Evaluation of the biostimulant activity of the seaweed extract *Sargassum fluitans* Børgesen (Sargassaceae) on germination and growth parameters of *Arachis hypogea*.

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Abstract: The objective of the present work is to evaluate the biostimulant activity of seaweed extract *Sargassum fluitans* Børgesen (Sargassaceae) on germination and growth parameters of arachis hypogea. The preparation of this extract consists in macerating 100 g of vegetable powder in 1 liter of sterile distilled water using a Blinder Blender. To evaluate the effect of the Total Aqueous Extract (E.T.A) on the germination of peanut seeds, the device put in place is completely randomized and consists of 3 repetitions. Five (05) objects are compared, these are: the batch of control bags, no solution is added except distilled water. In the other batches, 200 ml of solution + 5 mg / l (ETA1), 10 mg / l (ETA2) and 15 mg / l (ETA3) of extract powder are respectively made during a watering every two days. Analysis of the results indicates that seaweed extracts stimulate seed germination and groundnut leaf development. The treatment ETA3 with the dose 15 mg / ml had a better rate with 100% of germination. Treatment ETA2 with a dose of 10 mg / ml of solution and treatment of ETA1 with a dose of 5 mg / ml of solution had a germination rate of 80%. As for the control plant, it had a germination rate of 32%. The mineral composition of Sargassum fluitans extract could be used as a biostimulant in peanut culture.
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Keywords: Biostimulant, Total Aqueous Extract, *Sargassum fluitans*, seaweed, *Arachis hypogea*, Côte d'Ivoire.

Introduction

Since 2011, the coastal populations of Côte d'Ivoire regularly experience the massive influx of seaweeds, especially the *Sargassum natans* and *Sargassum fluitans,* which disturb socio-economic activities such as tourism, fishing and the hotel business. Innumerable initiatives carried out so far to address this scourge have failed since the phenomenon of stranding keeps increasing every year. Therefore it is necessary to explore other ways including that of valorization. Various studies revealed that seaweeds are largely used in various areas like human diet, extraction of phycocolloïds, pharmaceutical industry, cosmetics and above all in farming (**Elmtili *et al*., 2013**). In fact, seaweeds have been, for ages, used in coastal areas as soil fertilisers. Initially used in full, as organic compost, seaweeds are more and more used as liquid extracts (**Louvieaux, 2004**). Biostimulants have already been developed from natural extracts from seaweeds (*Aschophylumm nodosum* and *Sargassum johnstonii*) which contain much cytokinin and essential for plant nutrition (**Klarzynski *et al*., 2006**). Recent studies on the effect of seaweeds extracts have shown a better germination, flowering and fructification. As for (**Sivasankari *et al*., 2006**), they show that, besides the parameters mentioned above, a great seedling vigour, meaning that the stem was longer and the roots strong, due to the increase in dry and aerial biomass. Are those effects, as described, reproducible with the extracts of *Sargassum fluitans*? This study aims at providing an answer to that question. The objective of the present study is to evaluate the biostimulant activity of the extract of seaweed *Sargassum fluitans* Børgesen (Sargassaceae) on the germination as well as the growth parameters of the *arachis hypogea.*

I. Material And Methods

I.1. Material

The material used to collect and analyse the data consist of plant material and laboratory technical material besides the culture substrate.

The plant material consists of species of seaweed (*Sargassum fluitans*), see **Picture 1** and of peanut seed (*Arachis hypogaea*).

The Technical material consists of sampling equipment and of preprocessing of extraction, and the equipment for triphytochemical.

Picture 1: *Sargassum fluitans*

I.1.2. Characteristic of the culture substrate

Our culture substrate is from the soil, with sandy texture, collected at the National Floristic Center (CNF) of the University Felix Houphouët Boigny of Abidjan. The characteristics of the soil are reported in Tables I and II.

I.1.1. The Technical Material

I.2. Methods

I.2.1. The collect of seaweeds

The seaweeds were collected on the beach of the Atlantic Ocean in the city of Grand Bassam. The ones which landed on the beach were picked up, washed with sea water, put into plastic bags and taken to the laboratory.

Table I: Physical Characteristics of soil samples collected at 0-20cm

Size Distribution (%)					
Soil of CNF	Clay	Thin silt	Coarse Silt	Thin Sand	Coarse Sand
	10	1.5	1.75	22.05	64.7

Source: Gnimassoum, 2016.

Table II: Chemical characteristic of the soil samples collected at 0-20cm

pH		**Organic Material (mg/Kg)**					**Exchangeable Cations (mg/Kg)**				**Saturation Rate**	
pHeau	pHkcl	P	C	N	MO	C/N	K+	Ca++	Mg++	Na+	CEC	S/T
6.2	5.3	900	14700	1300	25284	11	0.06	1.342	0.4	0.06	6.56	28.31

Source: Gnimassoum, 2016.

