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Effect of seaweed saps on growth and yield improvement of green gram

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A field experiment was conducted during the *pre-kharif* season at Uttar Chandamari village in 2012 to study the effects of seaweed saps on growth, yield and quality improvement of green gram in new alluvial soil of West Bengal. The foliar spray was applied twice at different concentrations (0, 2.5, 5.0, 7.5, 10.0 and 15.0% v/v) of seaweed extracts (namely *Kappaphycus* and *Gracilaria*). Foliar applications of seaweed extract significantly enhanced the growth, yield and quality parameters. The highest grain yield was recorded with applications of 15% *Kappaphykus* sap + recommended dose of fertilizer (RDF), followed by 15% *Gracilaria* - sap + RDF extract resulting in an increase by 38.97 and 33.58% grain yield, respectively compared to the control. The maximum straw yield was also achieved with the application of 15% seaweed extract. Improved crop quality and nutrient uptake [nitrogen (N), phosphorus (P) and potassium (K)] was also observed with seaweed extract applications.

Key words: Seaweed saps, *Kappaphycus*, *Gracilaria*, recommended dose of fertilizer, green gram.

INTRODUCTION

Any improvement in agricultural system that results in higher production should reduce the negative environmental impact and enhance the sustainability of the system. One such approach is the use of bio-stimulants, which can enhance the effectiveness of conventional mineral fertilizers. Long-term indiscriminate use of them invites the crucial problem of soil health disorder vis-à-vis reduced input use efficiency, more precisely, fertilizer use efficiency. Due to these reasons the farmers are being compelled gradually day by day to turn towards various options like organic manures, bio-stimulants, growth regulators etc. One of such options is the use of seaweed extracts as plant nutrient bearing fertilizer. Marine bioactive substances extracted from marine algae are used in agricultural and horticultural crops, and many beneficial effects may be achieved in terms of enhancement of yield and quality. Liquid extracts obtained from seaweeds have recently gained importance as foliar sprays for many crops including various cereals, pulses and different vegetable species. Seaweed extracts contain major and minor nutrients, amino acids, vitamins, cytokinins, auxin and abscisic acid like growth promoting substances and have been reported to stimulate the growth and yield of plants, develop tolerance to environmental stress (Zhang et al., 2003), increase nutrient uptake from soil (Turan and Kose, 2004) and enhance antioxidant properties (Verkleij, 1992). Liquid extracts obtained from seaweeds have recently gained importance as foliar sprays for many crops including various grasses, cereals flowers andvegetable species (Crouch and Van Staden, 1994). In recent years, the use of seaweed extracts have gained in popularity due to their

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Table 1. Details of experimental treatment.

Treatment	Details	Time of application
T_1	2.5% *Kappaphycus*-sap + RDF	20 DAS and 40 DAS
T_2	5% *Kappaphycus*-sap + RDF	,,
T_3	10% *Kappaphycus*-sap + RDF	,,
T_4	15% *Kappaphycus*-sap + RDF	,,
T_5	2.5% *Gracilaria*-sap + RDF	,,
T_6	5% *Gracilaria*-sap + RDF	,,
T_7	10% *Gracilaria*-sap + RDF	,,
T_8	15% *Gracilaria*-sap + RDF	,,
T_9	RDF + Water spray	,,
T_{10}	7.5% *Kappaphycus*-sap + 50% RDF	,,

potential use in organic and sustainable agriculture (Russo and Beryln, 1990), especially in rainfed crops, as a means to avoid excessive fertilizer applications and to improve mineral absorption. Unlike chemical fertilizers, extracts derived from seaweeds are biodegradable, non-toxic, non-polluting and non-hazardous to humans, animals and birds (Dhargalkar and Pereira, 2005).

With the advancement of modern agricultural technologies, we somehow achieved food security for our country to feed its ever escalating population. But till date, unfortunately, we could not achieve nutritional security for our people. In West Bengal India, the growing of pulses in the cropping system is nowadays not a common practice, but it is well known that inclusion of any pulse crop in the sequence has all-round advantages. So, if we are to achieve nutritional security we must have to go for crop diversification in cropping pattern introducing crops like pulses for example, green gram, black gram etc. in *Zaid* (Summer) season and lentil, chickpea, garden pea etc. in *Rabi* (Winter) season. On the other hand, to nourish the protein energy malnourished (PEM) people and to maintain soil health properly pulse crops should be included in the cropping system. Green gram or mung bean (*Vigna radiata* L.) is the third most important food legumes grown and consumed in India and is a good source of proteins and minerals and its protein quality is similar to or better than other legumes like chickpea, black gram, peas, pigeon pea etc. (Kataria et al., 1989; Jood et al., 1998a).

