



**American Journal of Agricultural Research**  
**(ISSN:2475-2002)**



## **Bio-stimulant of seaweed source as an organic alimentative to Bellary onion: Bulb yield and Pyruvic acid levels**

**Kosalaraman Karthikeyan, Munisamy Shanmugam\***

Research and Development Division, AquAgri Processing Private Limited, B5, SIPCOT Industrial Estate, Manamadurai, Sivagangai District, Tamil Nadu, India.

### **ABSTRACT**

The influence of bio-stimulant Aquasap powder derived from the red seaweed *Kappaphycus alvarezii* was applied at 0.2% through foliar application on bellary onion at the establishment, vegetative, bulb formation and bulb development stage. Bulb yield and quality parameters such as total soluble solid, moisture content, ash content, organic content, pyruvic acid content, physiological loss and rotting percentage were studied. The crop yield in terms of bulb in treated plant was 31.65% over control with no statistical significant in the content of pyruvic between treated and control plants.

**Keywords:** *Kappaphycus alvarezii*, Onion, Bio-stimulant, Pyruvic acid, Organic nutrient, Aquasap powder.

### **\*Correspondence to Author:**

Munisamy Shanmugam  
Research and Development Division, AquAgri Processing Private Limited, B5, SIPCOT Industrial Estate, Manamadurai, Sivagangai District, Tamil Nadu, India. E-mail: [m.shanmugam@aquagri.in](mailto:m.shanmugam@aquagri.in)

### **How to cite this article:**

Kosalaraman Karthikeyan and Munisamy Shanmugam. Factors Bio-stimulant of seaweed source as an organic alimentative to Bellary onion: Bulb yield and Pyruvic acid levels. American Journal of Agricultural Research, 2016,1(2): 0001-0008.

**eSciencePublisher**

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## 1. INTRODUCTION

Nowadays organic farmers turn their sight towards manure, compost and organic fertilizer like seaweed which has extra amino acid, micro and macro nutrient. Purchasing and transporting of chemical fertilizer such as urea, DAP are bit expensive whereas seaweed acts as an alternate to conventional chemical fertilizer. The use of marine macro algae as fertilizer in crop production has been a long tradition in coastal areas all over the world. In India, huge quantity of macroscopic marine algae has been utilized directly as manure or in the form of compost by coastal people.

Liquid extract obtained from seaweeds have recently gained importance as foliar sprays for many crops including cereals, pulses, flowers, vegetables and spice. Seaweed extracts stimulate various aspects of growth and development resulting in around good health of the plants, while deliberating the effect of seaweed extracts on crops for aspects of root development and mineral absorption, shoot growth and photosynthesis and ultimately crop yield, even vegetative propagation can also be taken into consideration (Pramanick et al., 2013). In India, vegetable production was around 146.55 million tons for an area of 8.5 million hectare. The 4 major vegetables viz. potato (28.9%), tomato (11.3%), onion (10.3%) and brinjal (8.1%) were contributing 58.6% of total vegetable production in our country. Onion is the one of the major vegetable of India. In India production of onion in 2013 -2014 was nearly 163.09 lakh ton from an area of 9.56 lakh hectares. Tamil Nadu contributes 472.69 metric ton from an area of 39.97 hectares (ICAR-DOGR report). ([http://www.dogr.res.in/index.php?option=com\\_content&view=article&id=94&Itemid=98&Lang=en](http://www.dogr.res.in/index.php?option=com_content&view=article&id=94&Itemid=98&Lang=en)). The bio-stimulant Aquasap powder is rich in potash with high plant growth hormones such as auxin, cytokinin and gibberellins along with other primary and secondary nutrients (Table 1). The present experimental study describes the influence of bio-stimulant powder derived from *K. alvarezii* cultivated in Indian waters as an organic nutrient on the growth, quality and yield of onion.

