

## EVALUATION OF SOME PROMISING BREAD WHEAT LINES RESPONSIVE TO N<sub>2</sub>-BIOFERTILIZER UNDER NITROGEN LEVELS IN SANDY SOIL.

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### ABSTRACT

Irrational utilization of chemical fertilization and/or pesticides in many field crops has resulted in some environmental hazardous as well as increasing production costs. Therefore, development of new cultivars responsive to low level of fertilization and simultaneously yield sustainable are very needed. However, the Egyptian National Wheat Res. Program has established a new breeding program for biofertilizer response. Six promising bread wheat lines were screened from N-biofertilization breeding program conducted at El-Bustan region. These lines sustained their yield even with the application of only 50% of the nitrogen recommended dose (286 kg/ha) when the biofertilizer "Azottin" was used. This result encouraged us to evaluate these lines under five nitrogen fertilizer levels (25, 50, 75, 100 and 125% of the recommended nitrogen dose using N-biofertilization in sandy soil at El-Bustan region, El-Behera Governorate during 1997/1998 and 1998/1999 wheat growing seasons. The results showed that the interaction between genotypes and nitrogen levels was significant for the yield and its components, while the harvest index was not significant. However, the highest grain yields were obtained from the biofertilized lines numbers 15, 11 and 21 at 214, kg N/ha, lines number 8 and 21 at 286 kg N/ha and lines 8 and 15 receiving 143 kg N/ha. Moreover, they surpassed the grain yields of the control treatment (286 kg N/ha) by 0.80, 0.87, 0.55, 0.72, 0.59, 0.52 and 0.30 ton/ha (14, 16, 10, 13, 11, 10 and 5%), respectively, in addition, these seven combinations exceeded the commercial cultivar Sids 1 under control treatment by 1.46, 1.23, 1.04, 1.16, 1.08, 0.96 and 0.96 ton/ha (29, 24, 21, 23, 21, 19 and 19%), respectively. The results indicated that lines; 8 and 15 expressed high stability level in grain yield over the two seasons. They also, outyielded the commercial check Sids 1, under biofertilization with no more than 143 kg N/ha, saving 50% of the recommended dose. Such lines should save money and environmental health. Therefore, the two lines are highly recommended for yield tests in the advanced yield trials, which conducted all over the country through the National Wheat Research Program.

### INTRODUCTION

All the former wheat cultivars (*T. aestivum*) utilized in Egypt until the late sixties and early seventies did not need more than 100 kg N/ha (Hamissa *et al.*, 1971). With the introduction of the semi-dwarf cvs amounts needed reached in general, 143-190 kg/ha (Gomaa *et al.*, 1977), but it goes up to 216-286 kg N/ha according to location and soil type (Basilious *et al.*, 1992; Sabry *et al.*, 1994 and Mitkees *et al.*, 1998a & b). The extensive application of nitrogen fertilization in both old and new lands have raised serious environmental concerns due to the possibility of contaminating potable water sources, and subsequently creating health hazardous to human being and

living stocks as well. On the other hand, irrational utilization of chemical nitrogen fertilizers have been a serious problem due to the increase of production costs. This led to considerable interest in biological N<sub>2</sub>-fixation to reduce production costs and environmental hazardous, and hence, save human's money and health.

Many reports indicated that inoculation of seed or seedlings of various C<sub>3</sub> and C<sub>4</sub> plants with associative N<sub>2</sub>-fixing bacteria led to change in plant growth and yield (Dobereiner and Day, 1975; Von Bulow and Dobereiner, 1975; Eid, 1982 and Pohlman and McColl, 1982). However, Hubbell and Gaskine (1980) and Vlassak and Reynders (1981a, b) reported that N<sub>2</sub>-fixation might be of secondary importance due to growth regulating substances from the root-associated bacteria. Larson and Neal (1978) and Rennie (1981) reported that wheat is established as possessing a genotype effect and the challenge is to introduce the capacity for N<sub>2</sub>-fixation to a wider and more useful genetic base.