Considering each and every corner of the above discussion, an experiment was conducted to study the effect of different seaweed saps on growth and yield of green gram and assess the nutrient removal by seed and stover of this pulse crop.

MATERIALS AND METHODS

Experimental site and soil information

The field experiment was conducted during the *pre-kharif* season of 2012 on inceptisol at Uttar Chandamari village of Nadia district of West Bengal in India. The soil of the site was sandy clay loam with pH 6.45, organic carbon 0.57%, total nitrogen 0.055%, available P_2O_5 26.29 kg ha^{-1} and available K_2O 148.72 kg ha^{-1}. The climate of the region is humid subtropical. The experimental site is located at 22° 57' N latitude, 88° 20' E longitude and altitude is 7.8 m.

Experimental designs and treatments

The experiment comprised of ten treatments, the details of the treatments are mentioned Table 1.

Two sprays of *Kappaphycus* and *Gracilaria* extract were applied; one at the seedling stage [20 days after sowing (DAS)] and the other at the pre-flowering stage (40 (DAS). For proper adherence, extracts were mixed with proper surfactant (Active 80 at 0.5 ml L^{-1} of water) at the time of spraying. The total spray volume was 650 L ha^{-1} in each application. The treatments were distributed in a randomized block design with three replications. The plot size was 5 × 6 m. The recommended dose of fertilizer (RDF) for green gram was 20:40:40 kg ha^{-1} N, P_2O_5 and K_2O, respectively and all fertilizers were applied as basal. Date of sowing was 25th of February, 2012 and the crop was harvested on 5th May, 2012.

Preparation and chemical composition of liquid sea weed extract

The seaweed extract used in this study was obtained from *Kappaphycus* sp. and *Gracilaria* sp. The algae were handpicked from the coastal area of Rameswaram, T. N., India during September, 2011. It was washed with seawater to remove unwanted impurities and transported to the field station at Mandapum, Rameswaram. Here, samples were thoroughly washed using tap water. After that, fresh seaweed samples were homogenized by grinder with stainless steel blades at ambient temperature, filtered and stored (Eswaran et al., 2005). The liquid filtrate was taken as 100% concentration of the seaweed extract and further diluted as per the treatments. The nitrogen (N) content of seaweed extract (100% concentrate) was determined by taking 20 ml of filtrate which was oxidized and decomposed by concentrate sulphuric acid (10 ml) with digestion mixture (K_2SO_4 : $CuSO_4$ = 5:1) heated at 400°C temperature for 2½ h as described in the semi-micro Kjeldahl method [AOAC International, 1995, method No. Ba 4b-87(90)], and other nutrient elements were analyzed by inductively coupled plasma-optical emission spectroscopy (ICP-OES), after wet digestion of filtrate (20 ml) with HNO_3-$HClO_4$ (10:4) di-acid mixture (20 ml) and heated at 100°C for 1 h and then raise the temperature to about 150°C (Richards, 1954)

Table 2. Chemical composition of *Kappaphycus* sap.

Nutrient	Amount present	Nutrient	Amount present
Moisture	94.38 g/100 ml	Iron	8.58 mg/100 ml
Protein	0.085 g/100 ml	Manganese	0.22 mg/100 ml
Fat	0.0024 g/100 ml	Nickel	0.35 mg/100 ml
Crude fibre	0.01 g/100 ml	Copper	0.077 mg/100 ml
Carbohydrate	1.800 g/100 ml	Zinc	0.474 mg/100 ml
Energy	7.54 Kcal/100 ml	Chromium	3.50 mg/100 ml
Sodium	18.10 mg/100 ml	Lead	0.51 mg/100ml
Potassium	358.35 mg/100 ml	Thiamine	0.023 mg/100 ml
Magnesium	116.79 mg/100 ml	Riboflavin	0.010 mg/100 ml
Phosphorous	2.96 mg/100 ml	B-Carotene	0.0 mg/100 ml
Calcium	32.49 mg/100 ml	Iodine	160 mg/100 ml
Indole acetic acid	23.36 mg/L	Kinetin + Zeatin	31.91 mg/L
Gibberelin GA_3	27.87 mg/L		

Data courtesy: National Institute of Nutrition, Hyderabad, India (except growth hormone data generated by CSMCRI using quantitative MS-MS and LC-MS techniques).

