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***Original article***

# Effects of humic acid foliar application on millet (*Panicum miliaceum* L.) yield and some of the biochemical and physiological parameters under drought stress condition in Ramjerd region of Fars

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## *Extended abstract*

### Introduction
Water shortage is the most important non-biological limiting factor for achieving the potential of crop yield. The millet is one of the drought-tolerant species that the study of its specific traits can help to identify the effective mechanisms of drought coping. Humic acid contains abundant elements that improve soil fertility and increase the availability of nutrients to plants, thus affect their growth and yield, but in the millet, humic acid function on increasing tolerance to drought is unclear.

### Materials and methods
To evaluate the effect of humic acid foliar application on yield and yield components of millet under drought stress conditions, the experiment conducted in 2015 using a split-plot based on randomized complete block design (RCBD) with three replications. Irrigation regime considered as the main factor including two levels of normal irrigation and drought stress (irrigation cut off) at 50% flowering stage and sub-factors also included the foliar application of humic acid at five levels (1, 3, 5, 7% humic acid and pure water as control) in the Ramjeard region of Fars province.

### Results and discussion
The results of this study showed that drought stress caused a significant decrease in the most morphological and physiological traits such as leaves chlorophyll content, the number of grain per spikes, 1000-grain weight, grain yield and harvest index but also humic acid has greatly controlled the effects of drought stress, then, ultimately the performance of treatments with humic acid was more than the control treatment. The remarkable point in this study was the process of changes in biochemical traits, that by increasing the percentage of humic acid, biochemical traits such as proline, superoxide dismutase and peroxidase enzyme decreased, these changes showing a positive effect of humic acid in reducing the effects of drought stress. As a result, the use of humic acid foliar application increased the leaves chlorophyll content, the number of spikes $m^{-2}$, 1000-grain weight as well as grain yield, so that changes in 7% humic acid treatment were higher compared to the other treatments. The highest proline content was related to foliar application with water (control) under drought stress conditions, while the lowest proline content observed in 7% humic acid foliar application under normal irrigation conditions. The amount of superoxide dismutase under normal irrigation conditions was significantly lower than all drought stress treatments. Under drought stress conditions the highest rate was observed in foliar

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application with water (control), which could be confirmed by reducing the amount of superoxide dismutase as a result of using humic acid under stress conditions. The highest amount of peroxidase enzyme was observed in foliar application with water (control) under drought stress conditions. Generally, peroxidase content was significantly higher in drought stress treatments than in normal irrigation conditions. Under drought stress conditions, the highest chlorophyll a content was observed in the foliar application of 7% humic acid, similarly, the highest amount of chlorophyll b was observed in the foliar application of 7% humic acid, while there was no significant difference with foliar application of 5%. In this experiment, the number of grain per spikes affected by humic acid application and the interaction of drought stress and humic acid. Humic acid compensated for the negative effect of drought stress on the number of grain per spikes. Among drought stress treatments, using 7% humic acid increased the number of grain per spikes by 12% compared to control treatment. The comparison of two irrigation regimes showed the highest and lowest yield observation under normal irrigation and drought stress conditions, respectively. The yield under drought stress decreased by 56% compared to normal irrigation. Generally, we can conclude that even though drought stress significantly decreased the number of grains per spikes, 1000-grain weight and yield, but the variation trend of biochemical traits in drought stress treatments was significantly decreased proline, superoxide dismutase and peroxidase with increasing of the humic acid percentage, while the amount of chlorophyll a and chlorophyll b increased.

## Conclusions

It seems that humic acid can improve the morphological and physiological characteristics of the conventional millet in drought stress conditions. It seems that humic acid can play a role in improving biochemical and physiological characteristics of conventional millet under drought stress conditions, so that by reducing the effects of drought stress increased chlorophyll and ultimately improved the photosynthesis rate. Therefore, the yield and yield components of humic acid treatments were higher than foliar application with water (control).

***Keywords*:** Chlorophyll, Peroxidase, Proline, Superoxide dismutase

**Table 1. Some physicochemical properties of the soil at 0-30 cm depth**

| | |
|---|---|
| **Electrical conductivity (dS/m)** | 2.55 |
| **pH** | 7.9 |
| **Bulk density (g/cm$^3$)** | 1.55 |
| **Soil water content before planting** | 19.57 |
| **Saturated water content (percent)** | 48 |
| **Soil texture** | Clay loam |
| **Phosphorus (percent)** | 12 |
| **Available nitrogen (percent)** | 10.11 |
| **Potassium (percent)** | 10.2 |

**Table 2. Analysis of variance of biochemical and physiological traits measured in conventional millet under drought stress conditions and humic acid foliar**

| S.O.V | df | Proline | Superoxide dismutase | Peroxidase | Chlorophyll a | Chlorophyll b |
|---|---|---|---|---|---|---|
| **Replication** | 2 | 44.43 | 0.0003 | 0.0000312 | 0.40 | 0.20 |
| **Irrigation (I)** | 1 | 8200.53** | 0.4154** | 0.023460** | 210.78* | 6.98* |
| **Error a** | 2 | 35.03 | 0.0020 | 0.0000042 | 2.18 | 0.15 |
| **Humic Acid foliar(H)** | 4 | 1141.88** | 0.0046** | 0.0006133** | 53.01** | 2.17** |
| **I * H** | 4 | 829.78** | 0.0086** | 0.0013245** | 4.70** | 0.31ns |
| **Error b** | 16 | 10.43 | 0.00097 | 0.0000281 | 0.61 | 0.2 |
| **C.V (%)** | | *13.38* | *2.45* | *2.60* | *7.42* | *13.52* |

