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Full Length Research Paper

Exploring Allelopathic and Antifungal Properties of Padina pavonica (L.) Extract

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In this study, potential allelopathic of a brown alga Padina pavonica (L) was evaluated. Aqueous extracts of the alga obtained at room temperature / 24 h (E1), 50°C / 4 h (E2) and 100°C / 2 h (E3) were tested on the germination and early growth of crop plants and growth of three fungal strains: Fusarium graminearum, Penicillium expansum and Alternaria alternata. Also, a fractional, of the alga in three organic solvents with increasing polarity: hexane, chloroform and acetone, was carried out and estimated as well as that the seedling growth soil composition was estimated. Results revealed a perceptible allelopathic capacity of P. pavonica. Although the percentage of germination of seeds was not influenced or slightly stimulated compared to the control in the presence of the three kind extracts and the dry powder, root and shoot growth was clearly improved. Results varied according to extracts and the vegetable material dose. Extract prepared at room temperature (E1) was most favorable. Stimulation percentage varied between 15 and 78% for roots and between 1 and 67% for shoots. Fungal growth was strongly inhibited in the presence of extract E3 compared with E1. Thus, growth inhibition percentages were 95.83, 80.76 and 63.33% for F. graminearum, P. expansum and A. alternata respectively, in presence of E1, against 41.66, 72.30 and 51.66% in presence of E3. For organic extracts, the most spectacular seedling growth stimulations were recorded in the presence of the chloroform extract. This indicated that the active molecules had an average polarity. The incorporation of algal powder in the soil was shown very beneficial for the target species in particular with low dose (50 g/Kg). Moreover, the effect of the algal powder was similar to that obtained by the addition of chemical fertilizers. Finally, algal powder allowed a clear improvement of the chemical composition of the soil concerning its richness on calcium, magnesium and organic matter and did not affect its pH. Results might be considered very interesting, since molecules of this species could have a double effect in the crops: a fertilizing effect and an antifungal effect without deteriorating the physico-chemical properties of the soil.

Key words: Anti-fungal activity, aqueous extracts, chemical fertilizers, fertilizing capacity, growth, *Padina pavonica*, seeds.

INTRODUCTION

Molisch (1937) was the first to define the term allelopa-thy, in a broad sense to describe either positive or nega-tive biochemical interactions among all plant kinds. Rice (1984) included microorganisms and restricted the con-ceptual content of allelopathy exclusively to negative effects arising by the production and excretion of chemi-cal compounds from plants and microorganisms. Many plants proved to have the allelopathic potentials; generally they had negative effects, and could be used in biological pesticide production. Majority of works present the harmful effects of plant extracts and rare were works which indicate beneficial effects of plants. Marine plants and macro-algae constitute a richness to explore and exploit in several regions of the world (Ben Said et al., 2002). Recently, Inderjit and Dakshini (1995) gave an overview of allelopathic activities in aquatic habitats with particular emphasis on algae. Allelopathy is a prevalent natural phenomenon in aquatic ecosystem; however, it is difficult to study its effects among aquatic organisms under natural conditions because factors such as nutrient

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and light competition, temperature and pH change could totally mask allelopathic effects (Keating, 1977).

Padina pavonica (L.), a brown alga, was widespread in Tunisian littoral. It belonged to the family Dictyotaceae, order Dictyotales, subclass Isogeneratae, class Phaeophyceae (Kamenarska et al., 2002). Studies conducted on this specie were particularly interested in the nature and rate of carbohydrate, lipids, vitamins, mineral salts and other active ingredients (Al Easa et al., 1995; Wahheb, 1997; Ktari and Guyot, 1999; Kamenarska et al., 2002). The richness of this specie let think of its ex-ploiting for a positive purpose on the crops. Indeed, the over use of synthetic agrochemicals often caused envi-ronmental hazards, imbalance of soil microorganisms, nutrient deficiency, and change physicochemical proper-ties of soil, resulting in a decrease of crop productivity. So, incorporation of allelopathic substances into agricul-tural management might reduce the use of industrial pesticides and fertilizers and lessen environmental deterioration (Chou, 1999).