Table 3. Chemical composition of *Gracilaria* sap.

Nutrient	Amount present	Nutrient	Amount present
Ash	38.91 g/100 g	Calcium	295.50 mg/100 g
Crude protein	9.58 g/100 g	Copper	0.20 mg/100 g
Crude fibre	10.40 g/100 g	Zinc	1.00 mg/100 g
Crude lipid	2.00 g/100 g	Iron	67.35 mg/100 g
Saturated fatty acid	48.92% of total fatty acids	Manganese	4.16 mg/100 g
Total amino acids	889.78 mg/g of protein	Nickel	0.92 mg/100 g
Moisture	88.88%	Cobalt	0.24 mg/100 g
Vitamin C	28.50 mg/100 g	Sulphate	106.20 mg/100 g
Carbohydrate	45.92%	Chlorine	1170.00 mg/100 g
Potassium	8633.00 mg/100 g	Lead	1.11 mg/100 g
Magnesium	549.50 mg/100 g	Cadmium	0.14 mg/100 g
Phosphorus	278.50 mg/100 g	Sodium	158.50 mg/100 g

Source: Benjama and Masniyom (2012), and Sivakumar et al. (2009). *Gracilaria* extract also contains variable amount of phytohormones like Auxin, Cytokinin, Abscisic acid etc. (Yokoya et al., 2010).

(Tables 2 and 3).

Plant sampling

Data were taken through random sampling at 21 DAS, 42 DAS and 63 DAS to measure plant height, dry matter accumulation, crop growth rate (CGR) and leaf area index (LAI). CGR was computed with the help of the formula: $[(W_2 - W_1)/ (t_2 - t_1)]$ where, W_1= dry weight per unit area at t_1, W_2 = dry weight per unit area at t_2, t_1 = first sampling and t_2 = second sampling. LAI was computed by the ratio of leaf area to the area of ground cover. For measuring nodule number per plant, data were taken at 21 and 42 DAS. Data on yield attributes were taken randomly before harvesting. At maturity, green gram seeds and stover samples were collected from each plot, oven dried at 70°C to constant weight and ground to pass through a 0.5 mm sieve for chemical analysis. The N content was determined by the semi-micro Kjeldahl method [AOAC International, 1995, method No. Ba 4b-87(90)]. Phosphorus (P) content was determined by the Vanado-Molybdate yellow method (Jackson, 1973) and Potassium (K) content by flame photometry (Jackson, 1973).

Statistical analysis

Data were analyzed using analysis of variance (ANOVA) following randomized block design (Gomez and Gomez, 1984). Differences were considered significant at 5% level of probability.

RESULTS

Effect of treatments on growth of green gram

Foliar application of different sea weed saps along with RDF increased growth attributes of greengram

Table 4. Effect of treatments on plant height (cm), dry matter accumulation (g m^{-2}), CGR (g m^{-2} day^{-1}), no. of nodule per plant and LAI of green gram.

