



# Response of Cassava Root *Manihot esculenta* to Potassium-Rich Biostimulants Manufactured from Red Seaweed *Gracilaria salicornia* Under Semi-Arid Condition

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**Abstract** Seaweed extracts are proven to be potent biostimulants due to the presence of wide range of nutrients including mineral like potassium and other macro-micronutrients, plant growth hormones, amino acids, vitamins, glycine betaines and quaternary ammonium compounds in them. In the present study, potassium-rich powders were obtained from some commercially important tropical red seaweeds viz. *Kappaphycus alvarezii*, *Gracilaria salicornia*, *G. edulis*, *G. firma* and *G. heteroclada* and *Eucheuma spinosum* and checked for their biostimulant effect through green gram seedling bioassay study on in vitro using WinRhizo software, and we found that extracts of all species increased the root development at significant level ( $p < 0.05$ ) as compared to standard nutrient medium. *G. salicornia* of Indian origin which is naturally available on a commercial level ( $>1000$  t dry per year) was taken for further studies and manufactured 3 variants of potassium-rich biostimulants, viz. concentrated seaweed extract (CSE), potassium-rich water-soluble powder (PSP) and seaweed fortified granule (SFG), and tested their biostimulant activity on cassava root at farmer's field and found to increase the tuber yield to 17.40%, 20.09% and 22.19%, respectively, with high starch content, less incidence of cassava brown streak virus disease (CBSVD) and increased nutrient use efficiency over control plants. Based on the results of present study, potassium-rich biostimulant obtained from *G. salicornia* can be applied to cassava for yield and quality improvement.

**Keywords** Biostimulant · Tropical red seaweeds · Potassium · Cassava root · Tuber yield · NUE

## Introduction

Cassava (*Manihot esculenta*) is an important food for more than 500 million people in the world and ranked fourth as a food crop after rice, maize, and wheat [6, 24, 33]. Between 1972 and 2021, global production of fresh cassava grew substantially from 101 to 314 mt rising at an increasing annual rate 8.28% in 2006 and then decreased to 3.64% in

2021 [11]. Global production of cassava is expected to reach 335 million metric tons by 2026, a 1.2% increase from 2021 levels. Since 1966, there has been a 1.4% rise in supply. In 2021, Nigeria was the leading producer of cassava with 60.8 mt followed by The Democratic Republic of Congo, Thailand and Ghana (Internet, assessed on 27 Dec. 2023). Cassava production has increased considerably in recent years, due to its significance as a staple food crop and as an industrial crop for manufacturing of starch as well. Total market value of cassava starch industry was estimated to be USD 50.75 Billion and expected to reach USD 65.68 Billion in 2027 with compound growth rate of 6.2% between 2022 and 2027 (Internet, assessed on March 26, 2023).

There was perception that cassava can be cultivated in low fertility soil that are too impoverished to support other staple crops [15]; this is because cassava has an extensive root system and is able to utilize plant nutrients less

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accessible to other crops. Fertilizer use is generally very low in Africa and on root and tuber crops like cassava because farmers think that cassava does not require it or may be because they are contented with the low yields obtained from using limited inputs [1]. But Fermont et al. reported that poor soil fertility has been identified as the most important constraint to cassava production and soil with moderate amount of fertilizer is required to produce high tuber yield [12] and about 60–70% of cassava produced globally is used for human consumption [9].

In India, cassava is mainly cultivated in southern states; it is mainly used for human consumption, industrial applications, and animal feed sector in India. About 60% of the total cassava produced in India is used as a raw material to produce starch, sago, and dry chips [29], and it has scope for wider applications in food, paper and textile industries [22]. In recent years, cassava has been globally recognized as a potential candidate for bioethanol production due to its high carbohydrate content and ability to grow under low management conditions. In India, since the diversified uses of cassava are limited, it has a great scope for exploitation for bioethanol production.

Biostimulants from seaweed *Kappaphycus alvarezii* have been extensively studied in India on wide range of agricultural and horticultural crops in different agro climatic conditions for its improved productivity [17, 18, 21, 27, 28]. *Kappaphycus alvarezii*, *Eucheuma spinosum*, *Gracilaria heteroclada* and *G. firma* are the major commercial important red seaweeds, and they together contribute over 50% of total global seaweed production (Internet, accessed on July 02, 2023).

Potassium is a third important fertilizer required for the growth of plant and currently all potassic fertilizers that are available in the market are chemically derived ones. The potassium derived from red seaweeds with product name of “Potassium derived from rhodophytes” has been included under Fertilizer Control Order (FCO) of India in July 2018 S.O. 3265(E) with specification (FCO 2018, India). Tubers and root crops including cassava are potassium loving crops and they need large amount of potassium for growth and physiological function. The present investigation aimed to study the presence of potassium-rich biostimulants from some commercial important red seaweeds like *Kappaphycus alvarezii*, *Gracilaria salicornia*, *G. edulis*, *G. firma* and *G. heteroclada* and *Eucheuma spinosum* on laboratory and pilot scale studies. *Gracilaria salicornia* which is naturally available on a commercial level (>1000 t dry per year) in India was further taken to manufacture three variants of potassium-rich biostimulants on commercial scale and studied their impacts on the tuber yield; starch content nutrients use efficiency of cassava root *Manihot esculenta* in semi-arid region.

## Materials and Methods

### Production and Analysis of Potassium-Rich Biostimulants from Some Tropical Red Seaweeds

Total 10 samples from 6 red seaweeds viz. *Kappaphycus alvarezii* (cultivated in India, Sri Lanka, Philippines, and Indonesia), *Gracilaria salicornia* (wild collection from India and Sri Lanka), *Eucheuma spinosum* (Indonesia) *Gracilaria heteroclada* and *Gracilaria firma* (Philippines) and *Gracilaria edulis* (India) were studied in the present investigation. The dry samples of all seaweeds taken were pulverized on laboratory scale to obtain whole seaweed powder with particle size of 60 to 80 mesh and tested their potassium and mineral nutrients contents. Potassium-rich water-soluble biostimulants were also prepared on pilot scale from all the species studied as follows: Dry weed (whole weed) was taken in water at 1:6 ratio and held it for 60 min at room temperature ( $30 \pm 3$  °C) with mild agitation; then, aqueous extract was separated and filtered through cloth filter followed by bag filter (200 mesh) and concentrated through triple effect evaporator and dried by followed by spray drier (SSP Pvt. Ltd., Haryana, India) to get water-soluble potassium-rich biostimulant. *G. salicornia* was further taken to manufacture three variants of potassium-rich products viz. its aqueous extract was evaporated to produce concentrated seaweed extract (CSE), potassium-rich water-soluble powder (PSP) as described earlier, and seaweed fortified granule (SFG) produced by blending residue left post aqueous extract with gypsum at 5:95 ratio and applied on cassava root at field level and studied the tuber yield and other biochemical parameters.

The physical parameters, total nitrogen and total phosphorus were determined by methods described in Fertilizer Control Order, India (1985 and elemental nutrients were analyzed according to AOAC-2016 [2] by using Atomic Absorption Spectrophotometer (Agilent 240 AA). Amino acids analysis was performed on an Agilent 1260 Infinity 1 liquid chromatography system, equipped with a 1260 Quaternary pump (G7111B), 1260 standard auto sampler (G1729A), 1260 thermo stated column compartment, 1260 diode array and multiple wavelength detector (G7115A), and a Poroshell 120 EC-C18 column (100 mm × 4.6 mm, i.e., particle size 2.7 μm) [20].

### Green Gram Seedling Bioassay Technique

A preliminary *in vitro* trial on green gram seedlings grown under plant growth chamber (Labtop Growth Chamber-LGC-200) was conducted prior to cassava field trial. The green gram seedlings cut assay technique as described by Wilson et al. [36] was followed and Long Ashton Nutrient

solution—LANS [14] was used as a standard nutrient solution for evaluating biostimulant effect of the potassium-rich products prepared in the present study. Green gram seedlings were raised in plant growth chamber maintaining a temperature of  $25 \pm 1^\circ \text{C}$ ,  $70 \pm 3\%$  of relative humidity and 16/8 light cycle ( $150 \mu \text{mol m}^{-2}$ ). The 10-day-old seedlings were taken, removed their roots and stems were transplanted to the medium containing seaweed biostimulants. The root morphology of transplanted plants grown from stem cuttings was measured after 7 days of transplantation using root imaging software (WinRhizo Pro, Regent Instruments, Canada). A completely randomized design (CRD) was chosen for the experiment with three replications with different concentration of the products and total root length (cm) and root volume ( $\text{cm}^3$ ) were measured and results were statistically drawn.

### Preparation of the Field Experiment

Cassava root *Manihot esculenta* variety Co-3 was farmed in semi-arid zone and studied its response to potassium-rich biostimulants manufactured from the red seaweed *Gracilaria salicornia*. The experiment was carried out at R&D plot of AquAgri Processing Private Limited in Manamadurai, Sivagangai Dt., Tamil Nadu, India. (Latitude is  $9^\circ 42' 56'' \text{N}$  and longitude  $78^\circ 28' 2'' \text{E}$ ) during 2018. Green manure  $5.0 \text{ tons ha}^{-1}$  was applied and plowed the plots for 3 times. Irrigation and drainage channel of 30 cm depth and 40 cm width at intervals of 80 cm was plotted across the field borders and recommended dose of fertilizer was applied to control and trial plots at the rate  $60:60:120 \text{ kg N-P}_2\text{O}_5\text{-K}_2\text{O ha}^{-1}$ .

