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Physiological and Anatomical Study of the Effect of Aqueous Rice Husk Extract on *Triticum aestivum* Germination and Growth.

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ABSTRACT

Factorial experiment was conducted to study the effect of aqueous rice husk extract of different concentrations (10, 1, 0.1, 0.01)% and the control (Distilled Water) on germination and growth of wheat (*Triticum aestivum* L.) seedlings planted in pots (1kg). The results showed that aqueous rice husk extract at 10% concentration positively affect physiological characteristic like germination percentage, dry weight of leaves, shoot height whereas, no significant effect on the other growth parameters. This result was coinciding with the anatomical increament such as midrib width, lower midrib depth, adaxial epidermis cells number, mid vein width. While all rice husk concentrations caused a significant increase in the vascular bundle number and scleranchyma tissue width and roots.
Keywords: Rice husk , *Triticum aestivum*, anatomy, plant extract.

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INTRODUCTION

Wheat (*Triticum aestivum* L.) is the most important crop cultivated in Iraq. It is an annual plant belonging to the genus *Triticum* which includes common bread wheat (*Triticum aestivum* L.).Many attempts had used to improve growth and yield of economic plants by plant extracts (1-3).

Rice husk is one of the most widely available agricultural wastes in many rice producing countries around the world. Globally, approximately 600 million tons of rice paddy is produced each year. On average 20% of the rice paddy is husk, giving an annual total production of 120 million tones (4).Rice husk was used, mainly because of its effect on growth and biochemical parameters of plants (5) or it used to improve plant growth characters like rice(6)**,** water spinach(7) or its antioxidant activity (8).The major constituents of rice husk are cellulose, lignin and silica. (9) mentioned that rice husk biochar is highly alkaline (8.5-9.7) with a high ash content (36-44%) and high silicon, sodium, potassium, calcium and magnesium contents, it contains (KH_2PO_4), ($CaCO_3$), ($KCaCl_3$), ($KHCO_3$), ($K_2MgO_7P_2$), ($KMgPO_4 \cdot 6H_2O$), (KCl) and vaterite ($CaCO_3$).

These minerals are at least moderately soluble in water and are sources of plant nutrients when biochar is applied to soil and the silica, is a major constituent of rice husk biochar, it concentrated in outer epidermis cells including protuberances and hairs (trichomes) and also present in the inner epidermis. During growth, rice plants absorb silica and other minerals from the soil and accumulate it into their structures (10). In addition (11) concluded that rice husk composed of Na_2O, K_2O, Fe_2O_3, MnO, CaO, LOI. So, we use rice husk extract to improve the growth and developmental characters of wheat.

METHODOLOGY

Seed germination

Twenty five seeds were putting in a petridish containing filter paper Watmann no.1. supplemented with 15 ml of distilled water for control treatment or aqueous rice husk extract concentrations. Petridishes were grown in growth cabenet of 25 c°. Five replicates were used for each treatment.

Wheat planting

Ten seeds of *Triticum aestivum* L. were planted in 1 kg pots containing sand soil. Three pots were used to each treatments as replicates. Seeds of *Triticum aestivum* were watered with 100 ml of tap water every morning. Germination percentage, plant height, dry weight of shoot and root, and root length were measured after 15 days of planting.

Preparation of water extract solutions

Cold water extract were prepared by weighting 10 grams of rice husk in 100 ml of distilled water to get 10% concentration. Then the mixture was grinding 10 minutes and filtered by three layers of cheesecloth. This solution was used as a stock to prepare (1%,0.1%,0.01%) concentrations .

Primer detection of active compounds:

Procedure of (12) was used to investigate the presence of phenols, alkaloids, glycosides, terpenes, tanines, resins and flavones in rice husk extract. The presence of phenols was detected.

Slides preparation:-

A cross sections of *Triticum aestivum* L. leaves were made by hand sectioning using preserved in 70% alcohol (13) having blades in a transversal plan and drawn by camera lucida, then pictures were took to it.