In this chapter, assessment of fertilizing and antifungal potentialities of brown algae *P. pavonica* (L.) was investigated.

MATERIALS AND METHODS

Algal sample collection

The tested macro-algae (*P. pavonica*) was collected from Tunisian littoral (Mahdia, Tunisia) (35°30'N, 11°04' East) in July 2007, identified and carefully washed with distilled water immediately to remove attached remains and organisms.

Aqueous extracts

Fresh tissue of macro-alga was air dried for 6 days at room temperature and then ground into a fine powder using a mortar. Thirty grams of dry powder was mixed with 1000 ml distilled water (Einhellig et al., 1993) and shaken for i) 24 h at room temperature (E1) (Khanh et al., 2005), ii) 4 h at 50°C (E2) (Delabays et al., 1998) and iii) 2 h at 100°C (E3) (Mao et al., 2006). The mixture was filtered through a filter paper several times and kept at 4°C in the dark for further use.

Organic extracts

Sequential extraction was carried out in organic solvents with rising polarity: hexane, chloroform and acetone. Forty grams of algal powder were immersed in the organic solvent for 7 days at room temperature. Organic extracts were evaporated to dryness under reduced pressure in a rotary evaporator at 45 - 50°C. The residue was weighed and yield was determined (0.153, 0.467 and 0.277% for respectively hexane, chloroform and acetone). The extracts were tested at concentration 3000 ppm in the biological assays.

Petri Plate assay

Effect of aqueous extracts: Aqueous extracts were tested on four crop species: Dicot species: *Lens culinaris* L. (lens), *Lactuca sativa* L. (lettuce), and Monocot species: *Triticum aestivum* L. (wheat) and *Hordeum vulgare* L. (barley). Lettuce was known to be very sensi-

tive to allelochemicals (Leather and Einhellig, 1987; Ervin and Wetzel, 2003), whereas other species were selected because they represent the first food source in Tunisia. All plant seeds were surface sterilized with 0.525 g/L sodium hypochlorite for 15 min. The seeds were rinsed four times with deionized water, imbibed in it at 22°C for 12 h and carefully blotted using a folded paper towel (Chon et al., 2005). Without any dilution, ten milliliters of each extracts, contained 0.3 g of algal dry powder, were pippetted onto filter paper. The seedlings watered with distilled water were used as control. Thirty imbibed seeds of target species were separately placed on the filter paper in Petri dishes. They were covered and placed in a growth chamber at 24°C during the 14 h light period and 22°C during the 10 h dark period. The plates were illuminated with

400 µmol photons.m⁻²s⁻¹ photosynthetically active radiation (PAR), provided by a mixture of incandescent and fluorescent lamps. Treatments were arranged in a completely randomized design with three replications.

Cumulative germination was determined by counting the number of germinated seeds at 24 h intervals over a 144 h period and transformed into germination percentage. Shoot and root length and dry weight of recipient species were measured on all seedlings in each Petri dish on day 7 after placing seeds on the medium. Data were transformed to percent of control for analysis. The inhibitory or stimulatory percent was calculated using the following equation given by Chung et al. (2001): Inhibition (-)/stimulation (+) % = [(extract – control)/control] x100 with: Extract: parameter measured in presence of *P. pavonica* extract Control: parameter measured in presence of distilled water.

Effect of organic extracts

The three extracts concentrated from hexane, chloroform and acetone were dissolved in acetone (15 mg in 2 ml) to estimate their effect on germination and early growth of crop species. Two controls were considered, one in the presence of distilled water and another one was in presence of acetone in order to eliminate the effect of organic solvent. Filter paper placed in Petri plates, were soaked in distilled water, acetone or various organic extracts. The solvents were evaporated for 24 h at 24°C. After that, 5 ml of distilled water were added (final concentration: 3000 ppm) and 30 soaked seeds were put to germinate for 7 days. Germination, shoot and root length and dry weight of target species were determined and expressed as percentage of the control.