### Designing of Trial Plot

A Randomized Complete Block Design (RCBD) was selected for the experiment consisting of 16 treatments including control viz. Control with recommended fertilizer dose-RDF (T0), Potassium-rich soluble powder-PSP (T1-T5), Concentrated seaweed extract-CSE (T6-T10) and Seaweed fortified granule-SFG (T11-T15). Agronomic practices starting from plantation of stem cutting (stakes) to harvest of tuber were followed according to standard practice laid down by Tamil Nadu Agricultural University Crop Production Guide, 2020 guide [32].

### Application of Potassium-Rich Biostimulants

Dosage and application method of three variants of potassium-rich biostimulants manufactured from *G.*

*salicornia* on cassava is as follows: The product PSP was applied three times ( $100 \text{ g} \times 3$ ;  $250 \text{ g} \times 3$ ;  $1000 \text{ g} \times 3$  times) at third month, 5th month and 7 months after plantation, two times ( $500 \text{ g} \times 2$  and  $1000 \text{ g} \times 2$ ) at third and 5th month only. Product CSE and SFG were also applied at 3rd, 5th, and 7th month after plantation of crop with different dosage level.

### Yield Attributes

After manual harvest, fresh weight of tubers obtained from plot was recorded and calculated the per hectare yield of tuber and stover. Tubers and stoves obtained from 10 random plants from different treatments and control were sundried and used to measure the biochemical composition of the tuber and nutrient use efficiency of the plants. Harvest index was calculated as below and expressed as a percentage [4].

$$\text{Harvest Index (HI)} = \frac{\text{Economic Yield}}{\text{Biological Yield}} \times 100$$

Where, the economic yield refers to tuber yield and biological yield refers to total biomass yield.

### Biochemical Parameters

Biochemical composition like starch, crude fiber, protein, fat, and ash content of tubers of 10 randomly selected plants were tested. Determination of moisture content of chips of tuber was done using a typical standard analytical method, i.e., a sample of 50 g duplicate sample was weighed and dried at  $85 \pm 2^\circ \text{C}$  for 16 h. and checked the moisture content [3]. Dry matter content (DM) was estimated by collecting a representative duplicate tuber chip of 100 g drying it in oven at  $65 \pm 2^\circ \text{C}$  for 72 h and calculated the percentage of dry matter [8]. The moisture free tuber chips were incinerated at  $750^\circ \text{C}$  for 5 h and estimated the total ash and organic content of tuber [3]. Tuber sample was defatted with petroleum ether to estimate the total fat content as described in AOAC 2000 [3]; then, defatted tuber chips were digested with 1.5%  $\text{H}_2\text{SO}_4$  in reflux chamber for 30 min [23] and determined the crude fiber percentage.

Starch content was tested according to the method of Shittu et al. [30] as follows:

$$\text{SC (\%)} = \text{SG} - 100,906/0.004845 \text{ where}$$

$$\text{Specific gravity SG} = M_a/M_w - M_w$$

$M_a$  = weight of the cassava in the air (kg);  $M_w$  = weight of the cassava in the water (kg).

Cassava mosaic disease is the common viral disease recorded in Asia [35] and its prevalence in tuber and leaves were visually examined and recorded data.

### Nutrients use Efficiencies

Nutrients contents present in the tuber and stover were estimated to calculate the nutrient uptake and nutrient harvest index [31], nutrient use efficiency (NUE), agronomic efficiency (AE), physiological efficiency (PE), agro-physiological efficiency (APE) and nutrient efficiency (NU) were estimated using the following formulae [10]:

$$\text{Agronomy Efficiency (AE)} = \frac{G_f - G_u}{N_a} \text{ kg kg}^{-1} \quad (1)$$

where  $G_f$  is the tuber yield from seaweed biostimulant + RDF applied plots (kg),  $G_u$  is the tuber yield obtained from only RDF applied plot (kg), and  $N_a$  is the quantity of nutrient applied (kg):

$$\text{Physiological Efficiency (PE)} = \frac{Y_f - Y_u}{N_f - N_u} \text{ kg kg}^{-1} \quad (2)$$

where  $Y_f$  is the total biological yield (tuber + stover) from the seaweed biostimulant + RDF applied plots (kg),  $Y_u$  is the total biological yield from only RDF applied plot (kg),  $N_f$  is the nutrient accumulation in the biological yield obtained from seaweed biostimulant + RDF applied plot (kg), and  $N_u$  is the nutrient accumulation in the biological yield of RDF given plot (kg):

$$\text{Agrophysiological Efficiency (APE)} = \frac{G_f - G_u}{N_f - N_u} \text{ kg kg}^{-1} \quad (3)$$

where  $G_r$  is the tuber yield in the seaweed biostimulant + RDF applied plot (kg),  $G_u$  is the tuber yield from RDF given plot (kg),  $N_f$  is the nutrient accumulation by stover and tuber in the seaweed biostimulant + RDF given plot (kg), and  $N_u$  is the nutrient accumulation by stover and seeds in the RDF applied plot (kg). Nutrient efficiency = AE × APE, where, AE refers to agronomy efficiency and APE refers to agro-physiological efficiency.

### Statistical Analysis

The root length, root volume, crop biometric data and yield data recorded were subjected to the statistical analysis by one-way analysis of variance (One-way ANOVA) technique that was performed using SPSS Version 22 and

mean comparison between treatments were drawn using Duncan Multiple Range Test (DMRT) with 5% error degrees of freedom.

## Results

### Green Gram Seedling Bioassay

Potassium-rich biostimulants prepared from red seaweeds shown to enhance the root growth in terms of root length, root volume, tips, links, and crossings at significant level ( $p > 0.05$ ). Total root length in treated seeds were 39 to 44 cm whereas it was 25 cm in the control seed. Similarly, the root volume ranged between 0.33 cm<sup>3</sup> to 0.38 cm<sup>3</sup> in treated seeds and it was 0.23 cm<sup>3</sup> in control one (Fig. 1).

### Nutrients Profile of Some Commercial Important Tropical Red Seaweeds Powder Prepared on Laboratory Scale

Potassium content and other mineral nutrients of some commercial important red seaweeds is given in Table 1. Organic and ash content of seaweeds tested were almost in 1:1 ratio. Organic matter of *K. alvarezii* sourced from different countries ranged between 45.21 ± 1.1 to 48.20 ± 3.8%, but it was found slightly higher in other red seaweeds tested, i.e., 51.07 ± 1.8% in *E. spinosum* (Indonesia), *G. salicornia* with 51.09 ± 3.0% (India) and 53.38 ± 1.5% of Sri Lanka, 52.39 ± 1.2% (*G. edulis*), 49.02 ± 4.8% in *G. firma* and 50.88 ± 2.5% in *G. heteroclada* of Philippines waters, but no significant difference was found between seaweeds tested. Similarly, ash content was in the range of 42.50 ± 4.3% and 50.87 ± 4.3% from all seaweeds tested without significant difference between them. Sodium chloride in *K. alvarezii* of different origins were found less than other seaweeds tested viz. it was 5.59 ± 0.5% (India), 5.35 ± 0.3% (Sri Lanka), 6.63 ± 0.4% (Philippines) and 6.45 ± 0.2% (Indonesia) and it was found significantly higher in other seaweeds like *E. spinosum* (7.82 ± 0.5%), *G. salicornia* of Indian origin (5.19 ± 0.7%), *G. salicornia* from Sri Lanka (10.12 ± 0.5%), *G. edulis* (9.75 ± 0.9), *G. firma* (10.18 ± 1.3%) and *G. heteroclada* (10.26 ± 1.8%). But in the case of potassium chloride, *K. alvarezii* sourced from all 4 countries was found to have higher ( $p < 0.05$ ) than *E. spinosum* and Gracilarian species. Thus, it was