Treatment	Plant height (cm)			Dry matter accumulation (g m^{-2})			CGR (g m^{-2} day^{-1})		Nodule no. $plant^{-1}$		LAI		
	21 DAS	42 DAS	63 DAS	21 DAS	42 DAS	63 DAS	21 - 42 DAS	42 - 63 DAS	21 DAS	42 DAS	21 DAS	42 DAS	63 DAS
T_1	15.63	38.33	52.30	19.67	150.31	280.41	6.23	6.19	2.80	18.5	0.65	2.90	2.87
T_2	16.71	38.93	52.87	19.79	155.83	288.23	6.48	6.30	3.11	19.3	0.70	2.97	2.99
T_3	18.92	42.57	54.67	21.41	170.23	299.57	7.09	6.16	3.82	20.2	0.84	3.43	3.31
T_4	19.13	45.52	59.13	27.93	190.78	320.78	7.75	6.19	3.95	23.1	1.08	5.09	6.09
T_5	14.25	39.77	50.17	19.17	145.57	271.61	6.04	6.00	2.47	17.8	0.58	2.85	2.74
T_6	15.97	40.11	49.33	23.67	149.91	280.19	6.02	6.20	2.67	18.3	0.61	2.93	2.81
T_7	16.48	43.77	53.47	24.25	171.33	289.72	7.00	5.64	3.23	19.2	0.88	3.21	3.07
T_8	17.27	44.67	55.87	25.69	188.97	308.56	7.78	5.70	3.78	22.9	0.97	4.87	5.57
T_9	14.11	39.19	51.23	18.23	142.27	265.57	5.92	5.87	2.39	17.7	0.50	2.81	2.77
T_{10}	17.44	37.35	51.37	24.25	161.23	291.46	6.52	6.20	2.71	18.3	0.75	3.48	3.59
SEm(±)	0.93	1.74	1.41	1.90	8.51	6.70	0.56	1.25	0.34	1.26	0.08	0.18	0.06
CD at 5%	2.71	5.08	4.18	5.81	25.73	19.81	1.65	3.69	1.01	3.74	0.24	0.53	0.18

T_1: 2.5% *Kappaphycus* - sap + RDF; T_2: 5 % *Kappaphycus* - sap + RDF; T_3: 10% *Kappaphycus* - sap + RDF; T_4: 15% *Kappaphycus* - sap + RDF; T_5: 2.5% *Gracilaria* - sap + RDF; T_6: 5% *Gracilaria* - sap + RDF; T_7: 10% *Gracilaria* - sap + RDF; T_8: 15% *Gracilaria* - sap + RDF; T_9: RDF + Water spray; T_{10}: 7.5% *Kappaphycus*- sap + 50% RDF.

significantly over control (Table 4). In general, a gradual increase in plant height, dry matter accumulation and LAI was observed with increasing seaweed extract application.

Though these parameter are not significantly affected by foliar applications of seaweed extracts up to 5% concentration. Maximum plant height, dry matter accumulation, number of nodules per plant was recorded with 15% *Kappaphycus*- sap + RDF and was statistically at par with 15% *Gracilaria* - sap + RDF treated plot regarding all the observations taken at different DAS. In case of CGR during 21 to 42 DAS, the best result (7.75 g m^{-2} day^{-1}) was recorded with the treatment T_8 which was closely followed by T_4. Highest value of LAI was recorded with T_4 for all the observations.

Effect of treatments on yield attributes and yields of green gram

As per the data depicted in Table 5 the maximum numbers of branches per plant, pods per plant and seeds per pod were observed under the treatment T_4 (highest dose, that is, foliar application of 15% *Kappaphycus* sap along with RDF) which was closely followed by 15% *Gracilaria* - sap + RDF (T_8). The treatment T_4(15% *Kappaphycus*-sap + RDF) showed the maximum increase in yield over control to the extent of 38.97% and this treatment was followed by the treatments T_8(15% *Gracilaria* - sap + RDF), T_3 (10% *Kappaphycus*- sap + RDF), T_7 (10% *Gracilaria* - sap + RDF), T_2 (5% *Kappaphycus*- sap + RDF) and T_6 (5% *Gracilaria* - sap + RDF) recording 33.58, 27.28, 21.17, 19.77 and 13.86% yield increase, respectively over control. Similar kind of results reported for *Phaseolus aureus* (Bai et al., 2008). Increase in yield of several other crops like *Capsicum annuum* (Arthur et al., 2003), black gram (Venkataraman and Mohan, 1997) and canola plants (*Brassica napus*) (Ferreira and Lourens, 2002) are reported with the foliar application of seaweed extract.

