



ISSN NO. 2320-5407

Journal homepage: <http://www.journalijar.com>
Journal DOI: [10.21474/IJAR01](https://doi.org/10.21474/IJAR01)

INTERNATIONAL JOURNAL
OF ADVANCED RESEARCH

RESEARCH ARTICLE

Effects of water stress on two genotypes of Durum wheat (*Triticum durum* Desf.) and Common wheat (*Triticum aestivum* L.) inoculated with Frankia CCI3 Azospirillum brasilense and Mycorrhiza

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Manuscript Info

Manuscript History:

Received: 19 March 2016
Final Accepted: 19 April 2016
Published Online: May 2016

Key words:

wheat, inoculation, Azospirillum, Frankia, mycorrhiza, water stress.

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Abstract

This study aims to evaluate the response of common and durum wheat to inoculation by micro-symbionts. Two common and two durum wheat genotypes were inoculated by three inoculums Frankia CCI3 (IF), Azospirillum brasilense (IA) and Mycorrhiza (IM) in presence or in absence of water stress in a greenhouse assai. After the heading stage plants were collected. The parameters of growths and the dry weight of plants and roots were measured as well as SPAD index and nitrogen dosage. The obtained results are significant. They showed that the inoculation with the IM treatment enhanced leaves, ear and roots length. IA treatment has a favorable effect on the nitrogen accumulated in leaves of inoculated plants under control and water stress condition.

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Introduction:-

In Algeria, Cereals are an important part of human and animal food supplies (Karakas et al., 2011). Wheat culture occupies one million hectares; the major parts of surfaces are located in semi dry regions. Those regions are submitted to rough climatic constraints: winter cold, irregular pluviometry, hot and dry wind accompanied with strong temperatures at the end of plant cycle (Boufenar et al., 2006). This situation affects seriously the cereal efficiency (on average of 10q / Ha). Water stress induced disturbance at the plant physiological and metabolic level. It limits plant growth and cereal productivity (Kara et al., 2012).

Algeria is one of the biggest world importer countries of grain (Chikihi et al., 2013). The imports reached 7.4million tons in 2011 and 6.9million tons in 2012 (Touchan, 2016). In a context of sustainable agriculture, the inoculation of culture by beneficial bacteria (Frankia, Azospirillum and spore endomycorrhizae) represents an interesting alternative. When micro-symbionts are applied: on seeds, on plant surface, or on the ground, they colonize the rhizosphere or penetrate inside the plant cells and colonize that space. They also have a good impact on plant growth. The micro-symbionts can act directly on the plant via nitrogen fixation, phytohormone synthesis, modulation of hormonal balance and by desamination of éthylène 1-aminocyclopropane-1-carboxylate (ACC) precursor. These properties can improve the strain and the ramification of root system, what favors the absorption of water and minerals in host plants, particularly: wheat, barley, corn and rice (Vessey, 2003). This work intends to analyze the physiological answer of four inoculated varieties (by micro-symbionts) of durum and common wheat under water stress. And that by focusing in particular on characterizing the most tolerant varieties based on physiological (chlorophyll content), morphological (length of leaves, ear and roots) and biochemical approaches (dosage of nitrogen), as well as plant and roots dry weight.

Material and Methods:-

Plant material:-

The plant material used in our study is constituted of: Two common wheat varieties: Hidhab, Ain Abid and two durum wheat varieties: Boussellem, Waha,. Supplied by ITGC (Institut Technique des Grandes Cultures) of El Khroub/Constantine, Algeria).

Table 1:- Origin of the varieties of common wheat and durum wheat .

	Species	Pedigree	Origin
Hidhab	Common wheat	HD1220/3*KAL//NAC	CIMMYT Mexico selection to Elkhroub
Ain Abid	Common Wheat	AS81189A	CIMMYT/SPAIN selection to Elkhroub
Boussellem	durum wheat	Heider/marten//Huevo of oro	CIMMYT/SELECTION to Sétif
Waha	durum wheat	Plc/Ruff//Gta/3/Rte crossing	CIMMYT selection ITGC Elkhroub

Bacterial Material:-

Pure cultures of bacteria were prepared using Frankia CCI3 according to the method described by Diem (1983). The stumps of *Azospirillum brasilense* were firstly grown in NFB N-free semi solid medium (Baldani and Dobereiner, 1980) for 48h at 30°C; then a loop full of each culture was transferred separately to 100 ml NFB liquid medium without blue bromothymol, supplemented with 1% NH₄Cl (w/v) and incubated at 30°C, without shaking for 72h. After incubation, the cells were centrifuged for 10 min and washed twice with phosphate buffer pH 7.0 in order to eliminate any residue of culture. And we proceeded to mycorrhiza spores isolation from soil by wet sieving and sedimentation method "Wet sieving and Decanting."

Implementation of the essay:-

The essay was conducted under a greenhouse. Seeds were disinfected by emerging them 30 seconds in ethanol 70 ° and 20 minutes in sodium hypochlorite at 10 %, and then rinsed in sterile distilled water. After sterilization, seeds are germinated in Petri dishes on moisturized Wattman paper and then transplanted into plastic pots of 1.5 liters, filled with an equivalent mixture of earth and sand at the rate of 2 seeds by pot. The inoculation of varieties cultivated under greenhouse, was made by pure cultures of bacteria. The experiment was realized in six repetitions for the statistical analysis. Later, plants were collected in order to compare their root system. The irrigation of plants was regularly made up to the field capacity, until water stress application.