Antifungal activity assay

The antifungal activity was tested on *Fusarium gramineaum, Penicillium expansum* and *Alternaria alternata.* These fungi were provided by the phytopathology laboratory of Higher Institute of Agriculture -Chott-Mariem in Tunisia.

100 µl of extracts at 100 µg/ml were put in dug wells (5 mm broad and 20 mm length) of sterilized Petri dished containing Potato Dextrose Agar (PDA). The fungal plugs (0.4 mm in diameter) were placed opposite the well with 1 cm of the edge limps. Limps control consisted distilled water in the well. After an incubation for 72 h at 24 \pm 2°C, the mycelium development of pathogenic fungi in each Petri dish and the phytotoxic effect of *P. pavonica* extracts by measuring the distance covered by the mycelium. The percentage of growth inhibition was calculated as follows (Khanh et al., 2005): I

(%) = (1- Cn/Co) * 100 Where Cn was the distance covered by fungi in the presence of extracts and Co was the distance covered in the presence of distilled water. The antifungal effect was measured under a totally random design with three replications.

Glasshouse assay

For incorporation treatment, biomass of P. pavonica was mixed with

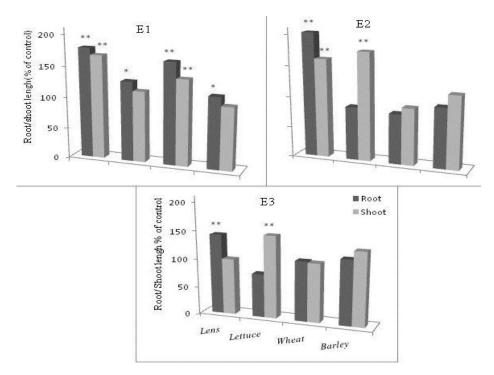


Figure 1. Root and shoot length of seedlings grown in the presence of aqueous extract prepared at room temperature (E1), at 50°C (E2) and at 100°C (E3). Differing statistically (Duncan test) from control are marked with one (P 0.05) or two (P 0.01) asterisks.

soil by vigorous shake in 20 cm diameter plastic bags. Four trials were carried out: soil without fertilization, fertilization with algal powder at two doses (50 and 100 g.kg⁻¹ of biomass (Khanh et al., 2005)) and fertilization with chemical fertilizer 0.5 g/Kg (Ammonitre). The pots were filled with the soil and residue mixture and twenty seeds per pot were planted. The growing medium was maintained near field capacity by sub-irrigation without nutrition solution (Fischer et al., 2000). The experiments were conducted in green-house for 15 days at 28 / 22°C day/night temperature. Germination percentage and seedling growth were measured as described previously. Data were transformed to percent of control for analysis.

Effect of P. pavonica residue incorporation on soil composition

After 15 days of germination, the mixture of soils and algal residue were analyzed for calcium, magnesium, carbon, organic matter and pH. Soil samples were air dried and then crushed. The sum of ions

 $(Ca^{2+} + Mg^{2+})$ was titrated with EDTA. The proportioning of calcium was determined by flame spectrophotometry and magnesium was calculated by the formula Mg (Méq/g) = ((Ca + Mg) – Ca). Carbon was proportioned by spectrocolorimetry at 590 - 600 nm and organic matter was calculated according to the formula: m.o. =1.72 x C (Aubert, 1978). The pH was measured by a pH-meter.

Statistical analysis

The laboratory bioassays and pot culture were conducted in a complete randomized design with three replications. Duncan – tests and ANOVA were performed on SPSS 13.0, for Windows program, to analyze treatment differences. The means were separated on the basis of least significant differences at the 0.05 probability level.