Effect of treatments on the uptake of nutrients by greengram

The use of the seaweed extracts significantly increased N, P and K uptake by grains at higher concentrations (10% and above) and reached maximum at 15% seaweed extract compared with control (Table 6). The highest N and K uptake by grain was recorded with the treatment T_4(15% *Kappaphycus*- sap + RDF) which was statistically at par with 15% *Gracilaria*- sap + RDF (T_8), 10% *Kappaphycus*- sap + RDF (T_3) and 7.5% *Kappaphycus*- sap + 50% RDF (T_{10}). 15% *Kappaphycus*-sap + RDF showed the maximum uptake of P by grain. In case of nutrient uptake by stover, 15% *Kappaphycus* - sap + RDF was observed to be the best and it was closely followed by 15% *Gracilaria* - sap + RDF and 10% *Kappaphycus* - sap + RDF. Our results confirm those findings previously reported by Crouch et al. (1990) who noted an increased uptake of magnesium (mg), K and calcium (Ca) in lettuce with seaweed concentrate application. Turan and Köse (2004), Nelson and Van Staden (1984), and Mancuso et al. (2006) also observed

Table 5. Effect of treatments on yield components and seed and stover yield of green gram.

Treatment	No. of branches plant^{-1}	No. of pod plant^{-1}	No. of seed pod^{-1}	Test weight (g)	Seed yield (kg ha^{-1})	Stover yield (kg ha^{-1})
T_1	14.3	58.0	9.4	30.24	1085.6	4157.9
T_2	14.8	59.5	9.5	30.88	1090.3	4199.5
T_3	16.4	69.4	9.8	30.03	1158.6	4372.1
T_4	19.0	76.0	10.5	31.25	1265.0	5220.3
T_5	13.7	57.3	9.3	30.18	995.3	3909.7
T_6	14.6	58.9	9.8	29.67	1036.5	4125.3
T_7	16.4	70.2	10.1	30.99	1103.0	4657.8
T_8	18.5	72.1	10.7	30.61	1216.1	5107.2
T_9	12.3	50.0	8.6	28.37	910.3	3712.7
T_{10}	15.9	62.8	9.9	30.37	1101.7	4298.3
SEm(±)	0.4	1.7	0.4	1.26	8.12	9.07
CD at 5%	1.2	5.1	NS	NS	24.33	27.12

T_1: 2.5% *Kappaphycus* - sap + RDF; T_2: 5 % *Kappaphycus*- sap + RDF; T_3: 10% *Kappaphycus* - sap + RDF; T_4: 15% *Kappaphycus* - sap + RDF; T_5: 2.5% *Gracilaria* - sap + RDF; T_6: 5% *Gracilaria* - sap + RDF; T_7: 10% *Gracilaria* - sap + RDF; T_8: 15% *Gracilaria* - sap + RDF; T_9: RDF + Water spray; T_{10}: 7.5% *Kappaphycus*- sap + 50% RDF.

Table 6. Effect of treatments on nutrient removal by seed and stover of green gram.

Treatment	Nutrient removal by seed (kg ha^{-1})			Nutrient removal by stover (kg ha^{-1})		
	N	P	K	N	P	K
T_1	45.33	3.35	17.93	85.47	4.33	58.37
T_2	46.31	3.89	18.05	91.93	4.67	60.11
T_3	50.97	4.51	19.67	101.67	5.67	70.67
T_4	57.67	5.85	22.97	143.33	7.83	80.95
T_5	41.65	3.22	16.88	81.23	4.03	55.53
T_6	45.23	3.51	17.53	89.77	4.52	59.82
T_7	48.82	3.93	17.97	98.05	5.25	66.85
T_8	55.09	5.21	21.69	133.33	6.93	76.64
T_9	38.11	2.78	14.37	73.11	3.35	48.89
T_{10}	47.13	4.03	18.99	97.89	4.99	64.50
SEm(±)	3.81	0.33	1.41	4.89	0.27	1.76
CD at 5%	11.23	0.97	4.13	14.47	0.85	5.29

T_1: 2.5% *Kappaphycus*- sap + RDF; T_2: 5 % *Kappaphycus*- sap + RDF; T_3: 10% *Kappaphycus*- sap + RDF; T_4: 15% *Kappaphycus*- sap + RDF; T_5: 2.5% *Gracilaria* - sap + RDF; T_6: 5% *Gracilaria* - sap + RDF; T_7: 10% *Gracilaria* - sap + RDF; T_8: 15% *Gracilaria* - sap + RDF; T_9: RDF + Water spray; T_{10}: 7.5% *Kappaphycus*- sap + 50% RDF.

increased uptake of N, P, K and Mg in grape vines and cucumber with the application of seaweed extract. The presence of marine bioactive substances in seaweed extract improves stomata uptake efficiency in treated plants compared to non-treated ones (Mancuso et al., 2006).