Under Egyptian conditions, several investigations were carried out to evaluate the potentiality of associative symbiosis e.g. *Azospirillum* spp. with sugarcane, maize, sorghum, rice, wheat and desert plant (Amer, 1978; Hegazi *et al.*, 1979; Omar, 1980; Fouad, 1981; and Eid, 1982). Results showed the abundance of rhizocoenses in the rhizosphere of the given systems. Eid *et al.*, (1986) found that grain yield of barley cv. Giza 121 and wheat cv. Giza 155 increased by 67 and 45% for the two cvs., respectively with N<sub>2</sub>-fixing bacteria, indicating the importance of the varietal response to such biofertilizers. Esaad Bedaiwi *et al.* (1997) found that biofertilization could save 40% of nitrogen requirements for wheat in the new lands. Also, Mitkees *et al.* (1998a) found that six out of 21 bread wheat genotypes produced the highest yields under biofertilization in addition to only half of the recommended nitrogen dose (286 kg/ha).

This study was undertaken to determine the response of those selected lines inoculated with Azottin biofertilization to different nitrogen levels in sandy soil.

## **MATERIALS AND METHODS**

Two field experiments were conducted in sandy soil at El-Bustan region in El-Behera Governorate, during 1997/98 and 1998/99 growing seasons. This experiment was carried out to study the effect of N<sub>2</sub>-biofertilization on response of six promising bread wheat lines obtained from the lines proved their superiority in primary yield trials, in addition to the commercial cultivar Sids 1 (Table 1), to five nitrogen fertilizer levels (71.5, 143, 214, 286 and 357 kg N/ha represented 25, 50, 75, 100 and 125% of the recommended level in the region "about 30, 60, 90, 120 and 150 kg N/fed", under biofertilization as seed coated at sowing comparing to the recommended treatment (286 kg N/ha) without biofertilization.

**Table 1: List of the studied wheat genotypes and their cross names and pedigrees.**

Genotypes	Name and Pedigree
Sids 1	Control
L-8	Nac/Vee"S"
L-9	Buc"S"/FLK"S"//Maya "S"/VUL "S"
L-11	Nac/Vee "S"
L-15	Seri # 3/Buc "S"
L-20	DW 15023/Snb "S"//Snb "S"
L-21	Juf/BJY "S"//Ures

The biofertilizer used was "Azottin", a mixture of the associative N<sub>2</sub>-fixing bacteria (Azospirillum, Bacillus, Klebsiella and Azotobacter) produced by Soils, Water and Environ. Res. Inst., ARC. During land preparation, phosphorus fertilizer was applied in a rate of 72 kg P<sub>2</sub>O<sub>5</sub>/ha in the form of mono-ammonium phosphate. Likewise, potassium fertilizer was added in two equal doses (57 kg K<sub>2</sub>O/ha each) applied as potassium sulphate 48% during land preparation and at booting stage. Nitrogen fertilizer ammonium nitrate (33%) was applied at six rates according to the studied treatments. Nitrogen fertilizer was splitted in all cases into six equal doses applied right before irrigation starting from plant emergence to the end of booting stage. The soil of the experimental site was sandy in texture having low available nitrogen as well as phosphorus and organic matter. However, chemical and mechanical analyses is presented in Table 2.

**Table 2: Soil chemical and mechanical analyses of the experimental sites.**

Analysis	Season	
	1997/1998	1998/1999
EC mmhos/cm	0.18	0.12
pH	8.2	8.2
N (ppm)	13	15
P (ppm)	11	16
K (ppm)	60	65
CaCO <sub>3</sub> %	5.5	5.8
OM %	0.50	0.45
Sand %	91.1	89.4
Silt %	1.1	1.1
Clay %	7.8	9.5
Texture	Sandy	Sandy

The experiments were laid out in a split plot design with four replications. The wheat genotypes were assigned to the main plots and fertilization levels to the sub-plots. The plot size was 4.5 m<sup>2</sup>. The preceding crop was maize in each season. Sowing was taken place during the last week of November in both seasons at row spacing of 15 cm using 400 seeds/m<sup>2</sup>. Fixed sprinkler irrigation system was used at 7-day intervals. Other cultural practices recommended for the region were followed. The crop was harvested on May 1<sup>st</sup> for the two seasons. Data were collected for grain yield, straw yield, number of spikes/m<sup>2</sup>, number of kernels/spikes and 1000 kernel weight. Collected data were subjected to statistical analyses according to Steel and

Torrie (1980). In addition, curve linear regression analysis (Steel and Torrie, 1980) was performed to illustrate the response to nitrogen levels under biofertilization condition. Applying principles of Eberhart and Russel (1966), stability study was performed over the six promising lines during the two seasons.