Statistical Analysis

The data were analyzed and the means were compared according to Least Significant Difference (LSD 0.05) , (14) by using SPSS program.

RESULTS

According to (Fig.1), treatment of wheat seeds by rice husk 10% concentration caused a significant increase in germination percentage about (58.6%) compared with the control treatment(Distilled Water).While, concentrations 10%, 0.1%, and 0.01% caused a significant increase about(56%,37%,56%) respectively in plant height (Fig.2) concurrent with a significant decrease in root length (Fig.3).

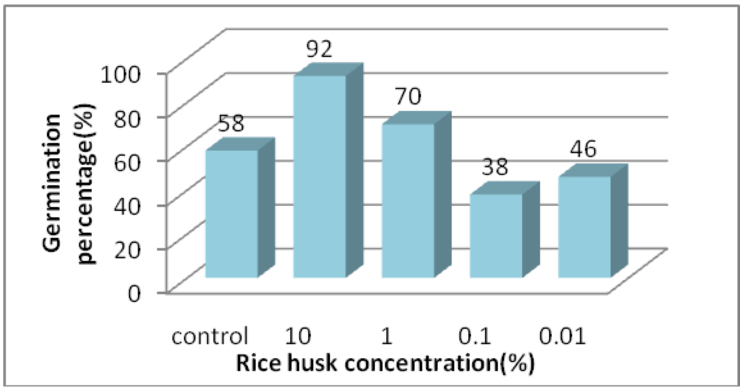

Fig (1):-The effect of rice husk concentrations on germination percentage. L.S.D.=14.6