Studied parameters:-

Leaves length:- Measures of leaf lengths have been taken to check the effect of bacterial strains and endomycorrhizae on the aerial part developments for both durum wheat and common wheat genotypes.

Root length:- After harvesting, measures were taken to see the effect of the bacterial strains and endomycorrhizae on root elongation.

Ear length:- For each variety, we measured ear length (beard not included).

Rate of total chlorophyll:- The rate of total chlorophyll was measured using a SPAD chlorophyll meter. Devise Calibration was performed by closing the vacuum clamp on itself. Then three test sockets are made on the sheet (top, middle, base) then the average value appears on the screen.

Nitrogen determination:- The Kjeldhal method was used to nitrogen determination as described by Rinaudo, (1970).

Plants dry weight:- Plants dry weight, expressed in grams, was determined after drying in an oven at 60° C for 96 hours.

Roots dry weight:- roots dry weight, expressed in grams, was determined as well after drying in an oven at 60 ° C for 96 hours.

Statistical analysis:- The results are statistically interpreted by an analysis of variance (ANOVA) and principal component analysis (PCA) through the XLSTAT software (2014) using XLSTAT 2014 software. The Newman-Keuls test lists the averages with a threshold of 5 % meaning.

Results:-

Growth parameters:-

The obtained results show genetic variability among the tested varieties. The variance analysis presented in tables (2, 3, 4, 5) showed a significant genotype, treatment and water regime effect. For leaf, root and ear length, data analysis shows that the higher significant values ($p < 0.0001$) are observed with IM treatment for all wheat varieties (Hidhab, Ain Abid, Boussellem, Waha) compared to controls and the other treatments. In case of stressed plants inoculated with IA, we noted that the growth of the variety Ain Abid (Table 3) is the highest compared to the others.

Root and plant dry weight:-

The effects of water stress on roots and plant dry weight are significant ($p < 0.0001$) tables (2, 3, 4, 5). These two parameters were measured in all wheat varieties. We observed that for root dry weight, the varieties Hidhab, Boussellem, Ain Abid with AI inoculum accumulated more dry matter in their roots, they seem to have the ability to develop a more important root system. While the variety Waha seems less responsive. The results of plant dry weight were significant ($p < 0.001$) for the IA inoculated and unstressed varieties Hidhab and Ain Abid, this treatment induced a higher response compared to controls. On the other hand Boussellem and Waha are distinguished under Mycorrhiza treatment. For inoculated plants under water stress, the greatest value of the root mass was noted in the variety Ain Abid inoculated IF compared with the control stressed, on the other hand the dry weight of plants is the highest recorded in waha inoculated by IA.

Rate of chlorophyll:-

The values of total chlorophyll levels indicate a difference between treatments. The non-stressed inoculated varieties have the highest chlorophyll content values compared to control plants and stressed inoculated plants. The evaluation of the chlorophyll content showed that all varieties respond negatively to water stress. Statistical analyzes tables (2, 3, 4, 5) showed a significant difference ($p < 0.0001$) between the inoculated plants and the controls ones. The highest values were obtained with the IF and IM treatments in Hidhab, Ain Abid and Boussellem, IA treatment in Waha As for the inoculated plants under water stress, the best treatment was IA with all varieties.

Nitrogen assay:-

For nitrogen assay analysis of variance (ANOVA) tables (2, 3, 4, 5) shows a significant difference ($p < 0.0001$) between the inoculated and control plants. The best treatment is AI for all varieties As well as under water stress, the highest values are obtained for the IA treatment of four the tested varieties.

Table 2:- Roots length, ear length, leaf length, rate of chlorophyll (SPAD), root dry weight, plant dry weight, nitrogen dosage, wheat plants Hidhab inoculated or not stressed or not with Frankia strains , Azospirillum and Mycorrhiza.

Variables	Treatment (T) ¹								SEM ²	Inoculation (Ino)		Significances (p-values) ³		
	Control	TS	IF	IA	IM	ISF	ISA	ISM		I	NI	T	Ino	T x Ino
Roots length(cm)	29.33 ^{abc}	30.88 ^{ab}	28.5 ^{bc}	29.5 ^{abc}	33.5 ^a	25.68 ^C	30.22 ^{abc}	29.32 ^{abc}	0.47	29.45	30.11	**	ns	**
Ear length (cm)	10.65 ^c	6.93 ^t	11.92 ^b	11.35 ^{bc}	11.35 ^{bc}	7.55 ^{ef}	9.25 ^d	8.5 ^{de}	0.36	8.79	10.48	***	*	***
Leaf length (cm)	20.3 ^{cd}	16.72 ^e	24.78 ^b	31.55 ^a	30.75 ^a	16.85 ^e	21.43 ^c	19.25 ^d	0.81	24.1	18.5	***	**	***
SPAD (SPAD Unit)	38.75 ^b	26.03 ^c	45.85 ^b	37.25 ^b	45.67 ^a	27.77 ^c	37.25 ^b	27.25 ^c	1.19	34.96	32.39	***	ns	***
Root dry weight(g)	2.18 ^b	3.21 ^a	1.19 ^b	3.11 ^a	1.60 ^b	1.60 ^b	1.19 ^b	1.16 ^b	0.15	2.96	1.64	***	**	***
Plant dry weight(g)	2.77 ^d	6.14 ^b	2.97 ^d	8.09 ^a	3.66 ^{cd}	5.31 ^b	6.13 ^b	4.88 ^{bc}	0.29	5.17	4.45	***	ns	***
Nitrogen dosage (%)	3.45 ^a	1.04 ^c	3.84 ^a	4.01 ^a	2.81 ^b	1.45 ^c	2.36 ^b	0.87 ^c	0.25	2.55	2.24	***	ns	***