## **RESULTS AND DISCUSSION**

The combined analysis over data of the two wheat growing seasons, 1997/1998 and 1998/1999 revealed that there were significant differences in grain yield, number of spikes/m<sup>2</sup>, 1000-kernel weight and harvest index (HI) due to the genotypes effect. Lines number 15, 8, 21 and 11 were statistically at par and gave the highest grain yields irrespective of fertilization treatments. Moreover, these four lines outyielded the commercial cultivar; Sids 1 by 0.66, 0.53, 0.47 and 0.46 ton/ha (13, 10, 9 and 9%), respectively. These increments generally could be due to more spikes/m<sup>2</sup>, large numbers of kernels/spike and heavy kernel weight as compared with the other genotypes. Meanwhile, differences in number of kernels/spike were not significant in that respect (Table 3). On the other hand, no significant differences in straw yield characters had been detected among the studied genotypes. These results are in general agreement with those obtained by Mitkees *et al.* (1989), Iman Sadek *et al.* (1995), Abo-Warda (1997), Hanna *et al.* (1997) and Iman Sadek and Abo Warda (1998).

Regarding nitrogen fertilizer effect, the results indicated that 214 kg N/ha (75% of the recommended dose) in presence of biofertilization, was sufficient to achieve the highest grain yield and saved 71 kg N/ha. Furthermore, this treatment exceeded the control treatment (286 kg N/ha without biofertilization) by 0.52 ton/ha (10%). This might be attributed to corresponding increases in number of the productive tillers expressed as number of fertile spikes/m<sup>2</sup>, number of kernels/spike and 1000-kernel weight. On the other hand, adding more nitrogen than 214 kg N/ha decreased the grain yield. In spite of the progressive increase in straw yield with increasing nitrogen supply in all plots treated with Azottin, the harvest index significantly decreased with more than 214 N kg/ha. This indicates that adding nitrogen more than 75% of the recommended dose stimulated the initiation of the nonproductive tillers. These results are in general agreement with those obtained by Mitkees *et al.* (1996), Essad Bedaiwi *et al.* (1997) and Mitkees *et al.* (1998a).

**Table 3: Grain yield and other agronomic traits of some new lines of bread wheat as affected by different Nitrogen levels under N<sub>2</sub> biofertilizer at El-Bustan region combined over 1997/98 and 1998/99 growing seasons.**

	Grain yield ton/ha	Straw yield ton/ha	Harvest index %	Number of spikes/ m <sup>2</sup>	Number of kernels/ spike	1000-kernel weight gm
<b>Genotypes (G)</b>						
Sids 1	5.14	12.0	30.2	429	40.5	41.3
L. 8	5.67	12.6	31.2	447	42.1	43.2
L. 9	5.50	12.4	30.8	427	41.5	43.6
L. 11	5.60	12.5	31.2	428	42.4	44.1
L. 15	5.80	12.4	32.0	445	42.1	43.3
L. 20	5.45	12.1	31.0	437	41.6	43.0
L. 21	5.61	11.9	32.2	456	42.1	42.5
LSD for G at 0.05	0.29	NS	1.1	21.0	NS	1.1
<b>Fertilization (N)</b>						
Azottin + 71.5 kg N/ha	4.85	10.4	32.0	383	39.3	41.3
Azottin + 143 kg N/ha	5.71	12.0	32.2	454	42.3	43.0
Azottin + 214 kg N/ha	5.92	12.4	32.4	458	43.6	44.1
Azottin + 286 kg N/ha	5.67	13.5	29.6	446	42.4	43.4
Azottin + 357 kg N/ha	5.69	13.7	29.3	456	41.8	43.6
Control 286 kg N/ha	5.40	11.6	32.0	434	41.0	42.7
LSD for N at 0.05	0.18	0.4	0.8	14	1.1	1.4
G x N Interaction	*	*	NS	*	*	*