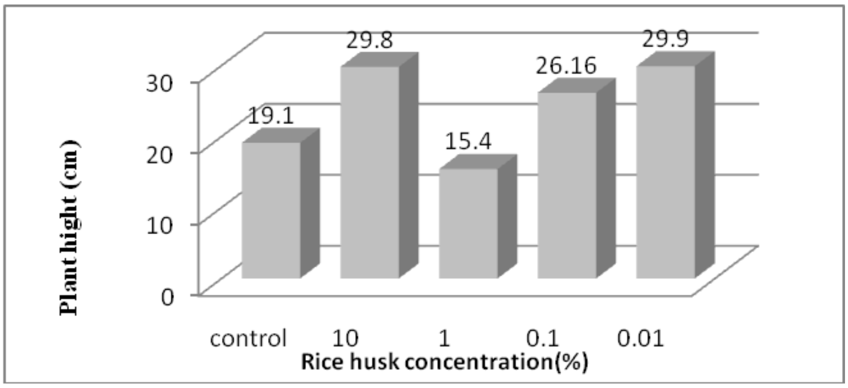

Fig (2):-The effect of rice husk concentrations on plant length. L.S.D.=3.8

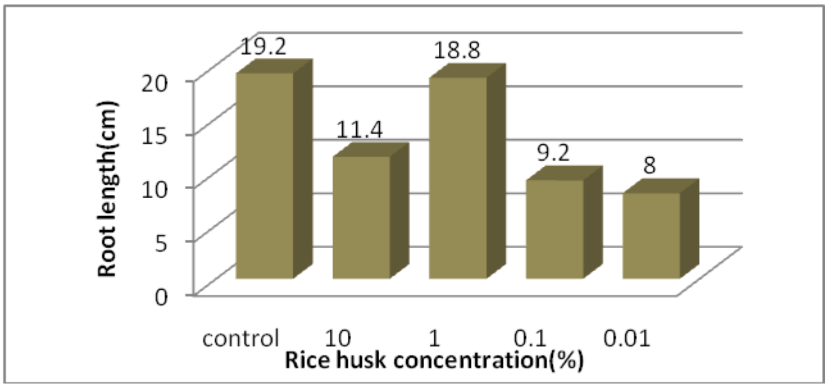

Fig (3):-The effect of rice husk concentrations on root length. L.S.D.=1.5

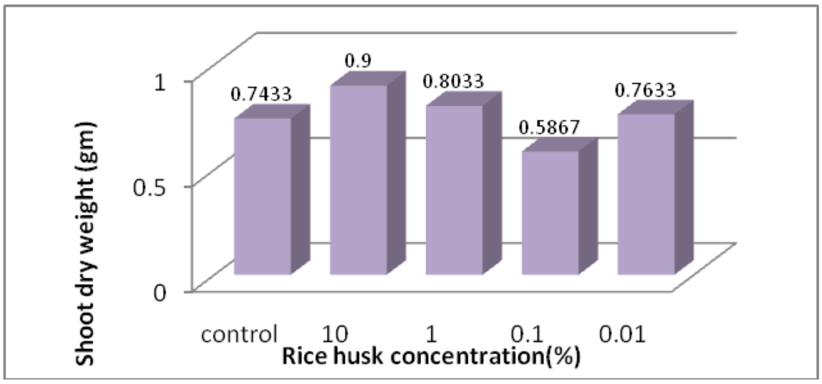

Fig (4):-The effect of rice husk concentrations on shoot dry weight. L.S.D.=0.3

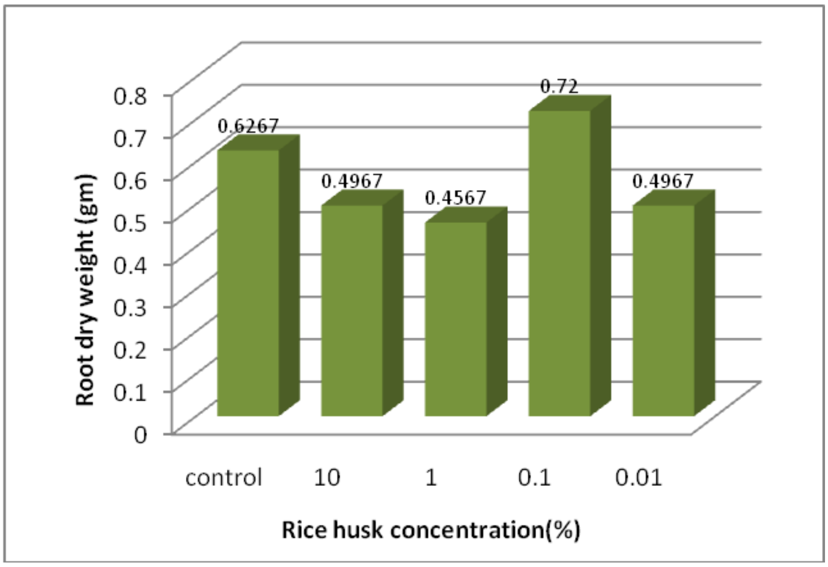

Fig (5):-The effect of rice husk concentrations on root dry weight. L.S.D.=0.2

Fig.4,5 showed that no significancy in the dry weight both in shoot and root of the wheat seedlings.

In addition, the research indicates a lot of anatomical changes in the midrib region including a gradual significant decrease in the midrib thickness, upper midrib depth, midvein, adaxial epidermis cells area and midrib scleranchyma thickness at lower epidermis Table (1) and Fig(8).While, the other parameters are increased significantly including midrib width, lower midrib depth, adaxial epidermis cells number, midvein especially at 10% rice huck extract concentration.

Table (1):-The effect of rice husk concentrations on anatomical characters of the midrib of the wheat.

Measurements / Treatments	Midrib thicknes (µ)	Midrib width (µ)	upper midrib depth (µ)	lower midrib depth (µ)	Lower epidermal cells No. of the midrib	adaxial epidermis cells number	midvein (µ)	midrib scleranchyma thickness at lower epidermis (µ)	adaxial epidermis cells area (μ^2)
Control	107.5	75	37.5	30	30	15	6.5	25	19.375
10%	100	87.5	28.125	40.625	32.5	25	7.175	18.75	17.25
1%	75	75	18.75	31.25	37.5	35	4.5	13.75	7
0.1%	80	100	31.25	28.125	30	32.5	3.75	12.5	6.6667

0.01%	50	62.5	21.875	14.375	22.5	35	5.75	21.875	8.95
L.S.D	3.7	3.7	1.7	1.7	1.7	1.7	0.3	2.4	0.2

Table (2):-The effect of rice husk concentrations on anatomical characters of the vascular bundle of the wheat.