* and ** and ns, indicate significantly different at 5% and 1% probability and are not significant, respectively

**Table 3. Comparison of the mean interaction effects of biochemical and physiological traits measured in conventional millet under drought stress conditions and humic acid foliar**

| Irrigation regimes | Humic Acid foliar application | Proline | Superoxide dismutase | Peroxidase | Chlorophyll a | Chlorophyll b |
|---|---|---|---|---|---|---|
| | | μg g-1 | ---mg g-1--- | | | |
| **Drought stress** | **Control** | 85.67a | 1.40 a | 0.24 a | 8.26 e | 1.31 e |
| | **1%** | 80.00a | 1.24 c | 0.21 c | 8.31 e | 1.51 e |
| | **3%** | 49.00 b | 1.27 bc | 0.21 c | 10.45 d | 2.06 de |
| | **5%** | 40.33 c | 1.31 b | 0.19 d | 13.91 c | 2.56 cd |
| | **7%** | 34.33 cd | 1.24 c | 0.22 b | 16.56 b | 3.12 bc |
| **Normal irrigation** | **Control** | 29.00 de | 1.03 d | 0.17 e | 13.51 c | 2.63 cd |
| | **1%** | 28.03de | 1.06 d | 0.13 g | 15.54 b | 3.16 bc |
| | **3%** | 25.67 de | 1.07 d | 0.18 e | 16.59 b | 3.03 bc |
| | **5%** | 23.33 e | 1.07 d | 0.16 f | 19.33 a | 3.93 ab |
| | **7%** | 20.67 de | 1.06 d | 0.15 f | 19.02 a | 4.23 a |

Means of each column followed by similar letters based on Duncan test at 5% are not significantly different

**Table 4. Analysis of variance of yield and yield components in conventional millet under drought stress conditions and humic acid foliar application**

| Source of variation | df | Number of spikes m-2 | Number of grain per spike | 1000-Grain weight | Grain yield | Harvest Index |
|---|---|---|---|---|---|---|
| **Replication** | **2** | 49.23 | 34.03 | 1.37 | 508.26 | 20.41 |
| **Irrigation (I)** | **1** | 3413.33** | 16614.67** | 15.93** | 15272.67** | 248.32** |
| **Error a** | **2** | 10.43 | 4.23 | 0.013 | 24.32 | 0.1552 |
| **Humic Acid foliar application (H)** | **4** | 377.42** | 478.25** | 0.57** | 488.28** | 11.69ns |
| **I * H** | **4** | 33.42* | 148.78** | 0.23* | 97.30** | 0.5974ns |
| **Error b** | **16** | 8.06 | 19.82 | 0.07 | 17.89 | 0.9783 |
| **C.V (%)** | | *3.87* | *2.74* | *8.91* | *13.61* | *16.68* |

* and ** and ns, indicate significantly different at 5% and 1% probability and are not significant, respectively

**Table 5. Comparison of the mean interaction effects of yield and yield components in conventional millet under drought stress conditions and humic acid foliar**

| Irrigation | Humic Acid foliar application | Number of spikes $m^{-2}$ | Number of grain per spike | 1000-Grain weight | Grain yield | Harvest Index |
|---|---|---|---|---|---|---|
| | | | | g | g $m^{-2}$ | % |
| Drought stress | Control | 97.00 $^{f}$ | 131.00 $^{g}$ | 1.71 $^{d}$ | 28.59 $^{e}$ | 6.67 $^{e}$ |
| | 1% | 102.67 $^{f}$ | 134.33 $^{g}$ | 2.17 $^{cd}$ | 31.70 $^{de}$ | 7.18 $^{de}$ |
| | 3% | 111.67 $^{e}$ | 137.33 $^{fg}$ | 2.38 $^{c}$ | 38.01 $^{de}$ | 9.08 $^{cde}$ |
| | 5% | 118.00 $^{de}$ | 141.67 $^{ef}$ | 2.07 $^{cd}$ | 37.18 $^{de}$ | 7.53 $^{de}$ |
| | 7% | 115.67 $^{de}$ | 146.33 $^{e}$ | 2.42 $^{c}$ | 40.15 $^{d}$ | 10.34 $^{bcd}$ |
| Normal irrigation | Control | 120.67 $^{cd}$ | 164.33 $^{d}$ | 3.26 $^{b}$ | 62.91 $^{c}$ | 12.01 $^{abc}$ |
| | 1% | 126.67 $^{bc}$ | 183.33 $^{c}$ | 3.33 $^{b}$ | 74.08 $^{bc}$ | 13.62 $^{ab}$ |
| | 3% | 131.00 $^{b}$ | 191.33 $^{b}$ | 3.32 $^{b}$ | 83.15 $^{ab}$ | 15.19 $^{a}$ |
| | 5% | 132.00 $^{b}$ | 199.67 $^{a}$ | 3.90 $^{a}$ | 84.39 $^{ab}$ | 13.52 $^{ab}$ |
| | 7% | 141.33 $^{a}$ | 187.33 $^{bc}$ | 4.22 $^{a}$ | 96.17 $^{a}$ | 15.23 $^{a}$ |

Means of each column followed by similar letters based on Duncan test at 5% are not significantly different