RESULTS

Effect of aqueous extracts on the germination and growth of test crops

The variance analysis showed that *P. pavonica* extracts had no significant effect on germination (P < 0.001). Indeed, germination percentage did not vary in presence of all extracts and kinetic was not influenced or slightly accelerated. However root and/or shoot growth was improved. This improvement was more significant when the seedlings were exposed to the extract obtained at room temperature (E1) and the stimulation percentage varied between 15 and 78% and between 1 and 67% for roots and shoots, respectively (Figure 1). Root growth was more or less inhibited in presence of the extract prepared at 50°C (E2) with a non significant respective reduction of 18.39, 13.23 and 1.75% for wheat, lettuce and barley, however, this extract improved growth of lens roots by inducing stimulation of 101.34%. The effect of the extract prepared at 100°C (E3), was marked on root growth of lens which was stimulated by 42.5%. For other species, a non significant difference compared to the control was recorded, except for the lettuce whose growth was inhibited by 23%.

Since, root growth of various target species was near to the control or was strongly stimulated in the presence of various kinds of *P. pavonica* aqueous extracts. The sensitivity of shoots varied according to target species and extracts (Figure 1). Globally, the behavior of shoots was

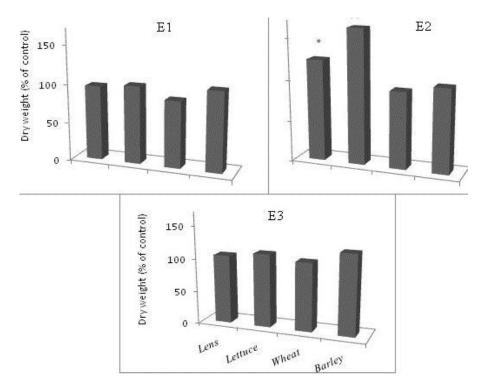


Figure 2. Biomass production by target species grown in presence of aqueous extract prepared at room temperature (E1), at 50°C (E2) and at 100°C (E3). Differing statistically (Duncan test) from control are marked with one (P 0.05) or two (P 0.01) asterisks.

similar to that of roots except for the lettuce which showed a better growth of shoots in the presence of E2 and E3. Shoot growth of the various target species was near to the control or was stimulated by an average factor of 1.2 in the presence of several of *P. pavonica* (Figure 1).Biomass production was significantly ameliorated for lens and lettuce in presence of E2 and E3. For other species, it was similar to the control (Figure 2).

Effect of organic extracts on the germination and growth of test crops

To determine the chemical group to which bioactive molecules of *P. pavonica* extracts could be owned three organic extracts: hexane, chloroform and acetone. Organic residues were dissolved in acetone, which required an acetone control. Results showed that this solvent did not affect germination and growth and the recorded re-sults would be allotted to the allelopathic compounds. Germination of the four species was similar to control in all cases (data no shown). Nevertheless, results indicate a stimulation of root/shoot growth for all species and in presence of the three organic extracts (Figure 3). The most important stimulations were recorded in the pre-sence of chloroform extract which showed a rise of root growth of 5.17, 80.32, 108.67 and 121.67% for lens, lettuce, wheat and barley, respectively. Similarly, this extract improved shoot growth of seedlings and the percentage of stimulation varied between 13 and 164%

(Figure 3). Acetone extract ameliorated significantly the growth of barley.

The effect of organic extracts on biomass production was not significant (Figure 4). Indeed, the quantity of dry matter was near to the control or slightly higher in all cases. However, an improvement of the biomass produc-tion in lettuce was recorded in the presence of the three organic extracts with a better stimulation in the presence of the chloroform extract (23.23%). For water content, the seedling hydration was near to the control or was improved in the presence of various organic extracts (Figure 4). In the presence of hexane extract a respective stimu-lation of 105 and 243% for lettuce and lens. A stimulation of an average of 51% was recorded for the monocotyle-dons in the presence of the same extract.