**Table 1** Nutrients profile of some commercial important tropical red seaweeds powder prepared on laboratory scale

| Origin of seaweed/nutrient profile     | <i>Kappaphycus alvarezii</i> |             |             |             | <i>Eucheuma spinosum</i> |             | <i>Gracilaria salicornia</i> |             | <i>G. edulis</i> |             | <i>G. firma</i> |             | <i>G. heteroclada</i> |
|--|------------------------------|-------------|-------------|-------------|--------------------------|-------------|------------------------------|-------------|------------------|-------------|-----------------|-------------|-----------------------|
|  | India                        | Sri Lanka   | Philippines | Indonesia   | Indonesia                | Indonesia   | India                        | Sri Lanka   | India            | Philippines | Philippines     | Philippines |                       |
| Organic matter (%)                     | 45.47 ± 2.1                  | 48.20 ± 3.8 | 46.80 ± 2.2 | 45.21 ± 1.1 | 51.07 ± 1.8              | 51.09 ± 3.0 | 53.38 ± 1.5                  | 52.39 ± 1.2 | 49.02 ± 4.8      | 50.88 ± 2.5 |                 |             |                       |
| Moisture (%)                           | 3.66 ± 0.4                   | 3.1 ± 0.3   | 4.25 ± 0.6  | 4.05 ± 0.2  | 5.03 ± 0.7               | 3.0 ± 0.2   | 4.12 ± 0.3                   | 5.75 ± 0.4  | 4.00 ± 0.5       | 3.10 ± 0.5  |                 |             |                       |
| Ash (om dry basis) (%)                 | 50.87 ± 4.3                  | 48.70 ± 2.8 | 48.95 ± 4.5 | 50.74 ± 3.5 | 43.91 ± 2.0              | 45.89 ± 4.2 | 42.50 ± 4.3                  | 42.86 ± 2.2 | 46.98 ± 5.6      | 46.02 ± 6.6 |                 |             |                       |
| Sodium (Na) (%)                        | 2.18 ± 0.2                   | 2.08 ± 0.1  | 2.46 ± 2    | 2.51 ± 0.2  | 3.04 ± 2                 | 2.02 ± 0.3  | 3.94 ± 0.3                   | 3.80 ± 0.4  | 3.97 ± 0.5       | 4.00 ± 0.5  |                 |             |                       |
| Potassium (K) (%)                      | 17.43 ± 0.8                  | 16.66 ± 1.3 | 16.33 ± 0.6 | 17.40 ± 1.0 | 12.28 ± 2.3              | 13.47 ± 1.2 | 15.96 ± 1.8                  | 13.54 ± 0.8 | 12.29 ± 1.4      | 11.15 ± 1.0 |                 |             |                       |
| Nitrogen (N) (%)                       | 0.58 ± 0.1                   | 0.71 ± 0.0  | 0.82 ± 0.2  | 0.61 ± 0.0  | 0.75 ± 0.0               | 1.25 ± 0.2  | 1.11 ± 0.2                   | 0.21 ± 0.2  | 0.41 ± 0.0       | 0.43 ± 0.0  |                 |             |                       |
| Phosphorous (P) (%)                    | 0.05 ± 0.0                   | 0.08 ± 0.0  | 0.16 ± 0.0  | 0.12 ± 0.0  | 0.07 ± 0.0               | 0.12 ± 0.0  | 0.09 ± 0.0                   | 0.11 ± 0.0  | 0.31 ± 0.0       | 0.33 ± 0.0  |                 |             |                       |
| Calcium (Ca) (%)                       | 0.38 ± 0.0                   | 0.45 ± 0.0  | 0.39 ± 0.0  | 0.28 ± 0.0  | 1.63 ± 0.0               | 1.72 ± 0.0  | 2.22 ± 0.0                   | 2.26 ± 0.0  | 1.10 ± 0.0       | 2.15 ± 0.0  |                 |             |                       |
| Magnesium (Mg) (%)                     | 0.66 ± 0.0                   | 0.52 ± 0.0  | 0.20 ± 0.0  | 0.29 ± 0.0  | 0.85 ± 0.1               | 1.39 ± 0.0  | 1.18 ± 0.0                   | 1.10 ± 0.1  | 1.16 ± 0.0       | 1.58 ± 0.0  |                 |             |                       |
| Iron (Fe) (%)                          | 0.08                         | 0.01        | 80.11       | 0.02        | 0.02                     | 0.17        | 0.11                         | 0.17        | 1.12             | 0.76        |                 |             |                       |
| Sulfur (S) (%)                         | 3.40                         | 3.41        | 2.88        | 2.80        | 1.85                     | 2.96        | 3.05                         | 3.12        | 2.88             | 1.70        |                 |             |                       |
| Boron (B) (mg kg <sup>-1</sup> )       | 2.10                         | 1.10        | 22.12       | 1.00        | 2.39                     | 1.75        | 1.33                         | 3.06        | 4.74             | 2.81        |                 |             |                       |
| Copper (Cu) (mg kg <sup>-1</sup> )     | 0.98                         | 1.75        | 0.23        | 0.36        | 0.58                     | 0.24        | 0.33                         | 1.08        | 0.30             | 0.70        |                 |             |                       |
| Zinc (Zn) (mg kg <sup>-1</sup> )       | 1.95                         | 2.28        | 0.34        | 0.12        | 1.61                     | 1.49        | 1.50                         | 4.33        | 3.30             | 3.80        |                 |             |                       |
| Manganese (Mn) (mg kg <sup>-1</sup> )  | 21.20                        | 35.07       | 40.20       | 38.50       | 5.55                     | 17.20       | 20.50                        | 1.87        | 28.10            | 13.00       |                 |             |                       |
| Molybdenum (Mo) (mg kg <sup>-1</sup> ) | BDL                          | BDL         | BDL         | BDL         | BDL                      | BDL         | BDL                          | BDL         | BDL              | BDL         |                 |             |                       |

BDL – below detectable limit (> 0.1 mg kg<sup>-1</sup>)

33.52 ± 4.1%, 32.04 ± 3.3%, 31.40 ± 2.2% and 33.50 ± 1.3% in *K. alvarezii* from India, Sri Lanka, Philippines and Indonesia, respectively, whereas it was 33.50 ± 1.3% in *E. spinosum* and 29.39 ± 1.6%, 25.90 ± 1.9%, 26.05 ± 2.0%, 23.65 ± 3.5% and 21.44 ± 1.2% correspond to *G. salicornia* from India and Sri Lanka, *G. edulis*, *G. firma* and *G. heteroclada*, respectively, and the same trend was observed in potassium content of all seaweeds tested.

The nitrogen and phosphorous content in all seaweeds tested were found less and varied marginally between them. Thus, the nitrogen level in *K. alvarezii* sourced from 4 different countries was 0.58 ± 0.1 to 0.82 ± 0.2%, and *G. salicornia* was tested to have little higher than other seaweeds, i.e., 1.25 ± 0.2 and 1.11 ± 0.1% of Indian and Sri Lankan material respectively but it was found less than 0.5% in *G. firma*, *G. heteroclada* and *G. edulis*. The plant secondary nutrients like calcium, magnesium and sulfur were tested considerably higher in all seaweeds. *E. spinosum* and all Gracilarian species were tested to have high amount of calcium (*E. spinosum*—1.63 ± 0.0%; *G. salicornia* India—1.72 ± 0.0%; Sri Lanka—2.22 ± 0.0%, *G. edulis*—2.26 ± 0.0%; *G. firma*—1.10 ± 0.0% and *G. heteroclada*—2.15 ± 0.0%) and magnesium as well (*E.*

*spinosum*—0.85 ± 0.0%; *G. salicornia* India—1.39 ± 0.0%; Sri Lanka—1.18 ± 0.0%, *G. edulis*—1.10 ± 0.0%; *G. firma*—1.16 ± 0.0% and *G. heteroclada*—1.58 ± 0.0%) compared to *K. alvarezii* of different sources (Calcium in *K. alvarezii* of India—0.38 ± 0.0%; Sri Lanka 0.45 ± 0.0%; Philippines 0.39 ± 0.0% and Indonesia 0.28 ± 0.0%; magnesium was: India—0.66 ± 0.0%; Sri Lanka 0.52 ± 0.0%; Philippines 0.20 ± 0.0% and Indonesia 0.29 ± 0.0%). Sulfur content ranged between 1.70% and 3.4% without clear trend between different seaweeds studied. The micronutrients like iron, boron, copper zinc and manganese were found to be less but can contribute largely to the plant growth, and none of the samples tested to have molybdenum and it was below detectable level (> 0.1 mg kg<sup>-1</sup>).

### Nutrient Profile of Potassium-Rich Biostimulant Produced on Pilot Scale from Some Commercial Important Tropical Red Seaweeds

Table 2 shows nutrient profile of potassium-rich biostimulant produced on pilot scale from some commercial important tropical red seaweeds. The organic content was between 3.20 ± 1.2% to 7.39 ± 0.0% and ash content was

**Table 2** Nutrient profile of potassium-rich biostimulant produced on pilot scale from some commercial important tropical red seaweeds