¹ legend : TS : control stressed; IF: inoculation with Frankia cci3, IA: inoculation with Azospirillum brasilense, IM :inoculation with Mycorrhiza, ISF: Stressed plant inoculated with Frankia cci3,ISA: Stressed plant inoculated with Azospirillum brasilense, ISM: Stressed plant inoculated with Mycorrhiza, I: Inoculation, NI: No inoculation,T:Treatment, Ino: inoculation. ² SEM: standard error of the mean. ³ significances: <0.1: *, <0.05: **, <0.001: ***, <0.0001: **** the results in the same line followed by the same letter are not significantly different, according to the test Newman- Keuls test (p < 0.05).

Table 3. Roots length, ear length, leaf length, rate of chlorophyll (SPAD), root dry weight, plant dry weight, nitrogen dosage, wheat plants Ain Abid inoculated or not stressed or not with Frankia strains , Azospirillum and Mycorrhiza.

Variables	Traitement (T) ¹								SEM ²	Inoculation(Ino)		Effects (pvalues) ³		
	Control	TS	IF	IA	IM	ISF	ISA	ISM		I	NI	T	Ino	TxIno
Roots length(cm)	25.5 ^d	30.62 ^{bc}	31.33 ^b	30.25 ^{bc}	34 ^a	20.33 ^e	29.55 ^{bc}	28.43 ^c	0.61	28.98	28.05	***	ns	***
Ear length (cm)	9.88 ^c	6.68 ^d	12.56 ^b	12.2 ^b	14.61 ^a	7.76 ^d	9.03 ^c	7.63 ^d	0.40	10.63	8.28	***	**	***
Leaf length (cm)	16.28 ^e	13.51 ^t	31.63 ^a	29.25 ^{bc}	30.25 ^{ab}	13.31 ^f	28.41 ^c	20.96 ^d	1.08	25.63	14.9	***	***	***
SPAD (SPAD Unit)	31.30 ^c	26.63 ^d	35 ^a	28.33 ^d	33.25 ^b	27.98 ^d	28.09 ^d	27.89 ^d	0.44	30.09	28.96	***	ns	***
Root dry weight(g)	1.65 ^{bc}	3.73 ^a	1.45 ^{bc}	2.22 ^b	1.72 ^{bc}	1.85 ^{bc}	1.19 ^c	1.13 ^c	0.13	1.59	2.68	***	***	***
Plant dry weight(g)	2.38 ^d	5.73 ^a	3.27 ^{cd}	4.34 ^{bc}	3.18 ^{cd}	5.01 ^{ab}	4.34 ^{bc}	3.91 ^{bc}	0.22	4.33	4.05	***	ns	***
Nitrogen dosage	2.97 ^a	1.33 ^c	3.22 ^a	3.73 ^a	3.18 ^a	0.36 ^d	2.14 ^b	0.82 ^{cd}	0.25	2.24	2.15	***	ns	***

¹ legend : TS : control stressed; IF: inoculation with Frankia cci3, IA: inoculation with Azospirillum brasilense, IM :inoculation with Mycorrhiza, ISF: Stressed plant inoculated with Frankia cci3,ISA: Stressed plant inoculated with Azospirillum brasilense, ISM: Stressed plant inoculated with Mycorrhiza, I: Inoculation, NI: No inoculation,T:Treatment, Ino: inoculation.. ² SEM: standard error of the mean. ³ significances: <0.1: *, <0.05: **, <0.001: ***, <0.0001: ****, the results in the same line followed by the same letter are not significantly different, according to the test Newman- Keuls test (p < 0.05).

Table 4:- Roots length, ear length, leaf length, rate of chlorophyll (SPAD), root dry weight, plant dry weight, nitrogen dosage, wheat plants Boussellem inoculated or not stressed or not with Frankia strains, Azospirillum and Mycorrhiza.

Variables	Traitement (T) ¹								SEM ²	Inoculation(Ino)		Effects(pvalues) ³		
	Control	TS	IF	IA	IM	ISF	ISA	ISM		I	NI	T	Ino	TxIno
Roots length(cm)	24.50 ^f	33.47 ^c	35.50 ^b	38.83 ^a	39.82 ^a	31.20 ^b	30.03 ^{de}	29.07 ^e	0.61	34.07	29.43	***	**	***
Ear length (cm)	9.63 ^{bc}	6.48 ^d	8.42 ^{bc}	12.42 ^a	13 ^a	8.42 ^{bc}	9.03 ^{bc}	9.92 ^b	0.35	10.01	7.93	***	**	***
Leaf length (cm)	19.95 ^c	15.33 ^d	19.78 ^c	23.25 ^b	23.65 ^a	14.50 ^d	23.92 ^a	20.08 ^c	0.73	21.52	17.42	***	**	***
SPAD(SPAD Unit)	36.43 ^b	28.66 ^c	39.50 ^a	40.47 ^a	39.72 ^a	28.22 ^c	27.34 ^c	26.69 ^c	0.92	33.52	32.12	***	ns	***
Root dry weight(g)	1.58 ^{bc}	2.94 ^a	1.16 ^c	2.34 ^{ab}	2.37 ^{ab}	1.56 ^{bc}	1.53 ^{bc}	1.19 ^c	0.08	1.71	2.39	***	**	***
Plant dry weight(g)	3.06 ^d	6.29 ^a	4.20 ^c	3.15 ^d	3.37 ^d	5.03 ^b	6.62 ^a	3.83 ^{cd}	0.20	4.41	4.9	***	ns	***
Nitrogen dosage	3.11 ^a	0.84 ^c	3.08 ^a	3.44 ^a	3.12 ^a	0.40 ^c	2.10 ^b	0.95 ^c	0.21	2.18	1.41	***	ns	***