Effect of residue incorporation of *P. pavonica* in soil on germination and growth

The initial bioassay was necessary and often used to evaluate the allelopathic potentialities of plant species (Chon et al., 2005; Kato-Noguchi and Tanaka, 2003); however, pot test was desired in order to indicate the effects that could be reproduced under natural conditions (Corrêa et al., 2008). Thus, the incorporation of algal powder to the soil was carried out to evaluate its effect on growth of test crops under natural conditions and to compare with those of the extracts. Moreover algal powder was compared with the chemical fertilizer used usually in

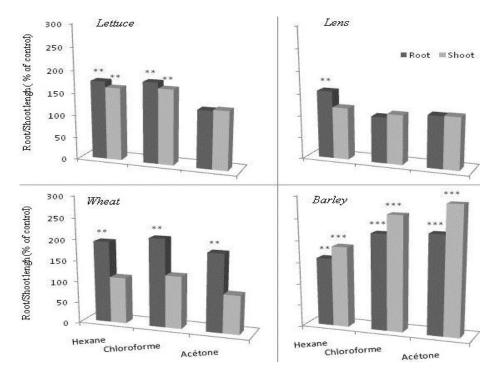


Figure 3. Root and shoot length (% control) of target species in the presence of three organic extracts. Differing statistically (Duncan test) from control are marked with one (P 0.05), two (P 0.01) or three asterisks (P 0.001).

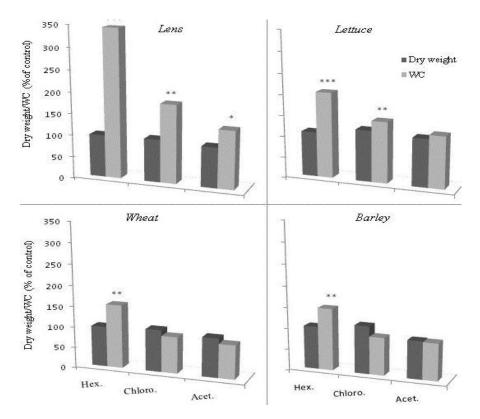


Figure 4. Biomass production and water content (% of control) of target species in the presence of organics extracts: hexane (Hex.), chloroform (Chloro.) and acetone (Acet.). Differing statistically (Duncan test) from control are marked with one (P 0.05), two (P 0.01) or three asterisks (P 0.001).

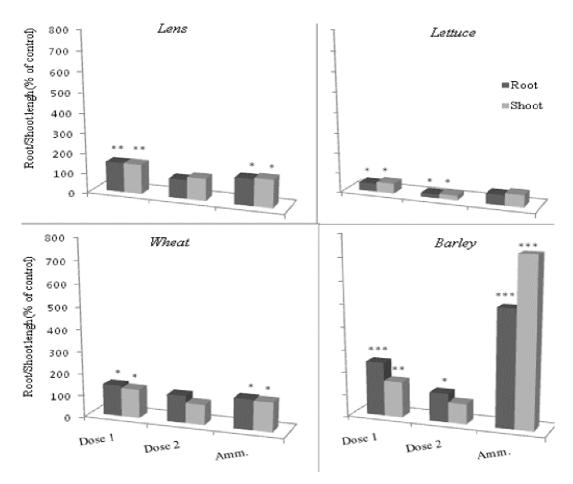


Figure 5. Effects of residue incorporation with peat, from thalli of *P. pavonica* at 50 g/Kg (dose 1) and 100 g/Kg (dose 2) and incorporation of industrial fertilizer (Amm.) on root and shoot length of test crops, 15 days after germination, expressed in % of control. Differing statistically (Duncan test) from control are marked with one (P 0.05), two (P 0.01) or three asterisks (P 0.001).