| Origin of seaweeds/nutrient profile      | <i>Kappaphycus alvarezii</i> |             |             |             | <i>Euचेuma spinosum</i> | <i>Gracilaria salicornia</i> |             | <i>G. edulis</i> |
|--|------------------------------|-------------|-------------|-------------|-------------------------|------------------------------|-------------|------------------|
|  | India                        | Sri Lanka   | Philippines | Indonesia   | Indonesia               | India                        | Sri Lanka   | India            |
| Organic matter (%)                       | 3.55 ± 1.3                   | 6.22 ± 0.3  | 3.20 ± 1.2  | 4.05 ± 1.0  | 7.39 ± 0.8              | 4.20 ± 0.1                   | 4.56 ± 1.0  | 6.30 ± 0.5       |
| Moisture (%)                             | 9.32 ± 1.0                   | 7.98 ± 0.4  | 6.68 ± 0.8  | 7.79 ± 0.7  | 9.96 ± 1.1              | 7.22 ± 0.8                   | 5.44 ± 0.7  | 8.59 ± 0.3       |
| Ash (on dry basis) (%)                   | 87.13 ± 2.2                  | 85.80 ± 1.5 | 90.12 ± 1.8 | 88.16 ± 1.4 | 82.65 ± 2.6             | 88.58 ± 2.5                  | 90.00 ± 2.8 | 85.11 ± 4.3      |
| Bulk density                             | 1.05 ± 0.1                   | 1.00 ± 0.1  | 1.02 ± 0.0  | 1.05 ± 0.0  | 1.05 ± 0.1              | 1.04 ± 0.1                   | 1.04 ± 0.0  | 1.05 ± 0.0       |
| pH (1% solution)                         | 7.84 ± 0.2                   | 8.25 ± 0.1  | 7.88 ± 0.3  | 7.85 ± 0.0  | 8.50 ± 0.1              | 7.70 ± 0.2                   | 8.80 ± 0.2  | 8.10 ± 0.0       |
| Nitrogen (as N) (%)                      | 0.32 ± 0.1                   | 0.40 ± 0.0  | 0.66 ± 0.4  | 0.89 ± 0.2  | 0.65 ± 0.1              | 0.69 ± 0.3                   | 0.44 ± 0.3  | 0.50 ± 0.2       |
| Phosphorous (as P) (%)                   | 0.05 ± 0.0                   | 0.04 ± 0.0  | 0.07 ± 0.0  | 0.05 ± 0.0  | 0.11 ± 0.0              | 0.12 ± 0.0                   | 0.10 ± 0.0  | 0.05 ± 0.0       |
| Potassium (as K) (%)                     | 37.00 ± 2.1                  | 34.45 ± 1.7 | 37.90 ± 1.2 | 36.43 ± 1.6 | 35.15 ± 1.4             | 36.85 ± 1.1                  | 27.20 ± 1.1 | 25.17 ± 1.5      |
| Sulfur (as S) (%)                        | 1.89                         | 1.66        | 2.10        | 1.76        | 2.00                    | 1.81                         | 1.45        | 1.20             |
| Calcium (as Ca) (%)                      | 0.76                         | 1.10        | 0.76        | 0.80        | 1.22                    | 1.15                         | 0.65        | 0.92             |
| Magnesium (as Mg) (%)                    | 0.70                         | 0.451       | 0.30        | 0.58        | 0.82                    | 1.58                         | 1.45        | 0.77             |
| Sodium (as Na) (%)                       | 5.21 ± 0.4                   | 6.70 ± 0.4  | 6.66        | 4.70 ± 1.1  | 8.50 ± 0.8              | 6.35 ± 1.0                   | 14.02 ± 1.8 | 12.59 ± 1.2      |
| Iron (as Fe) (%)                         | 0.08                         | 0.11        | 0.10        | 0.12        | 0.38                    | 0.23                         | 0.28        | 0.25             |
| Zinc (as Zn) (mg kg <sup>-1</sup> )      | 27.40                        | 10.47       | 40.10       | 15.45       | 11.74                   | 36.51                        | 20.42       | 11.55            |
| Manganese (as Mn) (mg kg <sup>-1</sup> ) | 24.37                        | 14.00       | 11.70       | 13.30       | 1.881                   | 13.85                        | 21.19       | 0.88             |
| Copper (as Cu) (mg kg <sup>-1</sup> )    | 9.10                         | 5.52        | 3.00        | 0.59        | 14.12                   | 2.50                         | 2.22        | 8.50             |
| Boron (as B) (mg kg <sup>-1</sup> )      | 0.13                         | BDL         | BDL         | 0.18        | BDL                     | BDL                          | 0.18        | BDL              |
| Cobalt (as Co) (mg kg <sup>-1</sup> )    | 5.53                         | 11.13       | 7.00        | 9.20        | 11.90                   | 6.55                         | 5.10        | 5.11             |
| Silica (as SiO <sub>2</sub> ) (%)        | 2.82 ± 1.5                   | 3.13 ± 1.7  | 1.15 ± 0.5  | 0.88 ± 0.5  | 1.30 ± 0.6              | 1.66 ± 2.0                   | 2.55 ± 2.0  | 2.90 ± 1.4       |

BDL – below detectable limit (> 0.1 mg kg<sup>-1</sup>)

more than 85% in all the samples tested (*K. alvarezii* of India— $87.13 \pm 2.2\%$ ; Sri Lanka  $85.80 \pm 1.5\%$ ; Philippines  $90.12 \pm 1.8\%$  and Indonesia  $88.16 \pm 1.4\%$ ; *E. spinosum*— $82.65 \pm 2.6\%$ ; *G. salicornia* from India— $88.58 \pm 2.5$ ; *G. salicornia* from Sri Lanka— $90.00 \pm 2.8\%$ , *G. edulis*— $85.11 \pm 4.3\%$ ). The potassium content of *K. alvarezii* of different origin was: India— $37.00 \pm 2.1\%$ ; Sri Lanka— $34.45 \pm 1.7\%$ ; Philippines— $37.90 \pm 1.2\%$  and Indonesia— $36.43 \pm 1.6\%$ , *E. spinosum* was also tested to have high potassium content ( $35.15 \pm 1.4$ ), and in the case species of *Gracilaria*, potassium content of *G. salicornia* of Indian origin was higher than ( $36.85 \pm 1.1$ ) than one collected from Sri Lanka ( $27.20 \pm 1.1$ ) and *G. edulis* of Indian source ( $25.17 \pm 1.1$ ). But reverse trend was found in the case of sodium; thus, high level of sodium was found in *G. salicornia* of Sri Lanka ( $14.02 \pm 1.8\%$ ) and *G. edulis* ( $12.59 \pm 1.2\%$ ), whereas in *K. alvarezii* and *E. spinosum* it ranged between  $5.21 \pm 0.4\%$  and  $8.50 \pm 0.8\%$ .

Micronutrients like iron, zinc, manganese, copper, boron, and cobalt were found to present in all the samples tested but no clear trend was observed. Acid insoluble ash in the form of silicon dioxide was found considerably high level in all seaweed samples sourced from India and Sri Lanka than seaweeds of Philippines and Indonesian origin viz. *K. alvarezii* of India— $2.82 \pm 1.5\%$ ; Sri Lanka— $3.13 \pm 1.7\%$ ; *G. salicornia* of India— $1.6 \pm 2.0\%$  and Sri Lanka— $2.55 \pm 2.0\%$ ; *G. edulis*— $2.90 \pm 1.4\%$ ; *K. alvarezii* of Philippines— $1.15 \pm 0.5\%$ ; Indonesia— $0.88 \pm 0.5\%$  and *E. spinosum*— $1.3 \pm 0.65\%$ .

### Nutrient Profile of Potassium-Rich Biostimulant and Its Variants Manufactured on Commercial Scale from *Gracilaria Salicornia*

Potassium-rich water-soluble powder (PSP) and its derivatives like concentrated seaweed extract (CSE) and seaweed fortified granule (SFG) were manufactured on commercial

**Table 3** Nutrient profile of 3 variants of potassium-rich biostimulant prepared on commercial scale from red seaweed *Gracilaria salicornia*

| Physiochemical parameters               | Potassium-rich soluble powder (PSP) | Concentrated seaweed extract (CSE) | Seaweed fortified granule (SFG) |
|---|-------------------------------------|------------------------------------|---------------------------------|
| Moisture content (%)                    | $3.75 \pm 0.1$                      | $70.50 \pm 0.5$                    | $12.55 \pm 0.5$                 |
| Total organic matter (%)                | $5.28 \pm 0.6$                      | $4.65 \pm 0.7$                     | $6.10 \pm 0.4$                  |
| pH (at 1% solution)                     | $7.40 \pm 0.5$                      | $8.50 \pm 0.0$                     | $7.50 \pm 1.5$                  |
| Total soluble solids (%)                | $89.97 \pm 0.12$                    | $29.55 \pm 0.5$                    | $87.45 \pm 1.0$                 |
| Total ash content (%)                   | $84.20 \pm 0.23$                    | $24.90 \pm 1.1$                    | $81.35 \pm 1.5$                 |
| Specific gravity ( $\text{g cm}^{-3}$ ) | –                                   | $1.14 \pm 0.0$                     | –                               |
| Bulk density ( $\text{g mL}^{-1}$ )     | $0.96 \pm 0.5$                      | –                                  | $1.05 \pm 0.1$                  |
| Nitrogen (N) (%)                        | $0.80 \pm 0.0$                      | $0.19 \pm 0.0$                     | $0.11 \pm 0.0$                  |
| Phosphorous (P) (%)                     | $0.07 \pm 0.0$                      | $0.05 \pm 0.0$                     | $0.02 \pm 0.0$                  |
| Potassium (K) (%)                       | $34.90 \pm 0.5$                     | $18.20 \pm 0.3$                    | $0.22 \pm 0.2$                  |
| Calcium (Ca) (%)                        | $0.65 \pm 0.1$                      | $0.13 \pm 0.0$                     | $28.70 \pm 2.7$                 |
| Magnesium (Mg) (%)                      | $0.80 \pm 0.0$                      | $0.25 \pm 0.0$                     | $0.31 \pm 0.2$                  |
| Sulfur (S) (%)                          | $2.5 \pm 0.3$                       | $1.58 \pm 0.1$                     | $0.42 \pm 0.2$                  |
| Iron (Fe) (%)                           | $0.08 \pm 0.0$                      | $0.03 \pm 0.0$                     | $0.50 \pm 0.0$                  |
| Zinc (Zn) ( $\text{mg kg}^{-1}$ )       | $24.40 \pm 2.0$                     | $10.34 \pm 0.1$                    | $0.2 \pm 0.0$                   |
| Copper ( $\text{mg kg}^{-1}$ )          | $10.20 \pm 3.5$                     | $5.00 \pm 0.0$                     | $112 \pm 2.5$                   |
| Manganese (Mn) ( $\text{mg kg}^{-1}$ )  | $14.30 \pm 2.51$                    | $8.10 \pm 2.2$                     | $30.50 \pm 0.5$                 |
| Boron (as B) ( $\text{mg kg}^{-1}$ )    | BDL                                 | BDL                                | BDL                             |
| Cobalt (as Co) ( $\text{mg kg}^{-1}$ )  | 4.2                                 | BDL                                | BDL                             |
| Silica (as $\text{SiO}_2$ ) %           | $2.85 \pm 1.6$                      | $0.05 \pm 0.0$                     | $4.55 \pm 2.2$                  |
| Total amino acid (%)                    | $0.54 \pm 0.5$                      | $1.30 \pm 0.8$                     | $20.40 \pm 0.5$                 |
| Essential amino acids (%)               | $0.23 \pm 0.2$                      | $0.55 \pm 0.1$                     | $0.0 \pm 0.0$                   |
| Non-essential amino acids               | $0.31 \pm 0.2$                      | $0.75 \pm 0.2$                     | $0.0 \pm 0.0$                   |
| Plant growth regulators                 |                                     |                                    | $0.0 \pm 0.0$                   |
| Auxin ( $\text{mg kg}^{-1}$ )           | $0.0 \pm 0$                         | $15.00 \pm 10.0$                   | $30.35 \pm 10.5$                |
| Cytokinin ( $\text{mg kg}^{-1}$ )       | $56.7 \pm 15.0$                     | $8.50 \pm 5.0$                     | $15.12 \pm 5.5$                 |
| Gibberellins ( $\text{mg kg}^{-1}$ )    | $405.6 \pm 50.0$                    | $50.05 \pm 25.0$                   | $25.10 \pm 10.5$                |