Similarly, the interaction between genotypes and nitrogen levels was significant for the yield and its components, while, the harvest index was not significant (Table 4). However, the highest grain yields were obtained from the biofertilized lines number 15, 11 and 21 at 214, lines number 8 and 21 at 286 and lines 8 and 15 receiving 143 kg N/ha. Moreover, they surpassed the grain yields of the control treatment by 0.80, 0.87, 0.55, 0.72, 0.59, 0.52 and 0.30 ton/ha (14, 16, 10, 13, 11, 10 and 5%), respectively. In addition, these seven combinations exceeded the commercial cultivar Sids 1 under control treatment by 1.46, 1.23, 1.04, 1.16, 1.08, 0.96 and 0.96 ton/ha (29, 24, 21, 23, 21, 19 and 19%), respectively. Therefore, lines coated with Azottin, number 15, 11 and 21 at 214 kg or lines 15 and 8 even at 143 kg N/ha saved 71-144 kg N/ha. These increments in grain yield could be attributed to their high values for all yield components (Table 4). The obtained results are in harmony with those reported by Mitkees *et al.* (1996), Esaad Bedaiwi *et al.* (1997) and Mitkees *et al.* (1998 a and b).

Regression analyses for the averaged values over the two seasons showed that lines number 8, 15, 20 and 21 had the highest coefficients of determination (78.7, 82.7, 96.0 and 88.7%) as compared with 68% for Sids 1 (Table 5 and Fig. 1). Lines number 8, 15 and 21 yielded the maximum, i.e. 6, 12, 6.37 and 6.10 t/ha at 274, 247 and 218 kg N/ha (115, 104 and 90 kg N/fed.), respectively, outyielding the commercial check Sids 1 by 0.63, 0.88 and 0.61 t/ha (11, 16 and 11%), respectively, at 276 kg N/ha (116 kg N/fed). However, decreasing nitrogen application to 143 kg N/ha would insignificantly decrease the yield with no more than 0.25-0.50 t/ha (4-8%). These results are supported by those obtained by Mitkees *et al.* (1998a) and Esaad Bedaiwi *et al.* (1997).

**Table 4: Grain yield and other agronomic traits as affected by the interaction between some new lines of bread wheat and Nitrogen fertilization under N<sub>2</sub>- biofertilizer at El-Bustan region combined over 1997/98 and 1998/99 growing seasons.**