agriculture. Incorporation of dry algal powder in the soil gave similar results to those obtained by the incorpora-tion of a synthetic fertilizer (Ammonitre). Indeed, we have registered an average stimulation of 1.5 times of seedling growth in the presence of low doses of algal biomass or fertilizer (50 g/Kg) and a slightly inhibition (an average of 15%) at 100 g/Kg. An inhibiting effect of algal biomass was registered on lettuce growth especially at 100 g/Kg (Figure 5). Biomass production was significantly amelio-rated in presence of P. Pavonica residue at dose 1 for wheat, whereas similar results were obtained from other test crops and control (Figure 6). In the presence of dose 1, lens seedlings produced the same quantity of dry matter as the control, whereas in the presence of dose 2 and synthetic fertilizer, a reduction of 26.97 and 21.58% respectively. However, lettuce had slightly benefited from the algal matter at dose 2. The water content was near to the control for all target species except wheat which presented a hydration improvement in presence of algal matter at dose 1 and the lettuce whose water content significantly decreased in presence of dose 2 of the biological material (Figure 6).

Effect of residue incorporation of *P. pavonica* on soil composition

After 15 days of experiment, the chemical properties of soil with algal powder and with fertilizer were analyzed and compared. Algal incorporation did not influence the soil pH, which was comparable with the control and with that in the presence of chemical fertilizer. An average value of 8.3 in all cases (Table 1).

Concerning the nutritive elements, the quantity of calcium and magnesium which were very significant elements for the improvement of the soil texture were dosed. Results showed an increase in the quantity of calcium compared with the control in the presence of the algal biomass: 0.064 meq/g with dose 1 and 0.052 meq/g with dose 2. In the presence of industrial fertilizer the quantity of calcium as 0.09 meq/g. The important quantities of magnesium were also recorded in the presence of the algal powder compared with the control and chemical fertilizer. An increase of 0.114 meq/g and 0.176 meq/g in the presence of dose1 and dose 2 respectively was recorded compared with the control (0.0013 meq/g). The

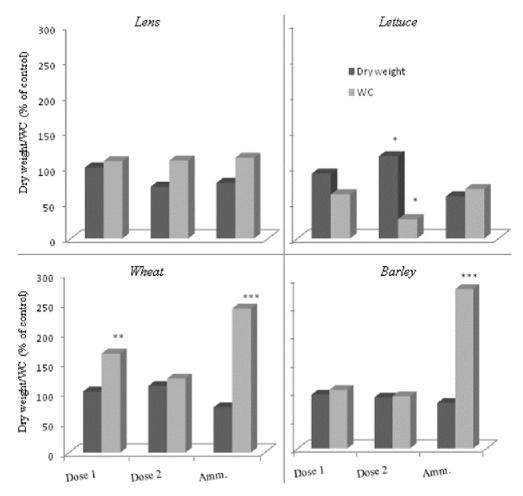


Figure 6. Effects of residue incorporation with soil, from thalli of *P. pavonica* at 50 g/Kg (dose 1) and 100 g/Kg (dose 2) and incorporation of industrial fertilizer (Amm.) on dry weight and water content (WC) of test crops, 15 days after germination, expressed in % of control. Differing statistically (Duncan test) from control are marked with one (P 0.05), two (P 0.01) or three asterisks (P 0.001).

Table 1. Contents in calcium, magnesium, carbon and organic matter and pH of							
the soil without (control) and in the presence of <i>P. pavonica</i> powder with two							
doses (50 g/Kg and 100 g/Kg) and of a chemical fertilizer (0.5 g/Kg)							
(Ammonitre), 15 days after germination.							

	Ca ²⁺ (Meq/g)	Mg ²⁺ (Meq/g)	carbon (%)	organic matter (%)	рН
Control	0.041	0.0013	0.37	0.6	8.3
Dose 1	0.064	0.116	0.57	1	8.4
Dose 2	0.052	0.178	0.65	1.1	8.3
Ammonitre	0.09	0.066	0.5	0.9	8.5

Fertilizer was of a less significant contribution (0.066 meq/g) compared with algal powder. Also, powder of *P. pavonica* improved the in organic matter (Table 1), which was essential to retain the nutritive elements and moisture in the soil as well as to nourish and shelter the organizations of the ground.