¹ legend : TS : control stressed; IF: inoculation with Frankia cci3, IA: inoculation with Azospirillum brasilense, IM :inoculation with Mycorrhiza, ISF: Stressed plant inoculated with Frankia cci3,ISA: Stressed plant inoculated with Azospirillum brasilense, ISM: Stressed plant inoculated with Mycorrhiza, I: Inoculation, NI: No inoculation, T:Treatment, Ino: inoculation.. ² SEM: standard error of the mean. ³ significances: <0.1: *, <0.05: **, <0.001: ***, <0.0001: ****, the results in the same line followed by the same letter are not significantly different, according to the test Newman- Keuls test (p < 0.05).

Table 5. Roots length, ear length, leaf length, rate of chlorophyll (SPAD), root dry weight, plant dry weight, nitrogen dosage, wheat plants Waha inoculated or not stressed or not with Frankia strains , Azospirillum and Mycorrhiza.

Variables	Traitement (T) ¹								SEM ²	Inoculation (Ino)		Effects (p-values) ³		
	Control	TS	IF	IA	IM	ISF	ISA	ISM		I	NI	T	Ino	TxIno
Roots length(cm)	24.91 ^e	33.9 ^a	32.25 ^{ab}	28.16 ^d	21.5 ^f	31.75 ^{ab}	29.01 ^{cd}	30.76 ^{bc}	0.61	28.91	29.41	***	ns	***
Ear length (cm)	9.93 ^b	6.50 ^c	9.65 ^b	13.18 ^a	13.35 ^a	9.65 ^b	8.82 ^b	9.57 ^b	0.35	10.35	8.22	***	**	***
Leaf length (cm)	21.85 ^c	16.15 ^e	22.50 ^b	25.62 ^b	26.75 ^b	13.72 ^f	28.72 ^a	19.92 ^d	0.73	22.87	19	***	**	***
SPAD (SPAD Unit)	38.10 ^b	28.79 ^d	41.83 ^a	36.13 ^c	41.70 ^a	27.62 ^d	28.31 ^d	27.09 ^d	0.92	33.95	33.45	***	ns	***
Root dry weight(g)	1.29 ^c	2.58 ^a	1.68 ^{bc}	2.19 ^{ab}	1.80 ^{bc}	1.72 ^{bc}	1.59 ^{bc}	1.24 ^c	0.08	1.7	1.94	***	ns	***
Plant dry weight(g)	3.54 ^e	7.37 ^a	3.84 ^e	3.50 ^e	3.64 ^e	5.30 ^e	6.22 ^b	4.42 ^d	0.20	4.49	5.45	***	*	***
Nitrogen dosage	3.14 ^a	1.21 ^c	2.48 ^{ab}	2.73 ^{ab}	3.01 ^a	0.40 ^d	2.19 ^b	0.73 ^{cd}	0.21	2.18	1.92	***	ns	***

¹ legend : TS : control stressed; IF: inoculation with Frankia cci3, IA: inoculation with Azospirillum brasilense, IM :inoculation with Mycorrhiza, ISF: Stressed plant inoculated with Frankia cci3,ISA: Stressed plant inoculated with Azospirillum brasilense, ISM: Stressed plant inoculated with Mycorrhiza, I: Inoculation, NI: No inoculation,T:Treatment, Ino: inoculation.. ² SEM: standard error of the mean. ³ significances: <0.1: *, <0.05: **, <0.001: ***, <0.0001: ****, the results in the same line followed by the same letter are not significantly different, according to the test Newman- Keuls test (p < 0.05).

Correlation among the studied parameters:-

The correlation analysis of the studied parameters was used to select the most discriminating variable. The ear length has been chosen. The majority of the correlations were highly significant ($p < 0.0001$), however, only values greater than 0.5 will be discussed. It seems that the highest correlation is the one between ear length (cm) with chlorophyll rate (SPAD unit) ($r = 0.70^{***}$), followed by the correlation between ear length (cm) and leaf length (cm) ($r = 0.50^{***}$) (Table 6).

Table 6. Correlation matrix of the measured parameters of durum and soft wheat

Variables	Roots length	Ear length	Leaf length	SPAD	Root dry weight	Plant dry weight
Ear length (cm)	0.24**					
Leaf length (cm)	-0.02	0.50***				
Rate of chlorophyll (Unit SPAD)	0.17*	0.70***	0.40***			
Root dry weight(g)	-0.13	-0.29***	0.02	-0.19**		
Plant dry weight(g)	-0.13	-0.57***	0.04	-0.54***	0.48***	
Nitrogen dosage (%)	-0.21**	0.23**	0.53***	0.30***	0.16*	0.03

Significances: < (0.1): *, < (0.05): **, < (0.001): ***, < (0.0001): ****.