Genotypes	Kg N/ha + biofertilizer					Control	kg N/ha + biofertilizer					Control
	71.5	143	214	286	357		71.5	143	214	286	357	
	Grain yield (ton/ha)						Straw yield (ton/ha)					
Sids 1	4.34	5.39	5.43	5.12	5.51	5.03	9.4	11.5	12.2	12.8	13.9	11.9
L. 8	4.76	5.99	5.68	6.19	5.93	5.47	10.8	12.3	12.6	14.4	13.9	11.5
L. 9	4.76	5.58	5.78	5.47	5.93	5.46	10.9	11.8	12.2	13.6	13.8	12.2
L. 11	5.01	5.93	6.26	5.09	5.94	5.39	10.1	12.7	13.1	13.0	14.3	11.6
L. 15	4.80	5.99	6.49	5.88	5.97	5.69	10.2	12.7	13.0	13.4	14.6	10.7
L. 20	5.08	5.43	5.69	5.80	5.47	5.25	10.5	11.5	12.1	13.3	13.4	12.0
L. 21	5.19	5.66	6.07	6.11	5.08	5.52	10.7	11.6	11.5	14.1	11.9	11.4
CV%	9						9					
LSD at 0.05	0.52						1.1					
	Harvest index %						Number of spikes/m <sup>2</sup>					
Sids 1	31.2	31.8	30.9	28.8	28.4	30.0	372	452	428	438	453	433
L. 8	30.8	32.6	31.1	30.2	29.9	32.4	358	485	454	471	479	436
L. 9	30.9	32.1	32.1	28.6	29.9	31.3	366	439	441	432	459	424
L. 11	33.2	31.7	32.3	28.5	29.5	31.8	398	446	459	392	462	414
L. 15	32.3	32.0	33.4	30.6	28.8	34.8	367	465	495	452	457	434
L. 20	32.7	32.1	32.0	30.2	28.7	30.6	400	431	438	455	455	443
L. 21	32.7	32.8	34.7	30.1	30.2	32.8	421	457	491	485	425	455
CV%	8						9					
LSD at 0.05	NS						37					
	Number of kernels/spike						1000-kernel weight (gm)					
Sids 1	38.0	40.5	42.9	41.3	39.1	41.1	39.6	41.1	43.1	41.3	41.8	40.8
L. 8	39.9	42.9	44.6	44.4	40.8	39.9	39.6	44.0	43.0	44.5	44.5	43.4
L. 9	38.1	42.3	42.5	41.0	42.8	42.3	42.5	43.1	44.8	42.1	45.4	43.8
L. 11	39.1	43.9	43.8	41.8	44.3	41.6	43.0	44.8	46.1	42.6	45.8	42.5
L. 15	38.6	43.1	43.3	44.6	43.3	39.6	41.6	43.5	44.6	44.0	43.8	42.1
L. 20	40.3	40.8	43.9	41.1	42.1	41.3	42.4	42.5	44.0	44.2	42.0	43.0
L. 21	41.4	43.0	44.3	42.4	40.4	41.1	40.0	41.8	42.9	45.4	41.8	43.3
CV%	7.2						6.0					
LSD at 0.05	3.0						1.8					

**Table 5: Regression analyses of response of some new promising bread wheat lines to Nitrogen fertilization under N<sub>2</sub>- biofertilizer**

Genotypes	Regression parameters			R <sup>2</sup> %	S-y	P <	Maximum, N		143 kg N/ha t/ha
	A	b	c				kg N/ha	t/ha	
Sids 1	3.697	0.0130	-235E <sup>-7</sup>	68.1	0.38	0.32	276	5.49	5.09
L. 8	3.855	0.0165	-302E <sup>-7</sup>	78.7	0.37	0.21	274	6.12	5.60
L. 9	4.205	0.0106	-174E <sup>-7</sup>	74.2	0.33	0.26	305	5.82	5.37
L. 11	4.510	0.0114	-231E <sup>-7</sup>	23.4	0.70	0.77	246	5.90	5.66
L. 15	3.439	0.0237	-780E <sup>-7</sup>	82.7	0.37	0.17	247	6.37	5.85
L. 20	4.385	0.0107	-212E <sup>-7</sup>	96.0	0.08	0.04	252	5.74	5.48
L. 21	3.857	0.0206	-474E <sup>-7</sup>	88.7	0.23	0.11	218	6.10	5.84

**Figure 1: Response of promising wheat lines to nitrogen fertilizer under N<sub>2</sub>-biofertilization.**

Stability study shown in Table (6) confirmed the pooled results for the two lines 8 and 15. They possessed the highest coefficients of determination (CD%) being 86 and 78%, respectively. The regression slope (b) for both lines did not significantly differ from unity (1.122 and 1.159) indicating their fitness for moderate environment and may respond for better ones. Also, deviation from regressions approached zero showing their good stability over the 12 treatments (environments).

