

## THE BIOPOLYMER, AGAR AGAR, AS A SOIL CONDITIONER Part I : Greenhouse Experiments

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### **Abstract**

The biopolymer Agar Agar, derived from red alga belonging to Rhodophyceae, was tried as a gel soil conditioner. Four greenhouse experiments were conducted for this purpose during the period from 1992 to 1994. Results obtained in this study show that seed soaking method could have a promising effect, as:

1: Soaking wheat seeds in Agar solution of 0.2 % significantly increased dry weight of plants grown on sandy soils by about 73 % over that obtained from control. A significant considerable enhancement effect for its combination with the three microelements Fe, Mn and Zn at 0.2% of each in chelated form was obtained (129% over control), while the response to these nutrients alone was little. However, there was a relatively slight response to either Agar, micronutrients or their combination in clayey soil.

2: Soaking cotton seeds in Agar solution of 0.2 % significantly increased the response of seed cotton yield to N fertilizer in clayey soil.

3: Soaking wheat seeds in Agar solution of 0.1% increased the efficiency of the biofertilizer Cerealin with wheat plants grown on sandy and calcareous soils, as their dry weight increased by 18% and 25% respectively over that obtained from Cerealin alone.

On the other hand, application of Agar solution to a calcareous soil as anticrusting agent at the rate equivalent to 0.025 % of soil weight, slightly improved the yield of sudan grass. Increasing such rate to 0.05% depressed the yield, indicating that these rates were much higher than that should be used with Agar, a result of economical importance.

### **INTRODUCTION**

At present time, synthetic soil conditioners can be included as biotechnology and as a major mean of increasing productivity in the agriculture sector. Soil conditioners, if used according to the established procedures, may increase water infiltration into soil containing clay, prevent soil crusting in calcareous soils, (Wallace and Nelson 1986), in-

crease water holding capacity, enhance nutrient efficiency and encourage soil microflora in sandy soils(Azzam *et al.* 1987).

However, the global demand for clean agriculture free from chemicals, like fertilizers, pesticides, synthetic soil conditioners, etc., is of much concern at moment, to protect the ecosystem from their adverse effects. Hence, natural polysaccharides are the most effective agents in stabilizing soil organic matter and clay aggregates; many researchers have used them as soil conditioners such as Wallace, 1986 and Ben Hur and Letey 1989). However, their use is so expensive, because they are biologically degradable and thus higher rates are needed for soil improvement. Wallace, (1986) noted that the excellent soil conditioning effect of the polysaccharide gaur, derived from gaur bean, disappeared within three weeks. Like wise, soils have been inoculated with micro-algae, to produce polysaccharides, as had been reported by Metting and Rayburn, (1983). Additionally, the same authors found that the effect of pure preparation of extracellular polysaccharides of the micro algae *C. mexicana* as comparable to the synthetic polymer Kriliun. Such situation prompted us to use the biopolymer, Agar as soil conditioner. It is a dried hydrophilic colloidal substance extracted from red algae belonging to Rhodophyceae. It consists chiefly of the calcium salt of the sulfuric acid ester of a linear polyglactose (Reynolds and Prasad, 1982). In contrast to the other natural polysaccharides, it is undegradable biologically, as its conditioning effect can last much longer. Additionally, it has a stabilizing and gellifying power. Because of these unique properties, it finds wide applications in the areas of biotechnology, microbiology, pharmacology, confectioners and where water retention is of importance. But in field of agriculture, it may be the first time to be used economically and practically. However, Abdel-Halim *et al.* (1994) found that Agar material showed a high potential as a new soil conditioner in sandy soil, when it was applied as a dry powder at the rate of 1.5g/kg soil, (equal to 1500kg/feddan) that is not accepted commercially. With seed soaking technique described in the present study, it should be possible to use only 0.1 kg of dry powder to prepare Agar solution of 0.1 or 0.2% that is sufficient for soaking seeds required for sowing one feddan.