PCA analysis Hidhab:-

The performed analysis is a PCA centered reduced. According to the Kaiser criterion, only two factors were selected (F1 and F2) which represent 71% of the total variance, which is quite good and can be used to identify the main parameters and the discriminating treatment. Thus, it seems that the first factor F1 represents 49% of the variance. It is positively correlated, and rather strongly, with the ear length, chlorophyll rate followed by nitrogen and root length which is less important. Regarding root and plant dry weight we notice that, there is a negative correlation. The inoculated plants with the tree treatments (IF, IA, IM) and the control plants are positively correlated however inoculated plants under water stress and stressed control (TS) are negatively correlated.

The axis 1 is therefore, in some ways, the overall result (in all settings and treatments considered). (Figure 1)

The axis F2 which represent 22% of the total variance is marked by an opposition. It has a lower inertia than the first, he opposed in one hand, plant dry weight and root dry weight (positive correlations), on the other hand, ear length and chlorophyll rate (negative correlations). It is therefore an opposition axis between photosynthesis and growth parameters and dry weight parameters (plants and roots). The axis represents F2 treatments inoculated plants under water stress (ISF, ISA, ISM), inoculated plants (IF) and the control plants were negatively correlated and in opposite inoculated plants (IA, IM) and the control stressed plants (TS) are positively correlated.(Figure 1).

PCA analysis Ain Abid:-

Successful factors (F1 and F2) represent 69% of total variance, F1 represents 52%. We notice that the first axis is distinguished primarily by ear length and nitrogen rate monitoring chlorophyll. Root length is of less importance. Concerning plant and root dry weight there is a negative correlation. Treatments axis F1 plants inoculated (IF, IA, IM) are positively correlated however, inoculated plants under water stress as well as control and stressed control (TS) are negatively correlated. Axis 1 represents the overall result (in all parameters and treatment considered) (Figure 1).

The axis F2 represents 17% of variance we notice a reverse trend which has a lower inertia than the first, these are the variables: plant and root dry weight (positive correlations), ear length and chlorophyll rate (negative correlations). It is therefore an opposition axis between photosynthesis or growth parameters and dry weight parameters (plant and root).

Treatments axis F2 inoculated plants under water stress (ISA) inoculated plants (AI) and control plants under stress (TS) are positively correlated in opposite of inoculated plants (IM), and the control plants inoculated plants under water stress are negatively correlated (Figure 1).