Measurements / Treatments	No.vascular bundle	lower epidermis cells number of V.B	Upper epidermis cells number of V.B	V.B thickness (μ)	sclerenchyma tissue width (μ)	sclerenchyma tissue thickness (μ)
control	22	37.5	30	60	3.12	6.25
10%	37.5	15	20	50.75	12.5	6.25
1%	35	23.25	29	51.25	12.5	6.25
0.1%	25	28.25	31.5	72.5	9.375	6.25
0.01%	32.5	32.5	32.5	45	9.375	6.25
L.S.D	3.7	2.9	2.9	2.9	2.3	0.4

All rice husk concentrations caused a significant increase in the vascular bundle number and scleranchyma tissue width, Table (2) and Fig(6). Wherease, lower epidermis cells number and vascular bundle thickness were decreased significantly. Upper epidermis cells number was decreased at 10% extract concentration,while the other concentrations had no significant effect on it compared with the control.

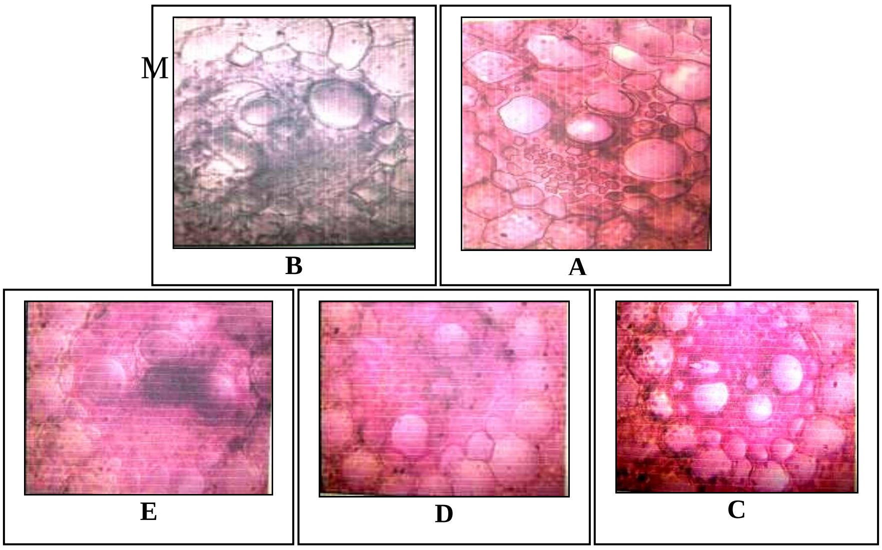

Figure (6):- Effect of Rice husk water extract on the midrib of leaves of *Triticum aestivum* L., (A) control, (B) 10%, (C) 1%, (D) 0.1%, (E) 0.01% . M=midvein, CL=chloranchyma tissue.

In the other hand, cuticle thickness was decreased with rice husk treatments especially 10% concentration. Indeed, sunken stomata was observed in 0.1% and 0.01% concentrations.

DISCUSSION

Several scientists conducted on plant extracts to improve plant growth. They used different plant parts like leaves (15), rice husk (8), bark (16) or rhizomes (17) because of their active compounds. In this study we used rice husk extract to improve germination and growth of *Triticum aestivum* L.