DISCUSSION

Seaweed extract a the rich source of several primary nutrients like K, P; secondary nutrients like Ca, Mg; trace elements like zinc (Zn), copper (Cu), iron (Fe), manganese (Mn) and beneficial elements like nickel (Ni), sodium (Na) etc. Sea weed extracts stimulate various aspects of growth and development resulting in around good health of the plants, while deliberating the effect of sea weed extracts on crops the aspects of root development and mineral absorption, shoot growth and photosynthesis and ultimately crop yield, even vegetative propagation can also be taken into consideration. Due to the presence of good amount of P in it, the liquid seaweed fertilizers (LSF) proliferate root development, enhance root to shoot ratio, thereby, making the plants more able to mine adequate nutrients from the deeper layer of soil and influence crop maturity as a whole. As P is the important constituent of Nitrate reductase (NADP), the niacin component of Vitamin-B complex, helps in photosystem-I to produce NADPH. As LSF is a very good source of K, it helps in regulating the water status of the plants, controls the opening and closing of stomata and thereby the photosynthesis to a large extent. The meristematic growth, translocation of photosynthates and disease resistance are also influenced by it due to the manifestation of good impact of K. Ca being present in seaweed extracts helps in enzyme activation, cell elongation and cell stability. LSF is the opulent source of secondary nutrients like Mg; hence, it helps in photosynthesis, phloem export, root growth and nitrogen metabolism. It also influences the N-fixation in legumes as it contains Mn. Mn is a constituent of several cation activated enzymes like decarboxylase, kinase, oxidase etc., and hence, essential for the formation of chlorophyll, reduction of nitrates and for respiration. The trace elements like Fe, Cu and Zn being present in considerable amount in seaweed extracts inspire redox reaction of respiration and photosynthesis, promote reduction of nitrates and sulphates and stimulate the cation activated enzymes. The organic constituents of seaweed extract include plant hormones which elicit strong physiological responses in low doses. A panorama of phytohormones and plant growth regulators are found in different seaweed concentrates and marine macroalgal extracts *viz.* Auxins, Gibberellins, Cytokinins etc. which simulate rooting, growth, flower initiation, fruit set, fruit growth, fruit ripening, abscission and senescence when applied exogenously. Seaweeds also contain a diverse range of organic compounds which include several common amino acids inter alia aspartic acid, glutamic acid and alanine in commercially important species. Alginic acid, laminarin and mannitol represent nearly half of the total carbohydrate content of commercial seaweed preparations. Seaweeds also contain a wide range of vitamins which might be utilized by the crops. Vitamins C, B, (thiamine), B_2(riboflavin), B_{12}, D_3, E, K, niacin, pantothenic, folic and folinic acids occur in algae. Although vitamin A is not present, the presences of its precursor carotene and another possible precursor fucoxanthin have been found. Apart from the above organic and inorganic constituents, there is an evidence of existence of different other stimulatory and antibiotic substances. These findings are in agreement with Jeannin et al. (1991), Vernieri et al. (2005), Kowalski et al. (1999), Zhang and Ervin (2008), Mancuso et al. (2006), Norrie and Keathley (2006) and Rayorath et al. (2008).

Thus, being a wealthy source of versatile plant nutrients, phytohormones, amino acids, vitamins, stimulatory and antibiotic substances, the liquid sea weed extract enhances root volume and proliferation, bio-mass accumulation, plant growth, flowering, distribution of photosynthates from vegetative parts to the developing fruits and promotes fruit development, reduces chlorophyll degradation, disease occurrence etc. resulting in improved nutrient uptake, water and nutrient use efficiency causing sound general plant growth and vigor ultimately reflecting higher yield and superior quality of agricultural products.

Conclusion

Thus, it can be concluded that the seaweed extracts are effective in increasing the growth parameters, yield attributes, yield vis-à-vis quality of green gram. The saps also enhance nutrient uptake by this grain legume crop. Presence of micro-elements and plant growth regulators, especially cytokinins (Zodape et al., 2009; Zang et al., 2008) in *Kappaphycus* and *Gracilaria* extracts is responsible for the increased yield and improved nutrition of green gram receiving foliar application of the aforesaid two saps.

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