BDL – below detectable limit ( $> 0.1 \text{ mg kg}^{-1}$ )

**Table 4** Treatment of cassava root with 3 variants of potassium-rich biostimulant prepared on commercial scale from red seaweed *Gracilaria salicornia*

| Variants of biostimulants                 | Treatment code | Dose-1 (application ha <sup>-1</sup> ) | Dose-2 (application ha <sup>-1</sup> ) | Dose-3 (application ha <sup>-1</sup> ) |
|---|----------------|--|--|--|
| Growth phase                              |                | 3 months DAP                           | 5 months DAP                           | 7 months DAP                           |
| Control (conventional RDF)                | T0             | RDF* (soil)                            | –                                      | –                                      |
| Potassium-rich water-soluble powder (PSP) | T1             | 100 g (foliar)                         | 100 g (foliar)                         | 100 g (foliar)                         |
|   | T2             | 250 g (foliar)                         | 250 g (foliar)                         | 250 g (foliar)                         |
|   | T3             | 500 g (foliar)                         | 500 g (foliar)                         | –                                      |
|   | T4             | 500 g (foliar)                         | 500 g (foliar)                         | 500 g (foliar)                         |
|   | T5             | 1000 g (foliar)                        | 1000 g (foliar)                        | –                                      |
| Concentrated seaweed extract (CSE)        | T6             | 125 mL (foliar)                        | 125 mL (foliar)                        | 125 mL (foliar)                        |
|   | T7             | 250 mL (foliar)                        | 250 mL (foliar)                        | 250 mL (foliar)                        |
|   | T8             | 500 mL (foliar)                        | 500 mL (foliar)                        | –                                      |
|   | T9             | 500 mL (foliar)                        | 500 mL (foliar)                        | 500 mL (Foliar)                        |
|   | T10            | 1000 mL (foliar)                       | 1000 mL (Foliar)                       | –                                      |
| Seaweed fortified granule (SFG)           | T11            | 4.0 kg (soil)                          | 4.0 kg (soil)                          | 4.0 kg (soil)                          |
|   | T12            | 8.0 kg (soil)                          | 8.0 kg (soil)                          | 8.0 kg (soil)                          |
|   | T13            | 10.0 kg (soil)                         | 10.0 kg (soil)                         | 10.0 kg (soil)                         |
|   | T14            | 15.0 kg (soil)                         | 15.0 kg (soil)                         | –                                      |
|   | T15            | 15.0 kg (soil)                         | 15.0 kg (soil)                         | 15.0 kg (soil)                         |

RDF-recommended dose of fertilizer; DAP-day after plantation; All treatments were given along with Recommended Dose of Fertilizer (T0)

\*RDF – NPK: 90–90–240 kg ha<sup>-1</sup>

scale from *G. salicornia* of Indian origin and their nutrient contents is given in Table 3 and these three products were applied on cassava root at field level as described in Table 4. Macro- and micronutrients of PSP manufactured on commercial scale was like one prepared on pilot scale (Table 2) without significant differences. Further, commercially manufactured SPS was tested to contain 0.54% of total amino acids (essential amino acid 0.23% and non-essential amino acid 0.31%) and 463.3 mg L<sup>-1</sup> of plant growth regulators (cytokinin 56.7 mg L<sup>-1</sup>, gibberellic acids 405.6 mg L<sup>-1</sup>).

The product CSE contained 29.55% total soluble solid which comprised of 4.65% organic matter and 24.90% ash content. The NPK in CSE were 0.19%, 0.05% and 18.20%, respectively, and the values 0.13% 0.25% and 1.58% corresponded to secondary mineral nutrients like calcium, magnesium, and sulfur. Product CSE also contained considerable amount of micronutrients (iron 300 mg L<sup>-1</sup>; zinc 10 mg L<sup>-1</sup>; copper mg L<sup>-1</sup>; manganese 8 mg L<sup>-1</sup>) with 1.30% of total amino acid (0.55% of essential amino + 0.75% non-essential amino acid) and 73.55 mg L<sup>-1</sup> of plant growth regulators (auxin 15.0 mg L<sup>-1</sup>, cytokinin 8.5 mg L<sup>-1</sup>, gibberellic acids 50.5 mg L<sup>-1</sup>) (Table 3).

Total organic content of SFG was 6.10% with 87.45% of ash, primary nutrients nitrogen, phosphorous and potassium were 0.11%, 0.02% and 0.22%, respectively, and 28.70%, 0.31% 0.42% corresponded to calcium

magnesium and sulfur. No amino acid was found in SFG but growth promoters like auxin, cytokinin and gibberellic acid were analyzed at level of 30 mg L<sup>-1</sup>, 15 mg L<sup>-1</sup> and 25 mg L<sup>-1</sup>, respectively (Table 3).

#### Tuber and Stover Yield of Cassava Root Treated with Potassium-Rich Biostimulants Manufactured from *G. salicornia*

Tubers were manually harvested, removed the dirt, and recorded the fresh weight. The total yield obtained from control plant (T0) was 24.50 t ha<sup>-1</sup> (stover yield 18.24 t ha<sup>-1</sup> and HI 57.32%) and the yield from PSP treated plants (T1 to T5) ranged from 23.40 t ha<sup>-1</sup> to 29.90 t ha<sup>-1</sup>, i.e., 22.04% higher than control in T3 plant (stover yield in T1 to T5: 17.15, 17.05, 18.20, 20.10 and 21.45 t ha<sup>-1</sup>, respectively) with HI ranging from 57.71 to 62.16. The tuber yield from CSE applied plant was: T6-22.05 t ha<sup>-1</sup>, T7-23.56 t ha<sup>-1</sup>, T8-28.60 t ha<sup>-1</sup>, T9-27.84 t ha<sup>-1</sup> and T-10—27.42 t ha<sup>-1</sup> with 17.40% higher tuber yield from T9 plants and stover yield obtained T6 – T10 were 15.44, 15.57, 18.52, 18.65 and 16.50 t ha<sup>-1</sup>, respectively, with 22.19% more tuber yield over control was recorded from T15 plants. High range of HI recorded from trial T2 (HI-61.19%) T3 (62.16), T5 (59.44%) of SPS treated, T8 to T10 of CSE treated plants and T13 & T14 of SFG plants (p > 0.05) (Table 5).



**Table 5** Tuber and stover yield of cassava root treated with potassium-rich biostimulants prepared from *Gracilaria salicornia*

