

# Amelioration Of Seaweed Liquid Fertilizer (Slf) Of Padina On The Growth And Biochemical Characters Of Cow Pea (*Vigna Unguiculata* (L.) Walp.)

N. Tensingh Baliah

P. Valarkkodi

Post Graduate and Research Department of Botany,  
Ayya Nadar Janaki Ammal College, Sivakasi, Tamil Nadu, India

**Abstract:** *The present study is aimed at to find the positive effect of Seaweed Liquid Fertilizer (SLF) of Padina in cow plants under nursery conditions. The application of SLF greatly increased the growth characters such as germination percentage, seedling vigour index, shoot and root length, plant fresh and dry weight. The results revealed that SLF @ 0.3% found to be more effective in increasing the shoot length of cowpea. There was marked differences observed in the root length and the effect was higher in 0.3% SLF and then there was gradual decrease in root length. The optimum concentration of Padina plant fresh as well as dry weight of cowpea was 0.3%. The seedlings treated with low concentration of SLF (0.1%) showed better results in chlorophyll content. Glucose content was significantly improved by SLF particularly 0.3% level, beyond and after this concentration there was a marked differences were observed. SLF of Padina in all the treatments resulted significant increase in protein content. The higher protein content was obtained with 0.3% followed by 0.4% of SLF application. Likewise, SLF plays a significant role in improving the NR activity of leaf of cowpea and the improvement from 0.1% to 0.3% and then there was gradual decrease in NR activity.*

**Keywords:** *Padina, SLF, cow pea, growth, biochemical attributes*

## I. INTRODUCTION

India is an agricultural country with approximately 70% of the population directly engaged in agriculture. The growing population is facing pressure on food production and to meet the increasing demand, farmers are using chemical fertilizers to enhance their crop production. Chemical fertilizers get accumulated in plants which lead to health problems in human beings due to bio-magnifications. Further, the mismanagement and excessive use of inorganic fertilizers creates problems in soil fertility and environmental pollution. Hence, a widespread need has arisen to go in for organic farming and cultivation. The efficiency of sole organic inputs in nutrient management was studied through the use of different types organic manures. Organic farming is a productive system, which reduces entirely the use of chemical fertilizers and pesticides, growth regulators and other agricultural chemicals. Organic

farming system is primarily aimed at cultivating the land and raising crops in such a way as to keep the soil alive and in good health. The advantages of organic farming are helping in maintaining environment health by reducing the level of pollution, reduce the human and animal hazards, helps in keeping agricultural products at a higher level and make it sustainable, reduce the cost of production, improve the soil health and ensure to maximum utilization of natural resources (Kilcher, 2002; Vogl and Darnhofer, 2004; Sasikala *et al.*, 2016).

Seaweeds are marine macro algae which form an important component of the marine living resources of the world. They occur in the intertidal, shallow and deep waters of the sea up to 180m depth and also in estuaries and backwaters. They grow on rocks, dead corals, stones, pebbles, solid substrata and on the other plants. Seaweeds usually grow vertically away from the substratum which brings them

closure to light; major environmental factors such as light, temperature, salinity, water motion and nutrient availability are related to seaweeds growth. Seaweeds are classified into three groups namely green (Chlorophyceae), brown (Phaeophyceae) and red (Rhodophyceae) based on their pigments like chlorophylls, carotenoids and phycobiliproteins (Khan *et al.*, 2009; Hong *et al.*, 2007). Now-a-days seaweeds are used in agriculture as potential organic fertilizer. The application of seaweed fertilizers for different crop plants is of great importance to substitute/supplementary to the inorganic fertilizers and to reduce the cost of production. The current research indicates that the extract from seaweeds are found to be superior to chemical fertilizers due to high level of organic matter, micro and macro elements, vitamins, fatty acids and growth regulators.

Seaweed extracts act as biostimulants mainly due to the presence of plant hormones. Main phytohormones identified in seaweed extracts are auxins, cytokinins, gibberellins, abscisic acid and ethylene. Auxins are responsible for elongational growth of plant tissues and apical dominance, cell division, plant movements and plant aging. Cytokinins are involved in cell division regulation affecting plant growth and rest period. One of the basic functions of gibberellins is initiation of seed germination, growth regulation, breaking bud dormancy, floescence and fruits development. Abscisic acid and ethylene are responsible for response to stress factors, inhibition of cell growth, and acceleration of plant aging. Furthermore, abscisic acid participates in regulation of seed germination (Gour Gopal and Ruma, 2011; Stirk *et al.*, 2003). Seaweeds the most widely used in agriculture due to their good biostimulant activity are red algae: *Corralina mediterranea*, *Jania rubens*, *Pterocladia pinnata*; green algae: *Cladophora dalmatica*, *Enteromorpha intestinalis*, *Ulva lactuca* and brown algae: *Ascophyllum nodosum*, *Ecklonia maxima*, *Saragassum spp.* etc (El- Sheekh and El- Saled, 2000; Oosten *et al.*, 2017).