## MATERIAL AND METHODS

The lack of information on using Agar as a soil conditioner prompted initiation of intensive work in greenhouse for three years during the period from, 1992-1994 (Part I) followed by four field trials through the period from, 1997-2000 (Part II ) to evaluate using Agar as a soil conditioner at the economic rate using an easy technique. However, it is to be mentioned that other tests were carried out at laboratory to help dissolving Agar material in water by acidifying the suspension with citric acid as had been described by Wallace *et al.* (1986) with gaur polysaccharide, but this technique failed to increase Agar solubility. Thus the most suitable technique was to prepare Agar stock solution of 10g/L of water, using heating to help dissolving, then diluted at the different required rates. Also, other pilot experiments (not reported here) were conducted in greenhouse using the high rates of synthetic polymers, suggested by El-Hady and Abou Saif, 1984; Wallace *et al* (1986) and Khadr *et al* (1988), or the natural polysaccharide, gaur, used by Wallace (1986). All of these approaches used amount much higher than should be used with Agar, a result of extreme practical importance.

### Greenhouse experiments :

Four greenhouse experiments were conducted to study the effect of Agar as a soil conditioner on:

1. **Response of wheat plant (*Triticum aestivum.c.v.Giza 164*)** to micronutrient application in sandy and clayey soils (of E.C 0.28 and 0.42 mmohs/cm in 1:5 soil water extract and pH 7.8 and 8.1 in 1:2.5 soil water suspension respectively) in 1992.
2. **Response of seed cotton yield (*Gossypium barbadnse c.v.Giza 75*)** to N fertilizer in clayey soil (E.C 0.24 mmohs/cm and pH 8.1) in 1993
3. **Efficiency of using the biofertilizer cerealin in wheat plant, *Triticum aestivum.c.v.Giza 164***, grown in sandy and calcareous soils (E.C 0.28 and 0.77 mmohs/cm in 1:5 soil water extract and pH of 7.8 and 7.9 in 1:2.5 soil water suspension respectively ) in 1993

**4. Growth of sudan grass (*Sorghum halepensis*,c.v. Giza 142) grown on calcareous soil, ( E C 0.16 mmohs/cm 1:5 soil water extract and pH of 7.9 in soil water suspension) in 1994**

**1. Effect of Agar on the response of wheat plant to micronutrient :**

In this experiment, Agar was applied by soaking seeds in Agar solution of 0.2% (2g/L) with the aim of coating seeds with a thin gel layer of Agar alone or Agar containing different micronutrients. Such technique may be similar to that developed by Hoyle (1983 a & b)[as cited by ( Wallace and Wallace (1986)], who applied spots of solution to the soil with each seed. Seeds of wheat were soaked for four hours in the following different solution :

1. Water (control)
2. Nutrient solution containing the three chelating micronutrients, Fe, Mn and Zn at the concentration of 0.2% of each (N sol.1)
3. Nutrient solution containing the five micronutrients, chelated Fe, Mn, Zn and Cu. of 0.2% of each, and B of 0.2% as boric acid (N sol.2)
4. Agar solution of 0.2%
5. Agar solution of 0.2% containing the previous mentioned three micronutrient (Nsol.1)
6. Agar solution (0.2%) containing the previous mentioned five micronutrients (Nsol.2)

Ten soaked seeds were planted in sandy and clayey soils placed in clay containers with 2kg soil/pot. P and K fertilizers were added to all treatments at rates of 15kg  $P_2O_5$  and 24 kg  $K_2O$  / feddan respectively, while, N was added in irrigation water at the rate of 50 kg N/feddan after ten days from planting. The treatments were replicated three times in a completely randomized block design. After 60 days from planting, fresh and dry weights were recorded.

**2. Effect of Agar on the response of seed cotton yield to N fertilizer.**

In this study, cotton plant was grown in a clayey soil in pots with 7 kg soil each. Four treatments were tried: soaking seeds for four hours in water (control) or in Agar solution of 0.2% under two levels of N fertilizer, 50 and 75 kg/ feddan as urea. P and K fertilizer were applied at either 15 kg  $P_2O_5$  or at 24 kg  $K_2O$  as super phosphate and po-

tassium sulphate, respectively, preplanting. Treatments were replicated three times in a split plot design. At harvest, seed cotton yield was determined (g/pot).