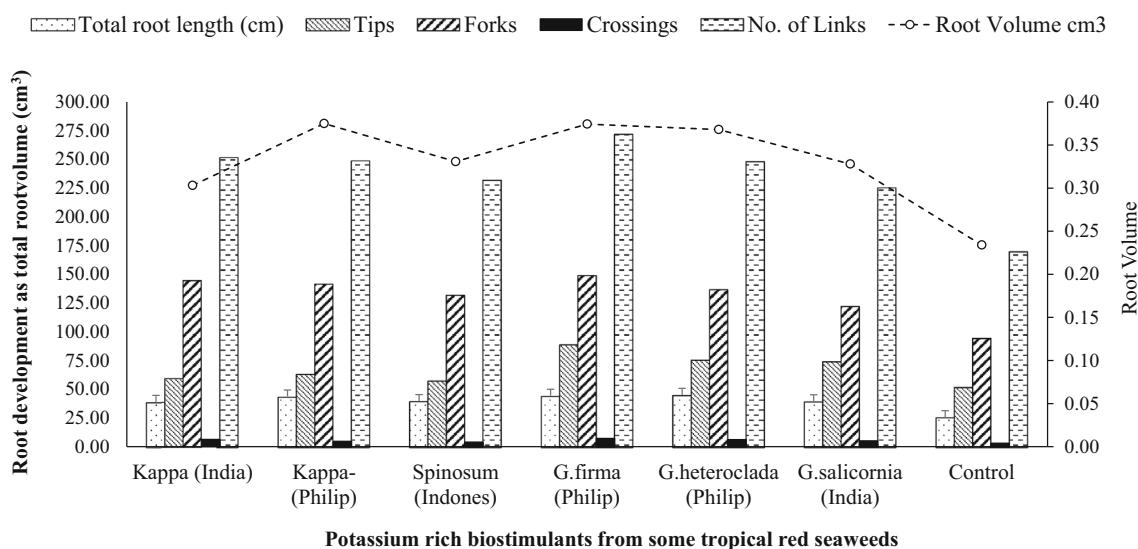
| Variants of biostimulants           | Treatment code | Tuber yield (ton ha <sup>-1</sup> ) | Stover yield (ton ha <sup>-1</sup> ) | HI                  |
|-------------------------------------|----------------|-------------------------------------|--------------------------------------|---------------------|
| Control (Conventional RDF)          | T0             | 24.50 ± 2.1                         | 18.24 ± 1.1                          | 57.32               |
| Potassium-rich soluble powder (PSP) | T1             | 23.40 ± 1.8                         | 17.15 ± 2.3                          | 57.71               |
|                                     | T2             | 26.88 ± 2.4                         | 17.05 ± 2.0                          | 61.19 <sup>ab</sup> |
|                                     | T3             | 29.90 ± 2.8 <sup>ab</sup>           | 18.20 ± 3.2                          | 62.16 <sup>ab</sup> |
|                                     | T4             | 27.30 ± 2.5 <sup>ab</sup>           | 20.10 ± 2.1                          | 57.59               |
|                                     | T5             | 28.50 ± 2.5 <sup>ab</sup>           | 21.45 ± 2.5                          | 59.44 <sup>ab</sup> |
| Concentrated seaweed extract (CSE)  | T6             | 22.05 ± 2.4                         | 15.44 ± 1.8                          | 58.82               |
|                                     | T7             | 23.56 ± 2.1                         | 15.57 ± 2.1                          | 60.2 <sup>ab</sup>  |
|                                     | T8             | 28.60 ± 1.8 <sup>ab</sup>           | 18.52 ± 2.1                          | 60.70 <sup>ab</sup> |
|                                     | T9             | 27.84 ± 2.0 <sup>ab</sup>           | 18.65 ± 2.6                          | 59.88               |
|                                     | T10            | 27.42 ± 2.1 <sup>ab</sup>           | 16.50 ± 2.6                          | 62.43 <sup>ab</sup> |
| Seaweed fortified granule (SFG)     | T11            | 24.10 ± 2.3                         | 17.28 ± 2.2                          | 58.24               |
|                                     | T12            | 27.16 ± 2.1 <sup>ab</sup>           | 18.28 ± 2.5                          | 59.77 <sup>ab</sup> |
|                                     | T13            | 30.10 ± 1.8 <sup>ab</sup>           | 17.55 ± 1.3                          | 63.17 <sup>ab</sup> |
|                                     | T14            | 31.18 ± 2.4 <sup>ab</sup>           | 19.25 ± 2.0                          | 61.83 <sup>ab</sup> |
|                                     | T15            | 30.05 ± 2.4 <sup>ab</sup>           | 19.90 ± 1.4                          | 60.16 <sup>ab</sup> |

All treatments were given along with RDF (T0); Values followed by different letters are significantly different at  $p < 0.05$

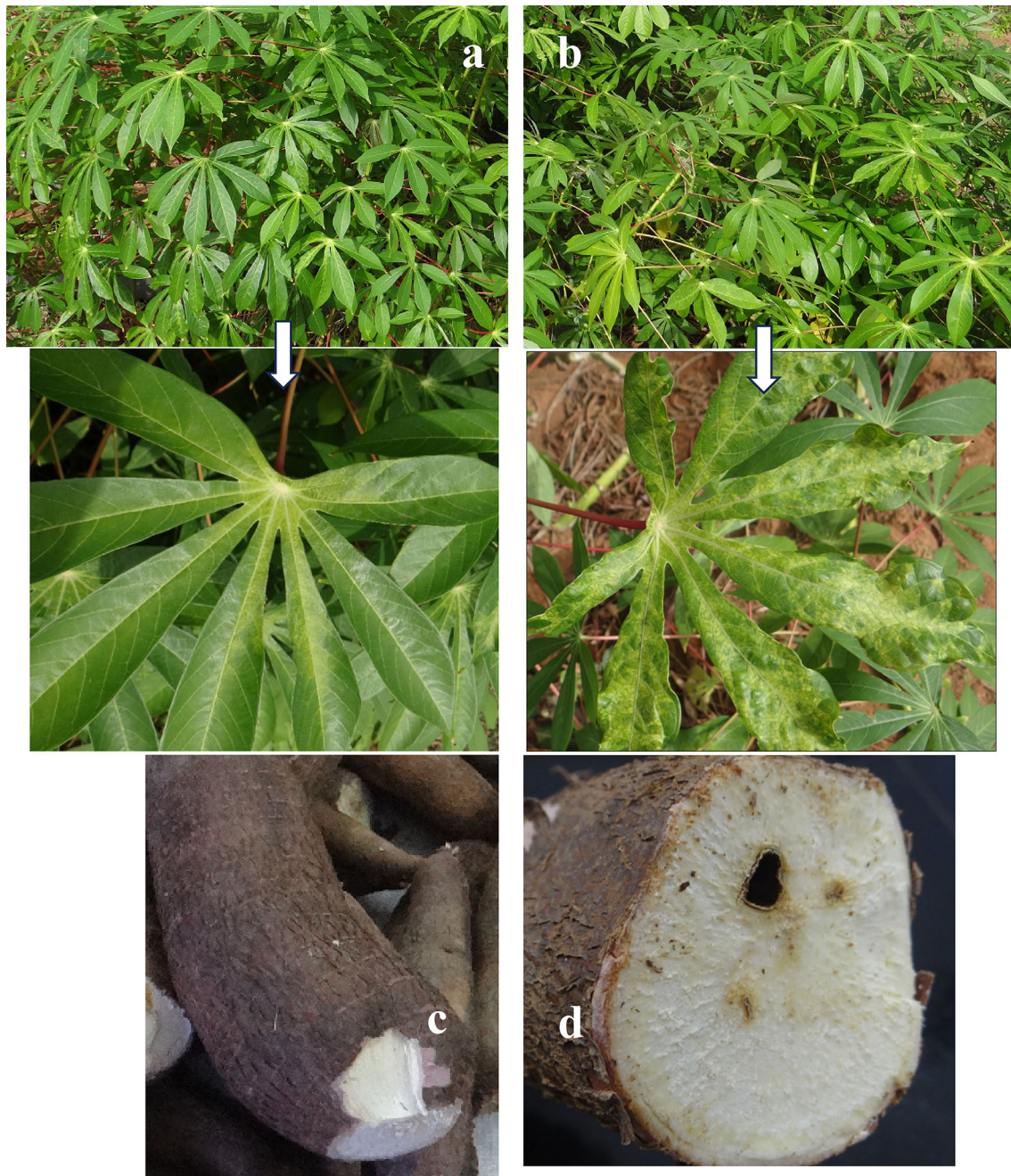
### Biochemical Composition of Cassava Tuber Treated with Potassium-Rich Biostimulants Manufactured from *G. salicornia*

Moisture content of fresh tubers ranged between  $32.44 \pm 2.2\%$  and  $37.10 \pm 3.1\%$  and in dry tubers it was from  $5.83 \pm 0.4\%$  to  $9.27 \pm 0.8\%$ . The starch content estimated on dry weight basis was  $80.14 \pm 1.2\%$  in control plants; the plants treated with SPS gave tuber with highest starch content, viz.  $86.1 \pm 1.8\%$ ,  $85.0 \pm 4.5\%$  and

$81.1 \pm 2.8\%$  from T3, T4 and T5, respectively, and thus, the higher starch yield over control was obtained from T5 which is 7.78% followed by T3 (7.44%) and T4 (6.06%) and T4 (6.06%). Plants from T8, T9 and T10 trial responded well to CSE and tuber yield increase over control was 7.08%, 7.09% and 9.12%, respectively ( $p > 0.05$ ). Seaweed fortified granule which was given to the plants through soil performed better than SPS and CSE which were applied through foliar method, and the results from T11 and T12 plants were not at significant level and T13,



**Fig. 1** Effect of potassium-rich biostimulant prepared from some commercial important tropical red seaweeds on the root development of green gram

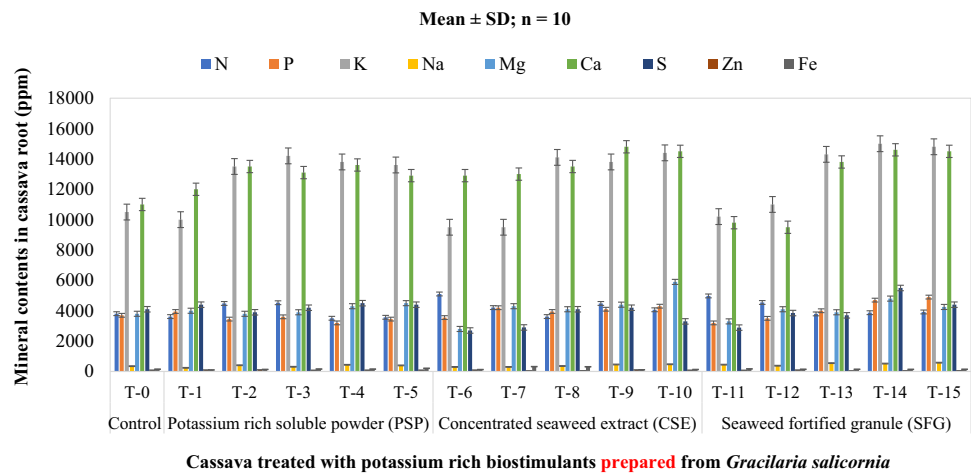


**Fig. 2** Difference between control and treated cassava root **a** Treated leaf found with less incidence of CMD; **b** Control leaf found with cassava mosaic virus disease (CMD); **c** Good quality tuber from treated plant; **d** Symptoms of CBSD in the tubers of control plant