Antifungal activity of extracts

Fungicides are chemical compounds having toxicological properties, used by the farmers to fight against the phytopathogenic fungi. These fungicides neutralize and reduce the activity of fungi but they remain a worrying and

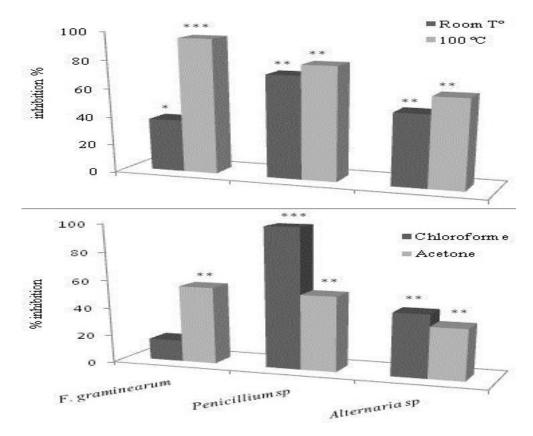


Figure 7. Effects of aqueous extracts (a) prepared at room temperature (Room $T^{\circ}C$) and at 100°C, and organic extracts (b) of *P. pavonica* on fungal growth. Differing statistically (Duncan test) from control are marked with one (P 0.05), two (P 0.01) or three asterisks (P 0.001).

frightening source of pollution and toxicity. To search for biological molecules with antifungal potential, *P. pavonica* extracts were evaluated on growth of three fungi strains: *F. graminearum, P. expansum* and *A. alternata.* Results revealed a strong toxicity of aqueous extracts on fungal growth (Figure 7). This toxicity was more significant when strains were exposed to the extract prepared at 100°C, thus the percentages of inhibition were 95.83, 80.76 and 63.33% for *F. graminearum, P. expansum* and *A. alternata* respectively (Figure 7).

The screening indicated that chloroform extract was most toxic for *P. expansum.* As shown by its complete inhibition. This was in contrast to *F. graminearum* whose growth inhibition was not significant in presence of this extract. Acetone extract presented an average inhibition percentage of 48% for the three receptive strains. The difference in behavior of the various fungal species indicated that various organic extracts did not contain the same molecules and those bioactive molecules with antifungal potentialities against *P. expansum* were extracted by chloroform and had a medium polarity.

DISCUSSION

Germination and growth of target species organs were not varied or more or less improved by *P. pavonica*

aqueous extracts. Differences in behavior of the organs and the target species could be allotted to the nature of allelochemicals present in extracts. Sanna et al. (2004) recorded a difference in sensitivity of micro-algae exposed to various types of filtrates of algae. Growth improvement of seedlings would be ascribable to the presence of bio-active substances of the growth like amino acids, nutriments, phytohormones, enzymes and vitamins released by P. pavonica thalli. Similar results were reported by Klarzynski et al. (2005, 2006) who recorded a growth stimulation of tomato and wheat plants by filtrates of Ascophyllum nodosum (brown alga). Morot-Gaudry (1997) reported that algal filtrates stimulated the enzymes responsible for minerals absorption, such enzymes responsible for minerals absorption, such reductase nitrate which was a significant element of plant nitrogen nutrition.