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In Fig.(1) germination percentage was increased significantly at 10% concentration while the low concentrations have no effect on seed germination. This result may be related to the increase of extract phenolic compound content as demonstrated in primary detection of active compounds of this work which may be led to increase its antioxidant activity. (8) mentioned that rice husk extract exhibited antioxidant activities and their antioxidant abilities coincidentally increased with the total phenolic content. In contrast, the antioxidant activity of rice may be the reason which caused the increase in shoot hight and shoot dry weight about 21% , 10%, and 8% in concentration 10% , 1% , and 0.01% respectively, Fig (2 and 4). An opposite results were found in root system.

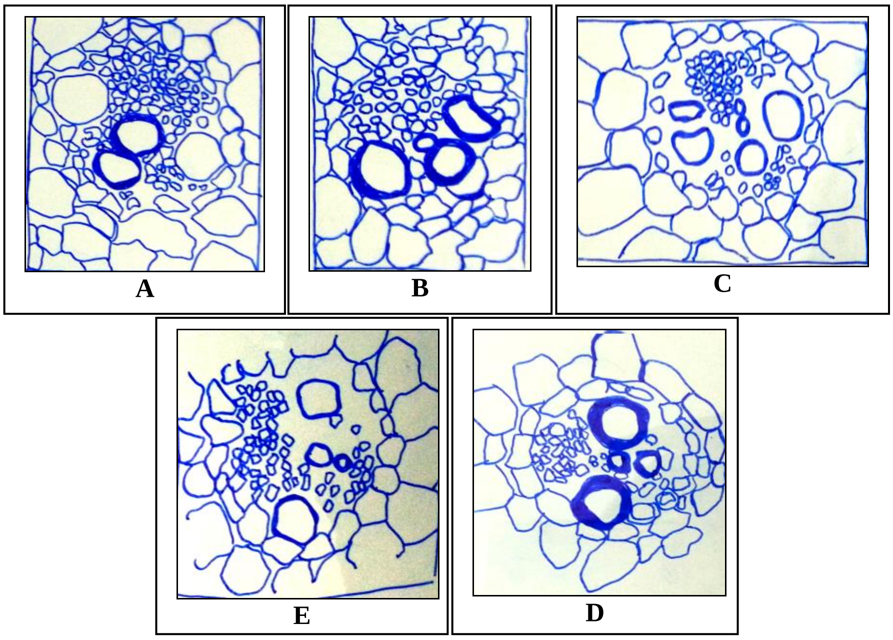


Figure (7):- Effect of Rice husk water extract on the midrib of leaves of *Triticum aestivum* L. draw by camera lucida, (A) control, (B) 10%, (C) 1%, (D) 0.1%, (E) 0.01% . M=midvein, CL=chloranchyma tissue.

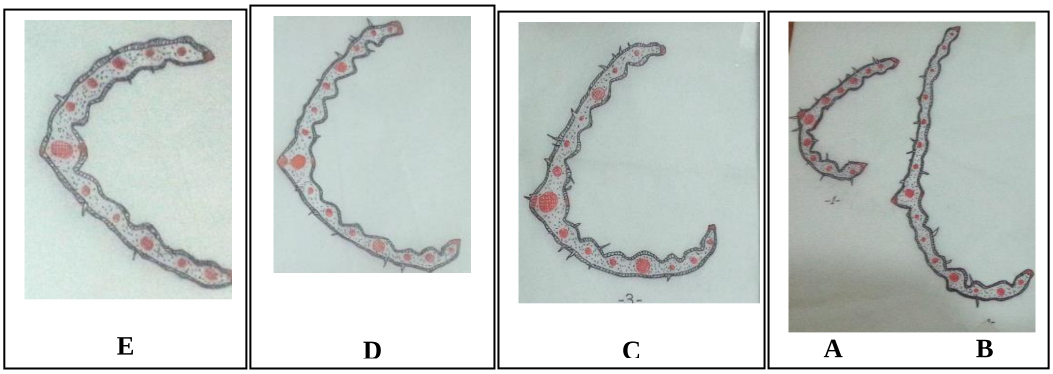
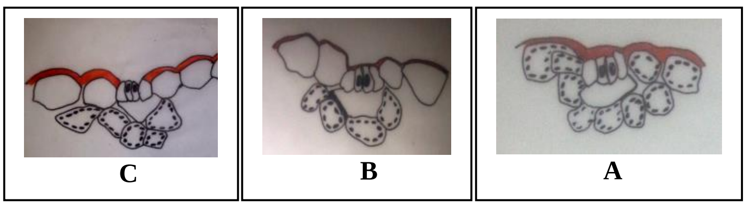