T14 and T15 had produced increased tuber yield (8.62%, 10.31% and 10.31%, respectively). Higher ash content was recorded from plants which gave significantly higher tuber yield, i.e., T3 (3.17%) and T4 (3.34%) and T5 (2.90%) of SPS applied plant, T8 (3.17%) and T10 (3.14%) from CSE applied plant and all granule treated plants (T11 to T15). Other biochemical parameters like crude fiber, protein and fat were recorded but no trend with respect to tuber yield

was observed (Table 6). The occurrence of cassava mosaic disease (CMD) on the leaves of control plants was  $18.5 \pm 4.5\%$  and  $16.2 \pm 5.0\%$  in the tubers, plants treated with SPS and CSE (foliar products) had less incidence of CMD on leaves (5.0–10.5%) and tuber (8–12%) (Fig. 2) whereas plants which were applied with SFG through soil application found to have significantly less incidence of CMD in tuber (3.3–6.7%) but without much impact on the

**Fig. 3** Nutrients content of Cassava tuber treated with potassium-rich biostimulants manufactured from red seaweed *Gracilaria salicornia*



**Table 6** Biochemical composition of cassava tuber treated with potassium-rich biostimulant and its derivatives prepared from *G. salicornia*

| Variants of biostimulants           | Treatment code | Moisture of fresh tuber | Moisture of dry tuber chips (%) | Starch (%)                    | Crude fiber (%) | Protein (%)    | Fat %          | Ash (%)                      |
|-------------------------------------|----------------|-------------------------|---------------------------------|-------------------------------|-----------------|----------------|----------------|------------------------------|
| Control (conventional RDF)          | T0             | 33.40 $\pm$ 3.2         | 7.32 $\pm$ 0.5                  | 80.14 $\pm$ 6.2               | 2.65 $\pm$ 0.2  | 2.38 $\pm$ 0.5 | 1.48 $\pm$ 0.0 | 2.67 $\pm$ 0.1               |
| Potassium-rich soluble powder (PSP) | T1             | 34.15 $\pm$ 2.3         | 6.52 $\pm$ 0.5                  | 77.12 $\pm$ 7.2               | 2.66 $\pm$ 0.3  | 1.62 $\pm$ 0.2 | 0.86 $\pm$ 0.0 | 2.78 $\pm$ 0.3               |
|                                     | T2             | 33.44 $\pm$ 3.2         | 5.95 $\pm$ 0.7                  | 82.80 $\pm$ 7.1               | 2.38 $\pm$ 0.2  | 2.80 $\pm$ 0.5 | 1.22 $\pm$ 0.4 | 2.15 $\pm$ 0.1               |
|                                     | T3             | 35.07 $\pm$ 4.1         | 7.33 $\pm$ 0.8                  | 86.10 $\pm$ 5.5 <sup>ab</sup> | 2.75 $\pm$ 0.2  | 2.83 $\pm$ 0.5 | 1.06 $\pm$ 0.8 | 3.18 $\pm$ 0.1 <sup>ab</sup> |
|                                     | T4             | 32.90 $\pm$ 3.9         | 8.82 $\pm$ 0.5                  | 85.00 $\pm$ 4.4 <sup>ab</sup> | 2.38 $\pm$ 0.1  | 3.19 $\pm$ 0.1 | 1.12 $\pm$ 0.3 | 3.34 $\pm$ 0.1 <sup>ab</sup> |
|                                     | T5             | 37.10 $\pm$ 1.2         | 7.58 $\pm$ 0.6                  | 87.18 $\pm$ 2.8 <sup>ab</sup> | 2.44 $\pm$ 0.0  | 2.23 $\pm$ 0.0 | 0.88 $\pm$ 0.1 | 2.90 $\pm$ 0.2 <sup>ab</sup> |
| Concentrated seaweed extract (CSE)  | T6             | 34.15 $\pm$ 3.5         | 7.06 $\pm$ 1.3                  | 77.46 $\pm$ 6.1               | 2.88 $\pm$ 0.1  | 3.19 $\pm$ 0.1 | 1.70 $\pm$ 0.5 | 2.20 $\pm$ 0.2               |
|                                     | T7             | 32.44 $\pm$ 2.3         | 8.61 $\pm$ 0.8                  | 84.00 $\pm$ 8.1               | 2.75 $\pm$ 0.9  | 1.63 $\pm$ 0.4 | 0.71 $\pm$ 0.2 | 2.46 $\pm$ 0.2               |
|                                     | T8             | 36.07 $\pm$ 3.0         | 7.55 $\pm$ 1.7                  | 85.81 $\pm$ 7.8 <sup>ab</sup> | 2.29 $\pm$ 0.1  | 2.26 $\pm$ 0.4 | 1.28 $\pm$ 0.0 | 3.17 $\pm$ 0.1 <sup>ab</sup> |
|                                     | T9             | 32.90 $\pm$ 2.2         | 6.69 $\pm$ 1.0                  | 85.82 $\pm$ 4.2 <sup>ab</sup> | 2.98 $\pm$ 0.2  | 2.80 $\pm$ 0.1 | 0.88 $\pm$ 0.0 | 2.88 $\pm$ 0.0               |
|                                     | T10            | 37.10 $\pm$ 3.1         | 7.89 $\pm$ 0.5                  | 87.45 $\pm$ 5.2 <sup>ab</sup> | 2.44 $\pm$ 0.2  | 2.54 $\pm$ 0.0 | 1.08 $\pm$ 0.2 | 3.14 $\pm$ 0.3 <sup>ab</sup> |
| Seaweed fortified granule (SFG)     | T11            | 35.15 $\pm$ 2.8         | 7.95 $\pm$ 1.4                  | 74.76 $\pm$ 6.2               | 1.94 $\pm$ 1.0  | 3.11 $\pm$ 0.1 | 0.93 $\pm$ 0.1 | 3.10 $\pm$ 0.1 <sup>ab</sup> |
|                                     | T12            | 33.18 $\pm$ 2.2         | 9.27 $\pm$ 0.8                  | 82.55 $\pm$ 7.1               | 2.21 $\pm$ 0.2  | 2.84 $\pm$ 0.1 | 0.76 $\pm$ 0.0 | 2.84 $\pm$ 0.2 <sup>ab</sup> |
|                                     | T13            | 32.00 $\pm$ 1.6         | 7.23 $\pm$ 1.0                  | 87.05 $\pm$ 9.5 <sup>ab</sup> | 2.25 $\pm$ 0.5  | 2.37 $\pm$ 0.0 | 1.20 $\pm$ 0.0 | 3.33 $\pm$ 0.7 <sup>ab</sup> |
|                                     | T14            | 36.60 $\pm$ 2.3         | 5.83 $\pm$ 0.4                  | 88.40 $\pm$ 6.4 <sup>ab</sup> | 2.03 $\pm$ 0.2  | 2.42 $\pm$ 0.2 | 0.44 $\pm$ 0.0 | 3.50 $\pm$ 0.2 <sup>ab</sup> |
|                                     | T15            | 35.32 $\pm$ 3.3         | 7.06 $\pm$ 0.6                  | 88.40 $\pm$ 8.2 <sup>ab</sup> | 2.16 $\pm$ 0.1  | 1.45 $\pm$ 1.2 | 1.25 $\pm$ 0.1 | 3.25 $\pm$ 0.8 <sup>ab</sup> |

All treatments were given along with RDF (T0); Values followed by different letters are significantly different at  $p < 0.05$

leaves (10.5–15.0%). Mineral nutrients like K, Na, Mg, Ca, Fe, S and Zn of tubers obtained from treated along with control plant are shown in Fig. 3. Results of nutrient use efficiency of plants applied with seaweed biostimulant against control plant are given in Table 7. There was improved agronomy efficiency (AE), physiological efficiency (PE) and agro-physiological efficiency (APE) at significant level with estimation of 23.63% to 40.36% of more nutrient uptake in the treated plants.