**Table (6): Stability parameters of the seven studied wheat genotypes over twelve treatments.**

Genotypes	Mean ± SE	Slop b ± Sb	S <sup>2</sup> d	CD%
Sids 1	5.14 ± 0.57	0.943 ± 0.166	0.8528	76
Line 8	5.67 ± 0.66	1.122 ± 0.168	0.9530	86
Line 9	5.50 ± 0.66	1.160 ± 0.136	0.6668	86
Line 11	5.60 ± 0.61	0.767 ± 0.270	2.3959	44
Line 15	5.80 ± 0.71	1.210 ± 0.194	1.2658	78
Line 20	5.45 ± 0.74	1.159 ± 0.241	1.9945	68
Line 21	5.61 ± 0.61	0.642 ± 0.299	2.9772	31

In conclusion, the two lines; 8 and 15 are highly responsive to Azottin biofertilizer under low levels (no more than 143 kg N/ha “50% of recommended dose”) of chemical nitrogen fertilizer. So, the exploitation of such genetic variability should be the best goal to maximize the wheat productivity under low inputs in sandy soil, saving money and environmental health. Therefore, these two lines are highly recommended for yield tests in the advanced yield trials conducted all over the country through the National Wheat Research Program.

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تقييم إستجابة بعض السلالات المباشرة لقمح الخبز للتسميد الحيوى تحت معدلات مختلفة من التسميد النتروجينى فى الأراضى الرملية  
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الزيادة العشوائية غير المرشدة فى إستخدام السماد الكيمايى والمبيدات فى كثير من المحاصيل الحقلية ينتج عنها تلوث للبيئة وزيادة فى تكاليف الإنتاج. ولذا كان لإنتاج أصناف جديدة ذات إستجابة عالية مع التسميد المنخفض وتحقق أيضاً فى نفس الوقت قدرة محصولية عالية الأهمية الكبرى. ومن ثم بدأ البرنامج القومى لبحوث القمح برنامجاً جديداً للتربية للإستجابة للتسميد الحيوى وفيه تم إنتخاب 6 سلالات مباشرة من قمح الخبز فى منطقة البستان بمحافظة البحيرة وقد أثبتت هذه السلالات قدرتها المحصولية العالية عند التسميد بمعدلات أقل وصلت إلى 50% من الجرعة السمادية النتروجينية الموصى بها (286كجم/هكتار) مع إستخدام التسميد الحيوى (أزوتين). وقد شجعت هذه النتائج على إجراء بحث لتقييم هذه السلالات بإستخدام 5 مستويات من النتروجين (25 ، 50 ، 75 ، 100 ، 125%) (من الجرعة الموصى بها من التسميد النتروجينى) مع التسميد الحيوى فى الأراضى الجديدة خلال موسمى 1998/1997 و 1999/1998 . وتوضح النتائج أن التفاعل بين السلالات ومعاملات التسميد كانت معنوية للمحصول ومكوناته. وقد حققت أعلى محصول للحبوب من السلالات المعاملة بالأزوتين رقم 15 ، 11 ، 21 عند 214 والسلالات رقم 8 ، 21 عند 286 والسلالات رقم 8 ، 15 عند 143 كجم نتروجين/هكتار. أيضاً تفوق محصول هذه المعاملات نفسها على معاملة الكنترول (286 كجم نتروجين/هكتار) بمقدار 0.80 ، 0.87 ، 0.55 ، 0.72 ، 0.59 ، 0.52 و 0.30 طن/هكتار (14 ، 16 ، 10 ، 13 ، 11 ، 10 و 5%) على الترتيب. هذا بالإضافة إلى أن السبع معاملات قد تفوقت أيضاً على الصنف التجارى سدس 1 تحت معاملة الكنترول بمقدار 1.46 ، 1.23 ، 1.04 ، 1.16 ، 1.08 ، 0.96 و 0.96 طن/هكتار (29 ، 24 ، 21 ، 23 ، 21 ، 19 ، 19%) على الترتيب. وقد أظهرت النتائج أن السلالتين 8 و 15 مستوى عالٍ من الثبات الوراثى خلال السنتين مع كلٍ من المعاملات المدروسة. أيضاً تفوقت هاتان السلالتان على الصنف التجارى سدس 1 تحت ظروف التسميد الحيوى مع 50% فقط من التسميد النتروجينى الموصى به. مثل هذه السلالات من شأنها أن تقلل تكاليف الإنتاج وكذا تقلل من التلوث البيئى ، ولذا يوصى بإختبار تلك السلالتين فى تجارب المحصول فى المراحل المتقدمة المقامة فى جميع أنحاء الجمهورية من خلال البرنامج القومى لبحوث القمح.