Seaweed extract are biodegradable, non-toxic, non-polluting and nonhazardous to human. It is well known that chemical fertilizers degrade the fertility of the soil by making it acidic, rendering it unsuitable for rising crops, however, seaweed extract besides increasing the soil fertility increases the moisture holding capacity and supplies adequate trace metals improving the soil structure. Seaweed Liquid Fertilizer (SLF), a blend of both plant growth regulators and organic nutrient input is eco-friendly promoting sustainable productivity and maintaining the soil health (Dhargalkar and Pereira 2005; Jayasinghe *et al.*, 2016). The growing agricultural practices need more fertilizers for higher yield to satisfy the need of food for human beings. Developing countries like India, utilization of seaweeds and their extracts will be useful for achieving higher agricultural production. In recent years, seaweed liquid fertilizer is widely used as foliar spray/soil treatment to increase yield in many commercial crops plants. The beneficial effect of seaweed extract application is as a result of many components that may work synergistically at different concentration. Seaweed liquid fertilizer at concentration showed maximum germination percentage, fresh weight, dry weight, root and shoot length, number of branches, leaf area, root nodules and content of total chlorophyll. (Ganapathy Selvam and Sivakumar, 2014).

SLF promoted seed germination and enhanced early seedling growth in green gram. (Venkataraman Kumar *et al.*, 1994; Tensingh Baliah *et al.*, 2017).

## II. MATERIAL AND METHODS

### COLLECTION OF SEAWEEDS

The marine algae *Padina* was freshly collected from the coastal region of Rameshwaram, Tamil Nadu, India. The collected algae were washed thoroughly with seawater to remove all the unwanted impurities, adhering sand particles and epiphytes. Samples were washed thoroughly using fresh water to remove the surface salt and then blotted to remove excess water.

### PREPARATION OF SEAWEED LIQUID FERTILIZER (SLF)

Seaweeds were shade-dried for four days, followed by oven-drying for 24 hours at 60°C. The dried seaweeds were used for the preparation of Seaweed Liquid Fertilizer (SLF) following the method of Rama Rao (1990). The filtrate thus obtained was considered as 100% SLF, from which different concentrations (0.1%, 0.2%, 0.3%, 0.4%, and 0.5%) were prepared by adding distilled water.

### NURSERY EXPERIMENT

Five treatments were given to the nursery plants by the soil application of 0.1%, 0.2%, 0.3%, 0.4%, and 0.5% aqueous seaweed extract. In each treatment, 100 ml aqueous extract was applied. The first treatment was given to 10-day-old seedlings. Thereafter, three treatments at interval of 10<sup>th</sup> days each were given up to 30<sup>th</sup> days. The control set was treated only with water.

### ANALYSIS OF GROWTH CHARACTERS

The growth characters such as seed germination (ISTA, 1985), seed germination index (AOSA, 1983), shoot length, root length, fresh, dry weight, seedling vigour index I and II (Kharb *et al.*, 1994) were analyzed in the treated and control plants. Plants were uprooted without causing any damage to the seedlings and it was thoroughly washed with tap water in order to remove soil and debris particle. Then the shoot and root length was measured with the help of meter scale expressed in centimeter. The fresh weight and dry weight of whole plant parts (shoot, leaves and root) was weighed using SHIMADZU electronic balance.

### ANALYSIS OF BIOCHEMICAL CHARACTERS

The biochemical characters such as chlorophyll, protein, glucose and nitrate reductase activity were analyzed in treated and untreated control plants. The total chlorophyll was estimated using the formula of Wellburn and Lichtenthaler (1984); protein was estimated by Lowry *et al.* (1951) method; glucose in the leaf tissue was estimated by Anthrone

(Jayaraman, 1981) and *in vitro* NRA was assayed according to Jaworski (1971) method.

### STATISTICAL ANALYSIS

Morphological parameters were determined with five independent replicates. Biochemical characters and enzymatic assay were carried out at least three times. The data were reported as mean  $\pm$  SE and in the figure parentheses represent the present activity.