## Discussion

Traditional agriculture has become the most-extensive and expensive food-producing segment in the world. Cassava is an important crop among tropical root crops, and it has multiple end-uses such as food, animal feed, and raw material for many industries; hence, the demand for this commodity is likely to increase. Though cassava is a drought-tolerant and water-efficient crop, being also

**Table 7** Nutrient use efficiency of cassava root applied with Recommended Dose of Fertilizer (RDF) and different variants of potassium-rich biostimulants prepared from red seaweed *Gracilaria salicornia*

| Variants of biostimulants           | Treatment code | Total nutrient uptaken (kg ha <sup>-1</sup> ) | Biological yield (kg ha <sup>-1</sup> ) | Tuber yield | Stover yield | AE                  | PE                 | APE                | UE                 | Nutrient uptake %   |
|-------------------------------------|----------------|---|---|-------------|--------------|---------------------|--------------------|--------------------|--------------------|---------------------|
| Control (conventional RDF)          | T0             | 105.8   | 42.74                                   | 24.5        | 18.24        |                     |                    |                    |                    |                     |
| Potassium-rich soluble powder (PSP) | T1             | 110.5   | 40.55                                   | 23.4        | 17.15        | 23.30               | - 0.47             | - 0.23             | - 5.45             | 44.08               |
|                                     | T2             | 122.4   | 43.93                                   | 26.8        | 17.05        | 26.78               | 0.07               | 0.14               | 3.84 <sup>ab</sup> | 46.04               |
|                                     | T3             | 138.6   | 48.10                                   | 29.9        | 18.20        | 29.80 <sup>ab</sup> | 0.16               | 0.16               | 4.91 <sup>ab</sup> | 51.00               |
|                                     | T4             | 135.8   | 47.40                                   | 27.3        | 20.10        | 27.20               | 0.16               | 0.09               | 2.54               | 57.75               |
|                                     | T5             | 136.0   | 49.95                                   | 28.5        | 21.45        | 28.40 <sup>ab</sup> | 0.24 <sup>ab</sup> | 0.13               | 3.76 <sup>ab</sup> | 56.58               |
| Concentrated seaweed extract (CSE)  | T6             | 130.8   | 37.44                                   | 22.0        | 15.44        | 21.90               | - 0.21             | - 0.10             | - 2.19             | 56.67               |
|                                     | T7             | 138.5   | 39.13                                   | 23.6        | 15.57        | 23.46               | - 0.11             | - 0.03             | - 0.67             | 54.50               |
|                                     | T8             | 146.1   | 47.12                                   | 28.6        | 18.52        | 28.50 <sup>ab</sup> | 0.11 <sup>ab</sup> | 0.10               | 2.90 <sup>ab</sup> | 57.71               |
|                                     | T9             | 143.1   | 46.49                                   | 27.8        | 18.65        | 27.74               | 0.10               | 0.09               | 2.48               | 60.88 <sup>ab</sup> |
|                                     | T10            | 147.6   | 43.92                                   | 27.4        | 16.50        | 27.32               | 0.03               | 0.07               | 1.91               | 59.63 <sup>ab</sup> |
| Seaweed fortified granule (SFG)     | T11            | 142.1   | 41.38                                   | 24.1        | 17.28        | 24.00               | - 0.04             | - 0.01             | - 0.26             | 61.50 <sup>ab</sup> |
|                                     | T12            | 147.8   | 45.44                                   | 27.1        | 18.28        | 27.06               | 0.06               | 0.06               | 1.71               | 59.21 <sup>ab</sup> |
|                                     | T13            | 145.5   | 47.65                                   | 30.1        | 17.55        | 30.00 <sup>ab</sup> | 0.12 <sup>ab</sup> | 0.14 <sup>ab</sup> | 4.23 <sup>ab</sup> | 61.58 <sup>ab</sup> |
|                                     | T14            | 148.5   | 50.43                                   | 31.1        | 19.25        | 31.08 <sup>ab</sup> | 0.18 <sup>ab</sup> | 0.16 <sup>ab</sup> | 4.86 <sup>ab</sup> | 60.63 <sup>ab</sup> |
|                                     | T15            | 142.0   | 49.95                                   | 30.5        | 19.90        | 29.95 <sup>ab</sup> | 0.20 <sup>ab</sup> | 0.15 <sup>ab</sup> | 4.59 <sup>ab</sup> | 61.88 <sup>ab</sup> |

Significant values ( $p < 0.05$  by LSD) are indicated with different letters; AE-agronomy efficiency, PE-physiological efficiency, APE-agro-physiological efficiency, UE-utilization efficiency; total fertilizer applied was 240 kg ha<sup>-1</sup>

exceptionally adapted to high soil acidity, it requires moderate amount of NPK nutrient to produce commercial yield. Cassava is a potassium loving crop; thus, enough K is required to be applied. The positive response of cassava yields to K application has been well documented [4, 25]. Also, the significant reduction in cassava yield in the absence of K fertilization during five consecutive cropping years was clearly demonstrated [8]. Furthermore, this yield reduction was considerably restrained by K application. Potassium fertilizers are commonly referred to as potassium and their content is measured as K<sub>2</sub>O. The K<sub>2</sub>O content of muriate potassium and sulfate of potassium is 60% and 50% respectively, in the present study, whole seaweed powder of *K. alvarezii* sourced from different countries had 16–17% of potassium, i.e., 19–20% K<sub>2</sub>O whereas other commercial important red seaweeds like *Eucheuma* and *Gracilarian* species were found to have 13.2–19.2% of K<sub>2</sub>O, but the powder of water-soluble extract of all red seaweed species studied contained 30 to 44% of K<sub>2</sub>O; therefore, this could a good source of natural potassic nutrient for cassava production. Both foliar and root application seaweed biostimulants tested in the present study produced a measurable response in cassava root but in varying level. Seaweed fortified granule (SFS) that was given through root had influenced at higher level on tuber yield as compared to SPS and CWE which were given through foliar mode. Application of SFS at 3 × 10 kg

(T13), 2 × 15 kg (T14) and 3 × 15 kg (T15) produced significant amount of tuber yield (20.62, 22.19 and 17.80%, respectively) with high starch content, whereas the increase in tuber yield from plants of SPS and CSE treated was 20.09% and 17.40%, respectively. It was also noticed that yield increase was not proportionately related to the volume of input of any biostimulant applied. The yield recorded from the plants applied through root application was high and it could be due to fact that it could have stimulated the plant and root growth more than foliar products. Plants treated with seaweed extract showed higher harvest index, yield, mineral contents, and biochemical constituents against control. Seaweed extracts have been demonstrated to serve as elicitors to plant defense responses against harmful microbial pathogens including virus thereby protecting crops from diseases and economic losses [16, 19, 34]. The less degree of infection in crops treated with seaweed extract is due to a general improvement of plant vigor, preformed resistance, induced systemic or systemic acquired resistance. Vera et al. reported that symptoms of the tomato chlorotic dwarf viroid and tobacco mosaic virus (TMV) disease in tobacco were significantly reduced when they treated with sulfated polysaccharides extracted from red seaweeds [34]. Similarly, when tobacco was applied with oligosaccharides extracted from seaweeds, a significant reduction of symptoms caused by tobacco mosaic virus was recorded

[16, 19]. In the present study, it was observed that incidence of cassava mosaic virus was found significantly less in treated plants, and it could be because seaweed extract might have improved the overall health and defense mechanism of plant against cassava mosaic virus.

Tubers obtained from treated plants had higher level of mineral ( $p > 0.05$ ); particularly, mineral content of tuber from plants treated through root application was found higher; therefore, it can be assumed that seaweed biostimulants has potential to increase the nutrient use efficiency of cassava significantly (Fig. 3).

Much work was not done on the effect seaweed extract on the cassava roots, but a few reports are available on its positive effect on the other tuber crop like potato [7, 13, 26]. Seaweed extract applied at two stages of growth (30 and 60 days after sowing) performed better and produced higher crop yield in potato [13]. A foliar application of product containing beneficial bacteria and humic acids at low concentrations resulted in a significant yield increase in cassava yield combined with low fertilization requirements [5]. Potassium contents were found to increase in the potato yield [13]. The response of cassava to potassium, nitrogen and phosphorus was studied by Canellas et al. and they found that K fertilizer was necessary for achieving higher tuber yields and a minimum of 30 kg ha<sup>-1</sup> of K in the presence of N and P was necessary to produce a significant result which confirms the importance K for cassava production [5]. There was improved agronomy efficiency (AE) found in all treated plants, but significant physiological efficiency was recorded only in a few plants (T3-T5, and T13-T15); similarly, agro-physiological efficiency (APE) was observed with T3, T4, T6 and T13 to T15. Total nutrient uptake in control plant estimated was 44.08%, whereas in plants of SFG treated, it was 59.21–61.88%, i.e., 34.31–40.36% higher nutrient use efficiency over control plant was observed. T3, T7–T10 plants were also found with significant level of nutrient uptake (13.67–17.42%).

## Conclusions

It is important to enhance the productivity of crops in limited growing area to satisfy the increased demand of foods. In this study, potassium-rich biostimulants manufactured from red seaweed *G. salicornia* when applied on cassava at minimal dosage level (500 g/spray × 3); a significant improvement in tuber yield, quality and nutrients use efficiency was observed. Potassic fertilizers are manufactured through chemical process, and it leads to more carbon foot printing on the environment. Seaweeds are considered as one of the major sources of food, feed and valuable chemicals, and currently, more attention is paid to promote the seaweed aquaculture around the world to meet

the demand. In the present investigation, red seaweeds are found to have considerable amount of potassium and its application on cassava crop could improve tuber yield with starch content and improved the nutrient use efficiency and less incidence of CMD in cassava.

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**Data Availability** The datasets generated during and/or analyzed during the present study are not publicly available due to the further research is still ongoing but are available from the corresponding author on request.

**Declarations**

**Conflict of interest** The authors declare no competing interests.

**Ethics Approval** Not applicable.

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