### **3. The effect of Agar on the efficiency of the biofertilizer Cerealin.**

In this study, two soils were used: sandy and calcareous soils. Seeds were soaked for four hours either in water, then coated with Cerealin, or in Agar solutions of either 0.1% or 0.2% then coated with cerealin. A control treatment was also included, wherein seeds were soaked in water. All treatments received the balanced compound commercial fertilizer named Foliar-x at the rate of 0.2% in irrigation water, once after planting. It contained 10%N, 7% P and 8% K, 2500ppm from each of chelated Zn and Fe and 3000ppm of Mn in addition to traces of Mg, S, B and Cu. Thus there were 4 treatments replicated three times in a completely randomized block design. Fresh and dry weights of plants were determined after 45days from planting. It may be worth to mention that the biofertilizer Cerealin is an inoculant for cereal crops containing *Bacillus polymyxa* as N<sub>2</sub> fixing bacteria in addition to phosphate dissolving bacteria (mon-culture) produced by General Organization for Agriculture, Egyptian, Ministry of Agriculture, Egypt; the Foliar.x fertilizer was produced by El Nasr Co. for chemicals and pesticides, Cairo.

### **4. Effect of Agar on the growth of sudan grass plant grown on calcareous soil.**

In this study, agar was used as anticrusting agent. The viscous liquid Agar could evenly coat soil aggregates and hold them together during emergence of seedlings. Four kg of calcareous soil were placed in clay containers samples of twenty homogenous Sudan grass grains were sown in containers, then the appropriate solutions containing Agar at the rates of either 0.025 or 0.05% of soil weight were poured over the soil in the containers. A control treatment without Agar was also included. The volume of the solution was adjusted to match the water holding capacity of the soil. Superphosphate and potassium sulphate were added before planting to all treatments at the rate of 30 kg P<sub>2</sub>O<sub>5</sub> and 24 kg K<sub>2</sub>O/ feddan, respectively, (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub> at the rate of 30 kg N/ feddan was added in irrigation water after ten days from planting.

The treatments were replicated three times in a completely randomized blocks. After 45 days from planting, no. of plants as well as fresh and dry weights of plants per pot were determined.

## RESULTS AND DISCUSSION

As previously mentioned, four greenhouse experiments were conducted to evaluate the ability of Agar biopolymer to act as a soil conditioner. The obtained results are presented and discussed as follows:

### 1. Effect of Agar on response of wheat plant to micronutrients

fresh and dry weight data presented in Table 1 emphasize the significant favourable effect of Agar on growth of wheat plants grown in sandy soil. Soaking seeds in Agar solution of 0.2% increased fresh and dry weight by 48.4 and 75.2% over control, respectively. Similar results were obtained by El-Hady *et al.* (1981), Khadr *et al.* (1988), Sadek (1998), using hydrogel synthetic soil conditioners, Wallace(1986) and Abdel-Halim *et al.*(1994) using the natural biopolymer, gaur and Agar respectively. However, those investigators applied dry polymers to the soil at higher rates, in the range of 0.5,1.0 and 2g/kg of soil (1120,2240, and 4480 kg/ha), which were not accepted in commercial practice at all. On contrary, by seed soaking technique used in the present study, only100g of Agar powder/ feddan was used to prepare soaking solution in addition of making application task much easier.

It seems that soaked seeds were coated with a thin layer of Agar that preserved water during the emergence of seedlings and, in turn, enhanced plant growth.

Further investigation of data recorded in the same table show that treating seeds with Agar not only increased the water use efficiency, but also improved micronutrient use efficiency in sandy soil. Soaking seeds in either Nsol1 or Nsol2 increased plant dry weight by about 19.3 and 83.7% over control, respectively, due to the inherent nutrient deficiency in sandy soils. Adding Agar to Nsol1 and Nsol2 improved their efficiency to achieve 128.9 and 105.4% over control respectively. It appeared that Agar with its gellifying and stabilizing power retained micronutrients around the seedlings against leaching loss. Such retaining effect might lead Nsol2 to be less effective

than Nsol1 in contrary to their use without Agar.

Table 1. Effect of Agar applied by seed soaking technique on growth of wheat plant in sandy and clayey soils (g/pot).