## III. RESULTS

### GROWTH CHARACTERS

The effect of SLF on the growth characters of cow pea was analyzed in the nursery plants. The growth characters such as germination percentage, germination index, seedling vigour index, shoot length, root length, number of leaves, fresh weight and dry weight were analyzed in the SLF treated and untreated control. The results revealed that there was a significant difference was observed in the rate of seed germination of cow pea and the rate was higher in SLF treatment over the control. The speed of germination index also varied according to nature of concentration of SLF. Little difference was shown in germination index among different concentration of SLF over the control. The values of the seedling vigour index of cow pea varied with the concentration of SLF. The highest value of seedling vigour index I was observed in plants treated with SLF @0.3% followed by 0.4%. In the case of seedling vigour index II, the index value was higher in 0.3% followed by 0.4% of SLF (Table 1).

S. No.	Treatment	Seed Germination (%)	Germination Index	Seedling Vigour Index - I	Seedling Vigour Index - II
1	Control	75	0.7	25.6	2.2
2	SLF - 0.1%	85	0.92	31.8	2.8
3	SLF - 0.2%	80	1.25	37.8	3.5
4	SLF - 0.3%	80	1.5	50.6	5.8
5	SLF - 0.4%	85	1.46	52.1	5.4
6	SLF - 0.5%	85	1.42	49.4	4.3

Table 1: Effect of SLF of *Padina* on the growth characters of cow pea (*Vinga unguiculata* (L.) Walp.)

The shoot length showed that the maximum length of 63.33cm in 0.3 % concentration of SLF, while the control plants showed only 34.1cm. In other concentrations of SLF also, better shoot length was noted as 37.4, 47.2, 61.3 and 58.1 at the concentration of 0.1%, 0.2%, 0.4%, and 0.5% respectively. The maximum length of root observed was 23.3cm in 0.3% concentration of SLF whereas; it was decreased as 21.3cm and 20.6cm with 0.4% and 0.5% SLF concentration respectively. But the control plant showed very less value of 12.0cm. The maximum plant fresh weight was noted in 0.3% concentration of SLF (17.2g) whereas, the control plant showed the minimum plant fresh weight of 8.2g. Compared the to control plants, the maximum plant dry weight was noted from 0.1% concentration of SLF up to 0.3% and then gradual decreases with the concentration of 0.4% and 0.5%. Among treated plants, the maximum plant dry weight was observed in plants treated with SLF at the concentration of 0.3% (Table 2).

S. No.	Treatment	Shoot length (cm)	Root length (cm)	Fresh weight (g)	Dry weight (g)
		34.1	12.0	8.20	2.90
2	SLF - 0.1%	37.4 $\pm 0.52$ (109)	15.0 $\pm 0.08$ (125)	9.60 $\pm 0.17$ (117)	3.26 $\pm 0.20$ (112)
3	SLF - 0.2%	47.2 $\pm 0.21$ (138)	18.3 $\pm 0.57$ (152)	11.16 $\pm 0.20$ (136)	4.40 $\pm 0.15$ (151)
4	SLF - 0.3%	63.3 $\pm 0.05$ (186)	23.3 $\pm 0.15$ (194)	17.20 $\pm 0.06$ (209)	7.26 $\pm 0.21$ (243)
5	SLF - 0.4%	61.3 $\pm 0.12$ (180)	21.3 $\pm 0.28$ (177)	15.02 $\pm 0.02$ (183)	6.40 $\pm 0.16$ (220)
6	SLF - 0.5%	58.1 $\pm 0.06$ (170)	20.6 $\pm 0.33$ (171)	13.03 $\pm 0.02$ (158)	5.00 $\pm 0.12$ (172)

Table 2: Effect of SLF of *Padina* on the biomass of cowpea (*Vinga unguiculata* (L.) Walp.)

### BIOCHEMICAL CHARACTERS

The biochemical constituents of *V. unguiculata* when grown with different concentration of *Padina* SLF showed better results over the control. At 0.3% concentration of SLF of *Padina* showed the maximum total chlorophyll content was noted (2.23mg/g. LFW) over the control (1.54mg/g. LFW). But, slight reduction was observed in 0.4% and 0.5% concentration of SLF (2.13mg/g. LFW and 1.97mg/g. LFW). The application of SLF of *Padina* significantly increased the carotenoid content. The result revealed that the effect was higher in 0.3% of SLF. There was not much variation in carotenoid content 0.3 and 0.4% of SLF.