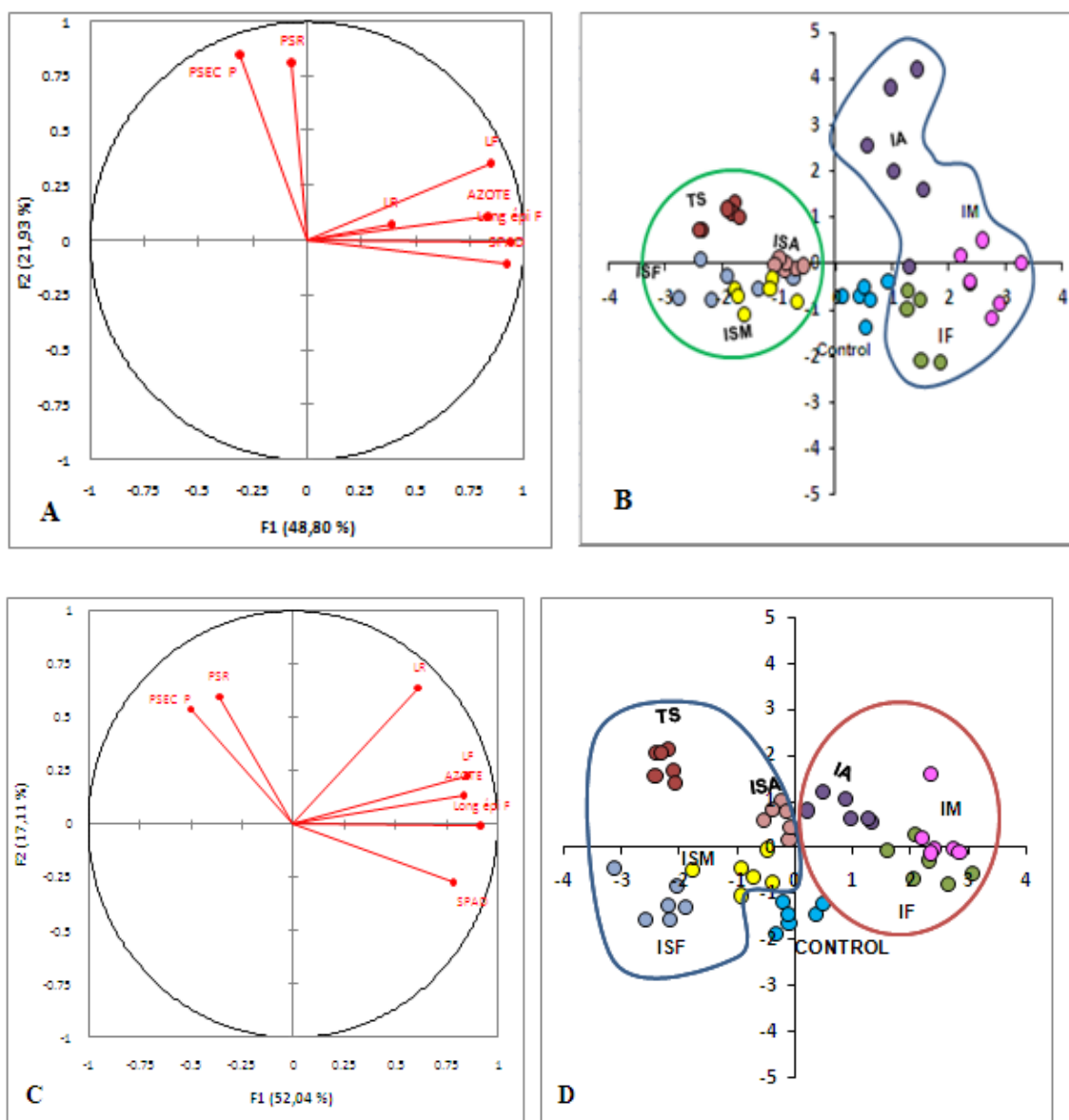


Figure 1:- Principal component analysis (A, B) Projection of measured parameters (length of ear, leaf and root, dry weight of the plant and roots, nitrogen content and chlorophyll) inoculated treatment and not inoculated respectively with Hidhab variety on the factorial F1 / F2 (mean \pm standard errors) ; (C, D) Projection of measured parameters (length of ear, leaf and root, dry weight of the plant and roots, nitrogen content and chlorophyll) inoculated treatment and not inoculated respectively with Ain Abid variety on the factorial F1 / F2 (mean \pm standard errors)

PCA analysis Boussellem:-

The factors (F1 and F2) represent 74% of the total variance, we notice that the first factor F1 represents 59% of the variance and is positively correlated with nitrogen and ear length, chlorophyll rate and leaf length. However root length, plant and root dry weight are negatively correlated. Treatments axis F1 plants inoculated (IF, IA, IM) and the control plants are positively correlated while the inoculated plants under water stress and control (TS) are negatively correlated (Figure 2).

The axis F2 represents 15% of the total variance, we observe that the variables are positively correlated and mostly in a relatively pronounced way, the second axis seems to be interpreted as a factor representing a global measure of the parameters. The axis F2 represents treatment inoculated plants under water stress (ISF, ISM) control are

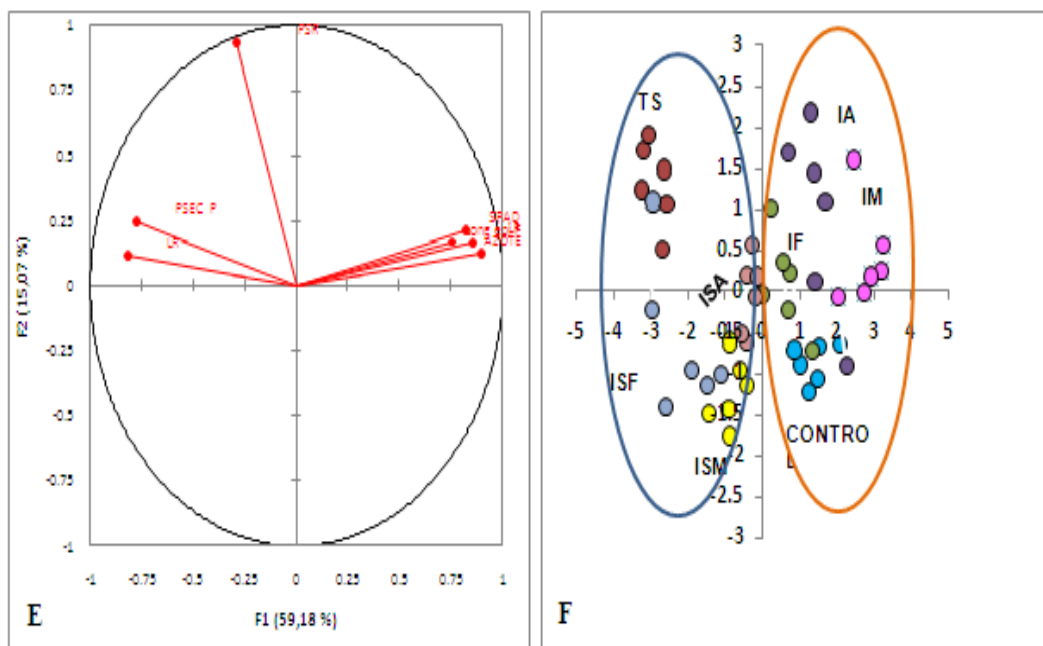
negatively correlated in the opposite inoculated plants (IF, IA, IM) and stressed control plants (TS) and inoculated plants under water stress (ISA) are positively correlated (Figure 2).

PCA analysis Waha:-

The two factors (F1 and F2) represent 89.79% of the total variance, which is good to identify key parameters and discriminating treatment. It is noted that the first factor F1 represents 73.46% of the variance; it is positively with nitrogen, plant dry weight, root dry weight and leaf length. Moreover root length, ear length and chlorophyll rate show a negative correlation.

The F1 treatments axis represents the inoculated plants under water stress (ISA) and the control plants are positively correlated unlike the inoculated plants under water stress (ISF, ISM) the control stress (TS) and inoculated plants (IF, IA IM) are negatively correlated (Figure 2).