Treatments	Sandy soil				Clayey soil			
	Fresh wt.		Dry wt.		Fresh wt.		Dry wt.	
	g/pot	Inc. %	g/pot	Inc. %	g/pot	Inc. %	g/pot	Inc. %
1-Soaking in water (control)	15.97	0	3.74	0	22.9	0	5.28	0
2- Soaking in N Sol.1	23.13	44.8	4.46	19.3	26.1	14	6	13.6
3- Soaking in N Sol.2	27.3	70.9	6.87	83.7	26.5	15.7	6.16	16.7
4-Soaking in Agar Sol.	23.7	48.4	6.45	72.5	26.2	14.4	6.03	14.2
5-Soaking in Agar+ N Sol <sub>1</sub> .	30.8	95.1	8.56	128.9	25.8	12.7	5.93	12.3
6- Soaking in Agar+ Nsol <sub>2</sub> .	27.5	72.2	7.68	105.4	26.4	15.3	6.1	15.8
L.S.D 0.05	6.41		2.09		2.92		N.S	

Inc%= increase% over control wt = weight

Comparing further the effect of agar on wheat grown in clayey soil with that in sandy soil in the same table, showed that it was much less effective in clayey soil. Such result was expected because clay colloids act like Agar in conserving water and nutrients; however, a result of much concern in this table was the superiority of wheat plant in sandy soils under the treatment Agar plus Nsol1 and Nsol2 to those grown in clayey soil, indicating that applying Agar with this easy technique could render sandy soil to a productive one. Sadek *et al.* (1998) obtained the same result with sudan grass using RAPG.

## 2. Effect of Agar on the response of seed cotton yield to N fertilizer in clayey soil.

As indicated in Table 2, increasing the rate of N fertilizer from 50 to 70 kg N/ feddan led to a decrease in seed cotton yield. Thus 50kg N/feddan could be relatively considered the optimum rate under this experimental conditions. At such rate, applying Agar by seed soaking method significantly increased the yield by 15.7% over 50kg N alone. Such improving effect can be ascribed to the enhancement effect of Agar on plant growth at earlier stages of growth.

Table 2. Effect of Agar applied by seed soaking technique on the response of seed cotton yield to N-fertilizer in clayey soil.

Treatments		Seed cotton yield	
N kg/fed	Soaking seed	g / pot	Increase %
5 0	in water (control)	16.42	0.0
	in Agar sol 0.2%	18.99	15.7
7 0	in water (control)	15.00	0.0
	in Agar sol 0.2%	16.10	7.3
L.S.D. 0.05 N		N.S	
Soak		0.335	
N x Soak		N.S	

Fed = feddan = 4200m<sup>2</sup> Sol = solution Soak. = soaking

### 3. Effect of Agar on the efficiency of the biofertilizer Cerealin:

It is obvious from data, presented in Table 3, that Agar increased the efficiency of cerealin in both sandy and calcareous soils, especially at the lower concentration(0.1%) wherein the increase in dry weight amounted to 21.5 and 27.0% respectively over the control. However, such increase did not reach the L.S.D. It seems that Agar furnished a suitable media for N<sub>2</sub>-fixing bacteria in Cerealin to give a sufficient effective biomass. Meanwhile, it saved them from washing away by irrigation due to its gellifying and stabilizing power, and being undegradable biologically as well. Azzam *et al.* (1987) came to the same conclusion using the hydrogel synthatic soil conditioner RAPG with peanut.

Table 3. Effect of Agar applied by seed soaking technique on the efficiency of biofertilizer cerealin with wheat plant in sandy and calcareous soils.

Seed treatments	Sandy soil				Calcareous soil			
	F. wt.	Inc.%	D. wt.	Inc.%	F. wt.	Inc.%	D. wt.	Inc.%
1-Control	9.11	0	1.49	0	11.22	0	1.89	0
2- Cerealin	10.09	10.8	1.53	2.7	12.26	9.27	1.92	1.6
3- Cerealin +A <sub>1</sub>	11.14	22.3	1.81	21.5	13.06	16.4	2.4	27
4- Cerealin + A <sub>2</sub>	10.03	10.1	1.51	1.3	13.47	20.1	2.15	13.8
L.S.D. 0.05	1.975		N.S					