At 0.3% concentration, the plants showed maximum concentration of glucose (55.66mg/g) and their increments were more compared to other treatments and control plants. The application of SLF of *Padina* in all the treatments resulted in a significant increase in protein content. The higher protein content was obtained with 0.3% followed by 0.4% of SLF application. Over the control plants, treated plants with SLF in all the treatments showed a significant increase in NR activity. The NR activity was higher in plants treated with SLF @ 0.3% with increase the concentration of SLF of *Padina*, there was a gradual increase in the NRA up to 0.3% and then gradual decrease in 0.4% and 0.5% of SLF (Table 3).

S. No.	Treatment	Total Chlorophyll (mg/gLFW)	Carotenoids (mg/gLFW)	Glucose content (mg/gLFW)	Protein content (mg/gLFW)	NRA ( $\mu$ moles/30 minutes)
1	Control	1.54 $\pm 0.12$ (100)	0.28 $\pm 0.10$ (100)	41.53 $\pm 0.02$ (100)	0.95 $\pm 0.03$ (100)	3.16 $\pm 0.36$ (100)
2	SLF - 0.1%	1.72 $\pm 0.20$ (111)	0.31 $\pm 0.12$ (110)	44.93 $\pm 0.03$ (107)	1.35 $\pm 0.02$ (142)	3.83 $\pm 0.10$ (121)
3	SLF - 0.2%	1.88 $\pm 0.15$ (115)	0.37 $\pm 0.15$ (132)	46.00 $\pm 0.13$ (112)	1.62 $\pm 0.01$ (171)	4.41 $\pm 0.08$ (139)
4	SLF - 0.3%	2.23 $\pm 0.22$ (144)	0.52 $\pm 0.24$ (185)	55.66 $\pm 0.04$ (134)	2.03 $\pm 0.04$ (214)	6.92 $\pm 0.18$ (218)
5	SLF - 0.4%	2.13 $\pm 0.20$ (138)	0.49 $\pm 0.14$ (175)	53.20 $\pm 0.02$ (128)	1.83 $\pm 0.2$ (193)	6.08 $\pm 0.16$ (192)
6	SLF - 0.5%	1.97 $\pm 0.25$ (127)	0.42 $\pm 0.39$ (150)	50.86 $\pm 0.05$ (122)	1.76 $\pm 0.05$ (185)	5.58 $\pm 0.15$ (176)

Table 3: Effect of SLF of *Padina* on the biochemical characters of cowpea (*Vinga unguiculata* (L.) Walp.)

## IV. DISCUSSION

## GROWTH ATTRIBUTES

SLF of *Caulerpa taxifolia* possesses fertilizer activity to enhance the germination and seedling growth in different crops. SLF are economical and ecofriendly alternatives to chemical fertilizers so this simple practice of application of SLF to different crops is recommended to attaining better germination and growth (Misal and Sabale, 2016). Likewise, SLF of *Gracilaria textorii* and *Hypnea musciformis* also significantly increased the seed germination, growth and yield parameters such as number of leaves, weight of fruits in crop plants such as brinjal, tomato and chilly. The mode of application of SLF also affected the seed germination percentage, nutrient uptake, growth (Immanuel and Subramanian, 1999) and yield of crops (Anantharaj and Venkatesalu, 2002; Jayaprakash *et al.*, 2016). The growth parameters increase mainly due to the presence of micro-nutrients, auxins and cytokinins and other growth promoting substances in SLF (Spinelli *et al.*, 2010). The mechanism of action is the enhancement of cell size and cell division, and together they complement each other as cytokinins are effective in shoot generation and auxins in root development (Liu and Lijun, 2011; Misal and Sabale, 2016).

The SLF of *Ulva lactuca*, *Sargassum wightii* and *Gelidella acerosa* induced maximum germination, root and shoot growth in Ragi; *Enteromorpha intestinalis* increased seed germination, root, shoot length and chlorophyll content of *Sesamum indicum* (Gandhiyappan and Perumal 2001); *Gracilaria edulis* extract supported higher growth, fruiting and flowering in *Ablemoschus esculentus* (Ramshubramanian *et al.*, 2004); *Gracilaria edulis* applied to soil bed maximum germination, growth and development in *Zea mays* and *Phaseolus mungo* was obtained (Lingakumar *et al.*, 2004); *Hydroclathrus* exhibited maximum per cent increase in the growth parameters of *Sorghum* (Ashok and Rathinavel, 2004); *Gracilaria verucosa* and *Chaetomorpha linum* as foliar spray significantly increased the vegetative growth of black gram, brinjal and tomato (Sethi and Adhikary 2009); *Sargassum johnstonii* induced significant increase in vegetative growth of *Lycopersicon esculentum* (Divya *et al.*, 2015).

