 0.04 | 100 | 5.7 | 2 | 12.3 | 0.5 | 1 | -0.5 | Sulla Willia |
| | | | | | | | | | | | | | |

Table 2. Variance analysis table to investigate the effects of cuttings and substrate on Grapes.

| Fs | Ms | Degrees of freedom | Sources of change |
|------|------|--------------------|--------------------------------|
| 5ns | 2 | 2 | Repetition |
| 11** | 7.6 | 2 | Grape variety |
| | 4 | 4 | The main factor error |
| | 3 | 8 | The main plot |
| 44** | 23.2 | 5 | Cutting bed |
| 77* | 22 | 10 | The mutual effect of substrate |
| | | | and grape varieties |
| | 3 | 15 | Sub-factor error |
| | | 30 | Subplot |
| | | 38 | All |

Ns. It is not significant at the level of 1 and 5%

DISCUSSION

In measuring the chemical properties of the used rooting media, pH and EC of the media were measured by saturated extract method. Chlorophylls and carotenoids were determined using a spectrophotometer.

^{*} It is significant at the 1% level

The percentage of organic matter and the amount of organic matter (OM) were also calculated and determined by drying the samples in an oven at 400 degrees.

Counting the seedlings during one month to 6 months from the beginning of the plan and the survival of the planted cuttings in two times (first in a plastic bag and pot and then direct planting in the soil and substrate in the greenhouse) by counting the number of leaves, leaf area, the length of sub-branches, the number of sub-branches, the length of the first leaf petiole, the weight of leaves in dry and wet state, the dry and wet weight of roots, the weight of the whole plant, chlorophyll a, chlorophyll b, carotenoids were recorded. Also, the physico-chemical properties of 8 substrates were determined and measured.

In this research, with a new perspective based on finding the most important soil factor affecting the growth of grape cuttings, while separating the units and the soil bed based on chemical, physical, biological, petrological and geomorphological characteristics, determining the morphology of their soil profile and some important laboratory characteristics of the sample were measured. Also, the freshness, area and yellowness of the leaves in the existing plant mass were scored by observation method.

Statistically, the highest mean number of leaves was observed in sand bed and the highest plant fresh weight was observed in pushal bed and mixed soil (sand, clay and manure). The amount of yellowing of the leaves produced by the cuttings was recorded after 6 months in the windy sand cultivation bed. The longest petiole length was obtained in sand and mixed beds (sand + fertilizer). The amount of photosynthesis and the area of leaves were obtained in manure and mixture (sand, clay, manure) and wood mulch. The highest percentage of carotenoid and amount of chlorophyll a was observed in the mixed substrate (manure, sand and clay).

The effect of substrate on the rooting of grape cuttings was significant. Also, the culture medium had significant effects on the photosynthesis and chlorophyll of hair leaves. According to the results obtained from the above research project, we briefly mention that Semnan province has a high variety of wild and cultivated grapes, especially in the cities of Shahrood and Bastam. Knowing their different traits from the point of view of botany, horticulture and investigating the genetic relationship of wild and cultivated grapes from the point of view of breeding programs and germplasm conservation in the future is of great importance which should be taken into account.

The degree of coordination of changes in the physical, chemical and morphological characteristics of the soil with changes in the average survival percentage of plants was analyzed in repetitions. The method used in this research can be used to study the relationship between soil and plants in natural environments as an alternative to common methods that examine soil independently of plants. Also, based on the model of this research, it is possible to establish a systematic method for land classification with the purpose of afforestation in dry areas. Depending on the type of grape, a pH of -5.5 to 7 is recommended for the soil. It is recommended that unbalanced acidity in terms of pH can be corrected with lime to increase pH and sulfur to decrease pH. Fertilizers suitable for strengthening the soil of grapevines are divided into two categories: fertilizers containing nitrogen, phosphorus, potassium, magnesium and calcium, and organic fertilizers. The branches that are selected and used for cutting must be fully mature, healthy, pest-free and woody. The diameter of the cutting at the upper end of the branch is between 8 and 12 mm. The length of the cutting should usually be between 25 and 45 cm.

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