A<sub>1</sub> = 1g Agar/L, A<sub>2</sub>= 2g/L F. wt. = Fresh weight, D. wt. = Dry weight  
inc. % = Increase % over control



#### 4. Effect of Agar on sudan grass grown on calcareous soil.

In this study, agar was used as anticrusting agent, because of its stabilizing power that could keep soil aggregates stable by holding them together better during wetting. Data in Table 4 show that treated soil with Agar solutions at the concentration of 0.025% of soil weight slightly improved the growth of sudan grass. On the other hand, soil treating with 0.05% Agar not only hinder the emergence of seedlings but also inhibited the growth of plants as compared to the control. Furthermore, visual examination of soil surface in pots showed a formation of a dry thin layer of Agar on the soil surface after three days from planting especially at the higher concentration.

Table 4. Effect of Agar applied in solution to calcareous soil on the growth of sudan grass plant.

Agar treatments	No. plants	Fresh wt.		Dry wt.	
		g/pot	Inc. %	g/pot	Inc. %
1- Control	15	112.8	0	29.6	0
2- 0.025% of soil wt.	12	110.8	-1.8	31.6	6.8
3- 0.05% of soil wt.	11	81.1	-28.1	19.33	-34.7
L.S.D. 0.05	1.997	7.394		1.714	

Inc.= Increase over control      wt.= weight

### CONCLUSION

The obtained results indicated that the biopolymer Agar proved to be a relatively promising new soil conditioner especially in sandy and calcareous soils. The simple and easy technique of soaking seeds in Agar solution of 0.2% used in the present study, gives it potential commercial use. It competes with other soil conditioners because of its vegetal nontoxic origin, red alga, in addition to its gellifying and stabilizing power and its resistance to any microbial degradation.

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## استخدام البوليمر الحيوي، آجار كـمـحسـن للتربة الجزء الأول - تجارب الأخص في الصوبة

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أقيمت أربع تجارب أخص في الصوبة لتقييم استخدام مادة الآجار المستخرجة من الطحالب الحمراء كمحسن للتربة نظرا لخواصها الطبيعية كمادة غراواتية غير قابلة للتحلل.

وقد أوضحت النتائج أن طريقة نقع البذور قبل الزراعة في محلول الآجار بتركيز ٠.١ أو ٠.٢ ٪ لمدة أربع ساعات قبل الزراعة أثبتت أنها طريقة فعالة.

فقد أدى نقع تقاوي القمح قبل الزراعة في محلول الآجار ٠.٢ ٪ إلى زيادة مؤكدة في وزن المادة الجافة لنبات القمح المنزرع في الأراضي الرملية بمقدار ٧٣ ٪ عن معاملة الكنترول . وقد زادت هذه النسبة إلى ١٢٩ ٪ عندما أضيفت عناصر الحديد والمنجنيز والزنك في الصورة المخيلية بنسبة ٠.٢ ٪ من كل منها إلى محلول الآجار بينما كان تأثير هاتين المعاملتين أقل كثيرا في حالة الأراضي الطينية.

كذلك أدى نقع بذور القطن في محلول الآجار ٠.٢ ٪ إلى زيادة مؤكدة في معدل الاستجابة للتسميد النيتروجيني عند زراعته في الأراضي الطينية.

كما أدى استخدام الآجار بمعدل ٠.١ ٪ لنقع حبوب القمح مع تلقيحها بالمخصب الحيوي السيراليين إلى زيادة فاعلية السيراليين عما لو استخدم بمفرده سواء في الأراضي الرملية أو الجيرية.

كان لاستخدام الآجار لمنع تكوين القشرة السطحية في الأراضي الجيرية تأثير على زيادة نمو نبات حشيشة السودان عند إضافته إلى التربة على هيئة محلول بمعدل ٠.٠٢٥ ٪ من وزن التربة بينما أدى استخدامه بمعدل ٠.٠٥ ٪ إلى تشبيط نمو حشيشة السودان مما قد يشير إلى إمكانية الحصول على نتائج أفضل عند استخدامه بمعدل أقل من ذلك كثيرا ولعل هذا يعتبر مهما من الناحية الاقتصادية.