The axis F2 represents 16.33% of the total variance, it shows that variables are positively correlated, and the second axis seems to be interpreted as a factor representing a global measure of the parameters. Treatments axis F2, the inoculated plants under water stress (ISF, ISM), the stressed control plants (TS) are negatively correlated, in the opposite inoculated plants (IF, IA, MI), stressed inoculated plants (ISA) and the control plants are positively correlated (Figure 2)



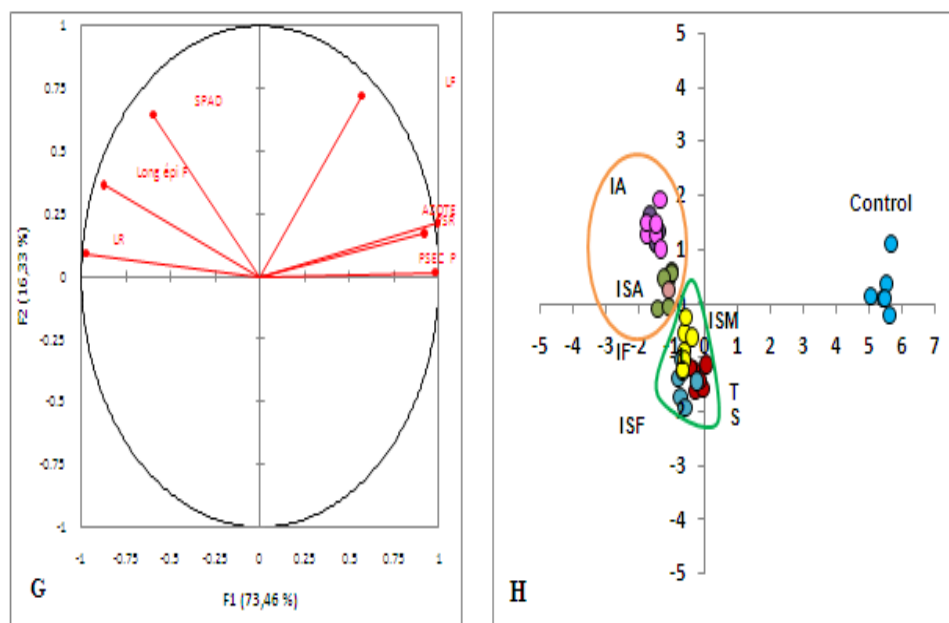


Figure 2:- Principal component analysis (E, F) Projection of measured parameters (length of ear, leaf and root, dry weight of the plant and roots, nitrogen content and chlorophyll) inoculated treatment and not inoculated respectively with Boussemel variety on the factorial F1 / F2 (mean \pm standard errors) ; (G, H) Projection of measured parameters (length of ear, leaf and root, dry weight of the plant and roots, nitrogen content and chlorophyll) inoculated treatment and not inoculated respectively with Waha variety on the factorial F1 / F2 (mean \pm standard errors)

Discussion:-

This study aims to compare between varieties of durum and common wheat under two water regimes and different treatments of microsymbionts strains. The results have shown that the effects of microbial inoculation on varieties vary according to the inoculants used. Stimulation of leaf length is observed with IM treatment that improves leaf length (+ 60%) compared to controls. Concerning the AI treatment, it improves the water-stressed plants behavior for all varieties. These results on the inoculation of *Azospirillum* are in agreement with those reported by Ramdani (2002) on durum wheat inoculation (*Triticum durum* var. Hedba3) by a *Azospirillum* sp., in the presence or in absence of nitrogen fertilizers, which resulted in a significant increase of leaf length (+ 12%). SH Li (1990) worked on *Prunus persica*, they said that leaf growth is sensitive to drought since limiting effect of water stress appears early and with intensity. Benmahioul (2009) also confirms our results and said that the reduction of aerial observed growth in seedlings can be explained by increasing levels of some growth regulators, including abscisic acid and cytokinins induced by stress. Furthermore the results for ear length showed that IM inoculation can improve growth in length (+ 67%) compared to controls. These inoculated plants have better growth compared with non-inoculated plants, this effect is maintained even under water stress condition. Similar results were observed by several studies (Jesus et al, 2004, Subramanian et al, 1996). The stimulation of growth by mycorrhizae is a result of an improvement in nutritional status and especially phosphate (Subramanian et al., 1997).

Furthermore IM treatment allowed an increase in root length (+ 60%) in Hidhab, Ain Abid and Waha. The IA treatment, under water stress, increased root length of all varieties over treatments and controls. Those results are in agreement with Dodd (1994), he reported that the extracellular mycelium *Glomus geosporum* and *G. monosporum* may extend a distance of 6 to 9 cm from the root. The effectiveness of mycorrhizal root systems is mainly due to an extension of the absorption surface and soil volume explored through hyphae fungal. Bizet (2014) showed that roots are able to grow in a more or less ground forced through physical support provided by the soil. The efficiency of extracting water from the soil by the roots is one of the adaptation characters that allow the plant to avoid or, more exactly, to delay its tissue dehydration. Moreover, Slama (2005) showed that the increase of absorption may be due to an extension in absorption depth and area, to growth rate and root extension. Simard (2014) noted that a decrease in the resistance to transport water to the roots is lost when there is addition of nutrients to the soil, thus demonstrating an indirect effect of mycorrhiza on plant water balance. This fact lead to this conclusion: in soils low

in nutrients, water is better absorbed by the plant, because the fungus allows it to draw a greater amount of minerals (especially phosphorus) thereby generating a good water absorption which became more important as the roots ensure their growth.