I.2.2. Preparation of the Total Aqueous Extract (ETA)

The preparation of that extract was carried out by means of the method described in (***Zirihi et al., 200***7); which consists in macerating 100g of plant powder in **1** liter of distilled water using a blender Blinder. In order to optimise the yield in extract, the maceration was carried out till the residue went out. The homogenate is filtered with cotton wool then on Whatman filter paper 3 mm. The resulting aqueous filtrate is vaporised through an incubator at 50°C to obtain a powder that makes up the Total Aqueous Extract (ETA).

I.2.3. Determining the doses of the various processings

Various successive concentrations are tested in order to determine the most appropriate dose for a better germination and growth of the peanut seeds. Thus, the various concentrations applied for each extract are the following: 200 ml of solution 5 mg/l, 10 mg/l and 15 mg/l.

I.2.4. The experimental Device

The device we used, is a device in total randomization including, as a factor, the Total Aqueous Extract applied at three levels (doses) or processings. Each processing is repeated three times.

I.2.5. Installation and the maintenance of cultures

The tested product on the grains of peanuts is the Total Aqueous Extract of the seaweeds *Sargassum fluitans*. Good quality seeds of peanut with germination potential, are carefully cleaned in a volume of sterilised distilled water and put to germinate in polyethylene bags filled with sand, sterilized in an oven at 50°C, at a rate of 5 grains per bag. No solution was added into the batches except distilled water. In the other batches, 200 ml of solution of 5 mg/l, 10 mg/l and 15 mg/l of extracted powder are brought in every two days respectively during the watering. For every processing, 3 repetitions are carried out. The test is carried out under semi controlled conditions in a greenhouse located in the Scientific Center of the university Felix Houphouët Boigny located in Bingerville. That greenhouse is submitted to the influence of the temperature and the cleaning of the environment. In average, the temperate inside is 26.4°C.

After the time frame allowed for the germination; i.e 8 days, a clearing was carried out to eliminate the vigorous seedlings and to let only one plant grow in each bag. Two days following this clearing, growth parameters measurements were started, and done once a month.

I.2.6. Parameters measured during the study

Two key parameters of the development cycle of the plant are measured: They are all about the germination (TG) and the development of the leaves of peanuts.

- **The Germination rate (TG)**

It all about the number of seeds that germinated out of the total of the seeds sown (**Côme, 1970**) and calculated using the following formula: $\boldsymbol{TG = \frac{n}{N} * 100}$;

n= number of seeds that germinated, N = Total number of the seeds tested;

- **The number of leaves**

The height is measured from the noose till the closing bud using a graduated ruler. The measurement of the height concerns just the growth phase of the plant as well as the number of leaves.

II. Results

II.1. Yield of the extraction

The aqueous extraction of 100 g of the powder of *Sargassum fluitans* gave a weight that is equal to 4.72 g. The extraction, which was repeated three times, helped obtain an average yield that is equivalent to 4.72±0, 64%.

II.2. Effect of the ETA on the agronomic parameters of the peanut

II.2.1. The Germination rate

Figure 1 highlights the various germination rate obtained in the various processings. Processing ETA3 with the dose of 15 mg/ml had 100 % as germination rate. Processing ETA2 with a dose of 10 mg/ml of solution and processing ETA1 with the dose of 5 mg/ml of solution had 80 % as germination rate. As for the Control Processing the germination rate was 32 %.

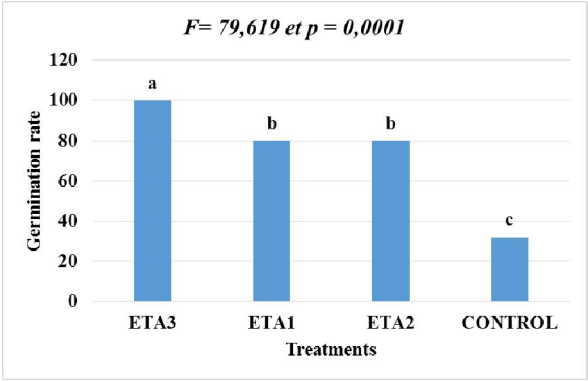

Figure 1: Change in the germination rate according to the various processings

II.2.3. Evaluation of the number of leaves according to the processings

With the Control Processing, the number of leaves varied from 4.22 to 13. 55. With the processing ETA1 the number changed from 5.44 to 16.77. As for processing ETA2, the number of leaves changed from 5.88 to 18.44. As for processing ETA3, the number of leaves increased from 5.44 to 13.55 (**Table III**).

Table III: Effect of the processings on the number of leaves according to the observation periods.

Processings	D+11	D+18	D+25	D+32
ETA1	5.44^{a}	9.22^{b}	13.44^{b}	16.77^{b}
ETA2	5.88^{a}	11.22^{a}	15.44^{a}	18.44^{a}
ETA3	5.55^{a}	7.55^{c}	11.55^{c}	13.55^{c}
Control Processing	4.22^{b}	6.77^{d}	9.77^{d}	13.55^{c}

Eleven days after the sowing, processings ETA1, ETA2 and ETA3, presented in group **a** showed the same number of leaves. These three processings were statistically different from the Control Processing presented in group **b**. we note that, in the following weeks, a significant difference between the three processings (**Figure 2**). Moreover, we note a similar group during the last week of processing ETA3 and of the Control Processing. The comparative study on the various processings reveals an increasing change starting from the leaves of the first week to the fourth week for all the plants. Nonetheless, processing ETA2 had the strongest leaves, followed by processing ETA1, compared with the other two processings.