The reasons behind the positive effect of seaweeds are presence of appreciable quantities of plant growth regulators (Mooney and Van Staden, 1985), cytokinin (Smith and Van staden, 1984), IAA (Abe *et al.*, 1972), gibberellins and gibberellins-like substances (Sekar *et al.*, 1995). Cytokinins and gibberellins, growth hormones detected in many species of *Sargassum*, might be responsible for beneficial effects to black gram. The increase in the growth variables at lower concentration of the SLF treated plant may be due to the uptake of magnesium, phosphorus, potassium, nitrate and iron from the seaweed extract of *S. myriocystum*. Dhargalkar and Pereira (2005) also reported similar findings with *Hypnea musciformis*, *Spatoglossum asperum*, *Stoechospermum marginatum* and *Sargassum* on the growth of crops such as *Capsicum frutescens*, *Brassica rapa* and *Ananas comosus*. El-Shora *et al.* (2016) reported that the growth parameters such as shoot length, root length, fresh weight of shoot and root of vegetable plant *Cyanopsis tetragonoloba* increased with the

application of optimum concentration of seaweed liquid fertilizer (SLF).

## BIOCHEMICAL ATTRIBUTES

The application of different concentrations of SLF (0.5%, 1%, 2.0%, 2.5%) when applied to soil bed, enhanced the chlorophyll a, b, protein, sugar, starch and nitrate reductive activity of Sorghum. SLF of *Sargassum polycystum* significantly enhanced the biochemical composition of the black gram when treated in soil during transplantation; SLF of *Gracilaria edulis* showed positive response on the photosynthetic pigment, total amino acid, protein and starch content of *Zea mays* (Lingakumar *et al.*, 2002). The liquid extract of *Gracilaria edulis* showed higher growth and biochemical characteristic in *Ablemoschus esculentus* (Ramshubramanian *et al.*, 2004). Higher chlorophyll, carbohydrate, and protein content were observed in *Dolichos biflorus* seeds by treating with 10% extract of *Caulerpa racemosa* and *Gracilaria edulis* (Anantaraj and Venkatesalu, 2002). The application of SLF increased chlorophyll a, b and xanthophyll pigments content in most of the concentration tested. Similar results were observed by Blunden *et al.* (1986). Seaweed liquid fertilizer of *U. fasciata* and *S. swartzii* enhanced the synthesis of biochemical constituents such as photosynthetic pigments, soluble sugar, soluble protein and lipid in C3 plants.

The extract of *U. lactuca* improved the accumulation of total carbohydrate, total protein and total lipid contents was also found maximum when the plants received with 1.0% SLF on marigold. At this condition their increments were more than 37.0%, 58.0% and 60.0% respectively. The application of aqueous seaweed extract in all the concentrations and treatments resulted in a significant increase in protein concentration. The higher protein concentration was measured with 10% seaweed extract application. Soil drench and foliar spray treated plants gave the best response. Such a rise in protein content may be attributed to the increased availability and absorption of necessary elements (Ca, Na, Mg, Cd and Zn) present in the aqueous extract that enhanced the efficiency of leaves. *Z. mays* and *P. mungo* responded well in maintaining high level of soluble proteins at 1% and 0.5%. *Sargassum* extracts application (Lingakumar *et al.*, 2004). Significant increase in the levels of protein content of *Sorghum vulgare* was recorded with 1.5% seaweed liquid extract prepared from *Hydroclathrus clathratus* (Ashok *et al.*, 2004). The increase in protein content at lower concentrations of liquid extract may be due to absorption of most of the necessary elements by the seedling (Anantharaj and Venkatesalu, 2001). The concentration of total soluble sugars and reducing sugars was maximum at higher concentrations of spray and drench treatments. This may be considered indicative of the fact that seaweed extracts stimulate various biological processes that increase the carbohydrate levels in tomato plants (Sivasankari *et al.*, 2006; Tensingh Baliah *et al.*, 2017).

## V. CONCLUSION

From the present study, it is clear that SLF of *Padina* possesses fertilizer activity to enhance the germination, seedling growth, biochemical components of cow pea. The application of SLF significantly increases the both growth and biochemical character of cowpea. In particular, 0.3% of SLF of *Padina* is the optimum/favourable for growth and biochemical response in cowpea. SLF are economical and eco-friendly alternative/supplementary to chemical fertilizers. This study is useful in demonstrating the potential of SLF of *Padina* as an option that provides competitive yields for organic farmers. However, the concentration of SLF should be standardized with respect to nature of crop plants.

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