Figure (8): Effect of Rice husk water extract concentrations on anatomical characteristic of *Triticum aestivum* L. Cross section of leaves draw by camera lucida. (A) control, (B) 10%, (C) 1%, (D) 0.1%, (E) 0.01%.



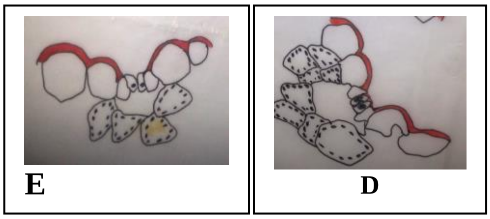


Figure (9): Effect of Rice husk water extract concentrations on anatomical characteristic of *Triticum aestivum* L. Cross section of stomata draw by camera lucida. (A) control, (B) 10%, (C) 1%, (D) 0.1%, (E) 0.01%.

The morphological changes were a result of anatomical changes occurs after rice husk treatments. (18) showed that the environmental and nutritional conditions during plant growth can influence cell differentiation, resulting in anatomical and physiological adaptations. So, the increase in shoot hight and shoot dry weight especially at 10% concentration Fig (2 and 4) was synchronized with the increase in midrib width, lower midrib depth, midvein Table (1) and Fig(7) and vascular bundle number Table (2) and Fig(8). Either responses should increase the efficiency of transport and the metabolite exchange capacity between wheat and other organs (19) causing adaxial epidermis cells number increase Table (1) as a result to the increase cytokinin transported from roots to the leaves which in turn causing cell number increament. This suggestion is compatible with the finding of (20) who showed that the primary functions of AHK genes, and those of endogenous cytokinins, are triggering of the cell division and maintenance of the meristematic competence of cells to prevent subsequent differentiation until a sufficient number of cells has accumulated during organogenesis. As well as the increase in mid vein diameter Table (1) may cause an increase leaf hydrolic conductivity which in turn increase water and minerals transport. This suggestion is compatible with (21). In addition the increase in scleranchyma tissues width Table (2) gives the plant more rigidity to overcome the adverse environmental conditions. This result is compatible with the finding of (22) who proved that toughness is generally correlated with traits such as the proportion of vascular tissue, fibre or sclerenchyma, and tissue density. Besides that we observed a sunked stomata in the leaves treated with 10%, 0.1% and 0.01% concentrations Fig(9) .This is compatible with the finding of (19) who demonstrated that sunked stomata formed in wheat plants treated with triadimefon was a result to the increase in epidermal cells depth. This morphological modification could result from the change in the direction of cellulose mocrofibril deposition in cell walls (23) and may be this was a kind of plant adaptation which control water loss allows plants to occupy habitats with fluctuating environmental conditions and so it can be predicted that stomata must be important contributors to speciation and evolutionary change (24). The decrease in cuticle layer in plant treated with 10% concentration Fig (9) was also observed. It may be refers to the presence of sufficient water content within the plant which scynocrinized with the increase in midvein diameter Table (1) and *et al.*, 2006 decrease cuticle layer. This result is compatible with the finding of (25-27) who showed that plant water deficit increases epicuticur wax load which related to low ABA content (28). The decrease in cuticle layer should increase transpiration. In the opposite manner lower rate of transpiration may be caused by sunked stomata (19). So, a balance in water contain was occurred during rice husk treatments resulting in more viable wheat plants. From this research we concluded that rice husk extract at 10% concentration improved the physiological and anatomical characteristics of wheat seedlings.

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