Caption: ETA1: 5 mg/ml; ETA2 : 10 mg/ml and ETA3 : 15 mg/ml of extracted solutions of seaweed. In the table, the lines with the letters (a or b) on the same column are not statistically different from the probability threshold α=5 % according to the Newman-Keuls test carried out.

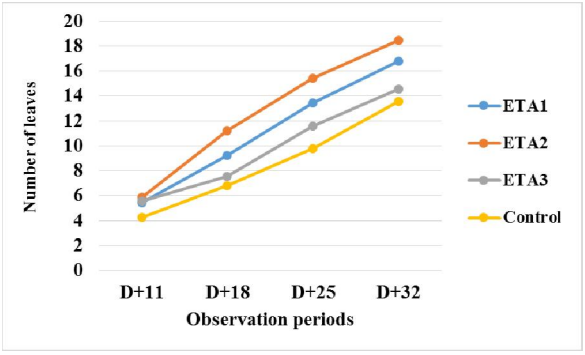

Figure 2: Change in the number of leaves according to the observation periods

III: Discussion

The seeds of peanut were sown in various culture environments. Compared with the Control environment (germination rate = 32%), the germination rate was 80% for the concentrations ETA1 and ETA2 and of 100% for the concentration ETA3. The Total Aqueous Extract in these environments rather than in the Control ones made the germination rate increase. This improvement must be due to the presence of soil nutrients, thanks to the extract applied. In fact, many positive effects are reported by **Sivasankari *et al*., 2006**; **Jolivet *et al*., 1991;**

Venkataraman *et al*., 1993; **Mohan *et al*., 1994;** El-**Sheekh** and **El-Saied, 2000** as for the use of seaweeds extracts in farming like the improvement of germination rate. **Sivasankari *et al*., 2006** showed an increase in the germination rate for the seeds of vine submitted to the extracts of *Caulerpa chemnitzia* (Esper) J.V. Lamououx (Caulerpaceae) and *Sargassum wightii* Greville (Sargassaceae).

All the processings stimulated the height growth of the peanuts. Comparatively to the Control Processing, processings ETA1 and ETA2 showed stronger growth on Day – 18 and on day – 25. That could be due to the presence of azote in the extract, knowing that it is the engine of the plant growth and contribute to the crop development of all the areal parts of the plant **(Ferreira and Lourens, 2002)**. The processings did not produce the expected impact on the height growth of the plant during the last week of experiment. Still, the Total Aqueous Extract of *Sargassum fluitans* used consists of nutrients that play many roles in the development of the plant. The lack of significant effect on the height growth of plants can be due to the saturation of the environment with the extract. In fact, for some biostimulants; like the seaweeds, it was proved that a high dose of product could have a phytotoxic effect, decrease the yields or even inhibit the microbial activity of the soils and reduce the growth (**Chen *et al*., 2002; Sivasankari *et al*., 2006**).

A significant increase in the number of leaves is noted in processings ETA2 and ETA1 in comparison with the Control Processing at the end of the experiment. That shows, the positive effect of the extract of *Sargassum fluitans*. The seaweed extracts are known for the ability to improve the physical properties of the soil, due to the fact that they contain trace elements which are involved in plant metabolism as the co-factor of many enzymatic reactions. The Total Aqueous Extract of the *Sargassum fluitans* contain nutrients which are important for the development of the plant. Seaweeds extract include a multitude of compounds (hormones, amino acid, micro elements); that implies a complex set of mode of actions that could justify the noted positive effects (improvement of the germination rate, foliar development).

Conclusion

The physico-chemical analyses revealed enough components with minerals contained in the extract of *Sargassum fluitans.* Minerals are essential in the development of cultures. This study has revealed that, the ETA of the *Sargassum fluitans*, increases the germination rate of seeds of peanuts and helps the foliar development of plants. As far as the height of the plants is concerned, the extract has revealed no effect.

Still, in order to bring more details about the effect of the ETA on peanuts, parameters such as content in water, the aerial dry biomass and chlorophyll content were measured. It appears that the plants processed with the *Sargassum fluitans* have averages superior to the Control Processing. Nonetheless, the statistics tests show no significant difference between the various processings.

The physicochemical composition of the extract of *Sargassum fluitans* makes for its use as biostimulant with the adults. But, it would be relevant to further the study till the total growth of the cultures in order to better notice the effects on all the phonological steps of the plant and use a great dose of extract. That would lead to revisit the extraction method in order to optimize the yield of extract; which is currently very low (4, 72±0, 64%). It would be relevant to compare the extract with reference products sold at the market in order to better rule on its effects.

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