In presence of organic extracts, the most important stimulations were recorded in the presence of chloroform extract which showed a percentage stimulation reaching an average of 142% for the two target organs. Kamenarska et al. (2002) recorded that *P. pavonica* was rich in sterol and lipid compounds which were significant components of cellular membranes and responsible for a great number of cellular functions. The principal sterol of *P. pavonica* in Mediterranean area was fucosterol (24-hydroperoxy-24- vinyl-cholesterol) (Ktari and Guyot, 1999). Moreover, P. pavonica contained a weak concentration of toxic allelochemical compounds like terpenoids and phenolic acids (Kamenarska et al., 2002). Indeed, these compounds were known to have inhibiting effects on germination and growth of various plants (Vyvyan, 2001). This could explain root growth inhibition recorded in lettuce exposed to the extract prepared at 100°C. This extraction probably allowed the release of a sufficient quantity of allelochemicals which induced the inhibition known by its sensitivity to allelochemicals (Chon et al., 2005; Abdelgaleil and Fumio, 2007). However, stimula-tion of germination and growth of target species under various P. pavonica extracts would be due to the presence of the stimulating allelochemicals. In fact, number of chemical compounds of terpenoid group were identified like stimulating germination such as strigol isolated from cotton (Cook et al., 1972), and from mays (Yoneyama et al., 2004), the sorgolactone isolated from sorghum (Hauck et al., 1992), and alectrol and orobanchol from red clover (Yokota et al., 1998).

Biomass production and growth were significantly ameliorated in presence of P. pavonica residue at low doses and a slightly inhibition at 100 g/Kg. Growth inhibition could be attributed to a release of toxic compounds following a degradation of the algal matter by soil micro-organisms (Schmidt and Ley, 1999) or the oxidation by the soil (Ohno, 2001). On the other hand, Andre (1994) reported that plants which pushed in a mixed ground of marine algae had a faster growth than those which push-ed in a ground with a comparable quantity of industrial fertilizer. The richness of the algae in mineral matter, basic elements for plants nutrition was well documented. In addition, Klarzynski et al. (2006) reported that algae powder was rich in carbohydrates, mainly brown algae were shown rich in mannitol, which was able to activate reductase nitrate enzyme and to increase the chlorophyll content.

Analysis of the soil mixed with the algal powder showed that this biological material did not have any effect on the pH, whereas it strongly enriched calcium and magne-sium. It was well known that these two elements played a key role in the improvement of the soil texture. They returned the structure of the more movable and more stable ground, regularized the pH and supported the

exchanges of ions necessary to the nutrition of the plants and created a medium favorable to the useful microbes of the ground (Soliner, 1983). Moreover, the powder of *P. pa-vonica* enriched the soil organic matter, compared with the

control and with that enriched by chemical fertilizer. The richness of the organic matter soil was a great important character since it represented a source of nutriaments. In addition, they enhanced the soil permeability to water and air, ensuring its stability and being used of support for fauna and flora (Soliner, 1983).

Results revealed a strong toxicity of aqueous extracts on fungal growth and the screening indicated that chloro-form extract was most toxic. Similar results were recorded by Bennamara et al. (1999) who showed that the methoxybifurcarenone (molecule extracted from a brown alga *Cystoseira tamariscifolia*) had an antifungal activity against three pathogenic tomato fungal strains: *Botrytis cinerea, Fusarium oxysporum. F.* sp. *lycopersici* and *Verticillium alboatrum.* Also, Kamenarska et al. (2002) reported an antifungal activity of ethanolic extract of *P. pavonica* against *Candida albicans*

Conclusion

Our research showed that P. pavonica was rich in stimulating natural substances for test crops growth without causing damage to the soil. P. pavonica could be used as a natural fertilizer and would be used in the develop-ment of biological manures in order to reduce the dependence of the chemical fertilizers in the agricultural production. Furthermore, the aqueous extracts of this alga proved endowed with a strong antifungal capacity against the fungi tested in this work. A result could be considered very interesting, since P. pavonica molecules had a double effect in the crops: a fertilizing and an anti-fungal effect without deteriorating the physico-chemical properties of the soil. The differences in behavior of seeds and fungi in the presence of various organic ex-tracts used enabled to have a preliminary idea on the nature of the bioactive molecules. Further studies are necessary to separate, purify and identify responsible molecules for the antifungal effects. The industrialization possibility of these natural bioactive products is a challenge that must be faced.

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