In this study, the tested wheat varieties have a considerable fall and highly significant ($p < 0.0001$) in chlorophyll content under water stress. However, with good water supply, results show values with an average order of 38 U.SPAD for all studied varieties. The chlorophyll content decrease in stressed plants is reported by many authors as one of the major cause's productivity and growth reduction (Guerfel, 2009; Ghobadi, 2011; Wang, 1997). These results confirm the observations of Booba (2009), who mentioned that the lack of water causes a drop in leaves chlorophyll content. The amount of chlorophyll leaves can be influenced by many factors such as leaf age, leaf position, and also by environmental factors such as light, temperature and water availability (Hikosaka et al., 2006).

According Chandrasekhar (2000), the reduction of chlorophyll is mostly due to lower thylacoidal protein content in chloroplasts and a decrease in photosystems in the cell thylacoïdale (Quartacci et al., 1995). According Tambussi (2007) a decrease in Chlorophylls levels disrupts the photosynthetic mechanism of the plant upper part, leaves and ears. It also severely alters the grain filling and thus affects the final yield. In the same context the work of (Fourkes, 2007; Ehdaia, 2008) indicate that the reduction of the chlorophyll disrupts the redistribution of assimilates stored by the rod to different parts plant disrupting growth.

Indeed, wheat absorbs nitrogen under normal conditions. However nitrogen deficiency occurs during rapid growth or when the plant is very stressed; which is characterized by significant leaf yellowing because nitrogen is not very mobile. The results showed that the stressed and non-stressed plants inoculated with the AI treatment have greater nitrogen leaf content (+ 80%) in Hidhab, Ain Abid and Waha. And the AI treatment has greater nitrogen leaf content stressed plants (+40%) compared to controls. According to App, (1980); Wetselaar, (1981), nitrogen balance studies have long demonstrated the important role of microorganisms in nitrogen fixation in rice. Nitrogen fixation can be high, according to Roger (1992) several types of microorganisms are involved and among them, heterotrophic nitrogen-fixing bacteria in rice rhizosphere: on and in the roots, and represent a significant proportion of the nitrogen requirements of the plant. The diazotrophs are extremely abundant (Thomas et al Bauzon 1982 Ladha et al 1987) and may represent up to 80% of the total microflora (Barraquio and Watanabe 1981).

The results for root and plant dry weight showed that the production of dry matter was improved by inoculation. This dry material was used to produce new roots, their proliferation (root volume), their elongation (increase in length) and their maintenance. (El Fakhri et al., 2010). IA treatment allows an increase in dry weight roots (+59%) in Bousellem and waha in comparison with controls and (+ 74%) in Ain Abid. On the other side IM treatment allows an increase in dry weight of the plant (+ 91%) in Boussellennm and Waha. These results were reported by several authors on various culturs such as the work of Pedraza (2010) on strawberry, the roots inoculated by *Azospirillum brasilense* strains had a high rate of root system infection where a high dry matter production in aerial part and especially root portion. This explains the accumulation of root dry weight in non-stressed inoculated plants and in plants inoculated under water stress. In argan tree (*Argania spinosa*), Nouaim (1994) observed that mycorrhizal could increase by 3 to 4 times the solids formed, while decreased from 40 to 50% the ratio root part / aerial part and showed the best efficiency of a mycorrhizal root system. The results of this study invalidate those of other authors (Dib (1992); Benlaribi (1990); Al Hakimi (1993), for the durum wheat, showing that the water deficit inhibits further growth of root system as that of above.

The PCA analysis shows that the two varieties of common wheat have behaved in the same way and similar results were observed in both varieties of durum wheat. On one hand the two varieties of wheat showed that increasing nitrogen content results not only in increasing chlorophyll rate chlorophyll but also leaf length, ear and root elongation.

On the other hand the two varieties of durum wheat showed that inoculation by microsymbionts varies considerably between treatments. We noticed that the fixing nitrogen increases chlorophyll levels, also helps leaves, ear and roots elongation, it also makes a significant reserve of dry matter. It is assumed that this result may be representative of the nitrogen fixation process by microorganisms. So we can say that mycorrhizae help the plant to absorb nutrients from the soil such as nitrogen that is absorbed in nitric or ammonium form. Nitrogen is a major constituent of the chlorophyll and proteins. According to Evans and Seeman (1989) between 50 and 80% of the nitrogen of the leaf is

allocated to photosynthetic proteins, this complex process of photosynthesis allowed the plant chloroplasts multiplication and its development.

Conclusion:-

Through this study, it appears that the best tested inoculation treatments for durum and common wheat under both water regimes are the mycorrhiza and *Azospirillum brasilense* (PGPR bacteria). Our tests show that the inoculated varieties are more tolerant to water stress (Boussellem with mycorrhiza and Waha with *Azospirillum brasilense*). The mycorrhizal fungi are among the most important soil organisms to consider. The mycorrhizae are involved in mineral nutrition, absorption of water and protection against abiotic stresses. Thus mycorrhizae can contribute as an alternative to establish and develop adequate agriculture as the use of chemical fertilizers has reached their limits. Today, agriculture must move towards more sustainable practices.

Acknowledgements:-

This work was supported by the Ministry of Higher Education and Scientific Research of Algeria. Authors thank the ITGC (Institut Technique des Grandes Cultures) for providing seeds. Authors thank also Mr Mohammed Gagaoua and Mrs Ryma Bouldje for their help.

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