## 2. MATERIAL AND METHODS

### 2.1. Preparation of trial plots

Seed of onion (Agri Found Dark Red) was sourced from Tamil Nadu Agriculture University,

Coimbatore. Experiment of onion trial was conducted in Agri R&D plot of AquAgri Processing Private Limited, Manamadurai, Sivagangai district, Tamil Nadu (Lat. 9° 42'55"N, Long. 79° 28'22"E). Farmyard was applied about 25 tonnes per ha ratio and ploughed 3 times and rotovator was used to break the soil finely. The size of the plots made was 12.6 m×5.8 m. The nurseries created were transplanted in the field with distance of 20cm and irrigated weekly intervals by flooding method.

### 2.2. Application of Aquasap powder

Commercially manufactured Aquasap powder (Batch no: 09102011-11) was collected from stock of AquAgri Processing Private Limited, India. Bellary onion was treated with 0.2% Aquasap powder at 4 times through foliar application. First dosage of foliar spray was applied in establishment stage of 10-15<sup>th</sup> day of sowing. Second, third and final dosages were given during vegetative (35-40 d), bulb formation (60-65 d) stage and bulb development stage (75-80 d) respectively (Karthikeyan and Shanmugam 2016 b).

The onion was harvested when it was about 75% of leaves got dried off then the bulbs were plucked manually. The data on plant weight, plant length, nos. of root, root length, nos. of leaves, leaves weight, leaves length, leaves thickness, bulb weight, diameter of bulb, diameter of bulb neck, total soluble solid, moisture content, ash content, organic content, pyruvic acid content, physiological loss and rotting percentage were recorded.

### 2.3. Analysis of physiochemical parameters

#### 2.3.1. Total solid and moisture content

Bulb was weighed and crushed to get juice through musclin cloth and centrifuged and solid content of liquid was estimated by gravimetric method. Onion was sliced into 0.5 to 1 cm sizes and dried at 80°C for 4 hrs to calculate its moisture content.

#### 2.3.2. Organic matter

5 g of the dry sample was measured into a previously weight porcelain crucible. The sample was burnt to ashes in the muffle furnace at 550°C for 4 hours. When it had become completely ashes, it was cooled in the desiccator and organic and ash content was estimated.

#### 2.3.3. Estimation of pyruvic acid

Pyruvic acid level of onion obtained from treated and control plants were estimated as per the

**Table 1:** Physico-chemical properties and active ingredient profile of bio-stimulant Aquasap powder from seaweed *K. alvarezii*

Physicochemical Parameters	Units	Results
Total organic matter	g/100g	8.31
Electrical conductivity (1% solution)	mS	4.63
pH (1% solution)	-	7.90
Moisture content	ml/100g	1.90
Total ash	g/100g	89.64
Bulk density	g/cc	0.76
<b>Macro nutrients</b>		
Nitrogen (N)	g/100g	0.54
Phosphorous (P)	g/100g	0.03
Potassium (K)	g/100g	34.50
<b>Secondary nutrients</b>		
Calcium (Ca)	g/100g	0.57
Magnesium (Mg)	g/100g	1.15
Sulphur (S)	g/100g	1.54
<b>Micro nutrients</b>		
Zinc (Zn)	mg/kg	47.82
Manganese (Mn)	mg/kg	34.29
Sodium (Na)	g/100g	7.34
Boron (B)	mg/kg	119.20
Iron (Fe)	mg/kg	0.05
Silica (SiO <sub>2</sub> )	g/100g	3.48
Chloride (Cl)	g/100g	39.35
Copper (Cu)	mg/kg	15.84
Cobalt (Co)	mg/kg	48.13
<b>PGR</b>		
Auxin	%	0.06
Cytokinin	%	0.07
Gibberellin	%	0.18
<b>Amino Acids</b>		
Threonine	g/100g	0.07
Valine	g/100g	0.01
Methionine	g/100g	0.02
Isoleucine	g/100g	0.12
Leucine	g/100g	0.14
Tyrosine	g/100g	0.11
Phenylalanine	g/100g	0.03
Histidine	g/100g	0.02
Lysine	g/100g	0.05

Arginine	g/100g	0.04
Tryptophan	g/100g	0.12
Proline	g/100g	0.02
Alanine	g/100g	0.12
Aspartic Acid	g/100g	0.07
Glycine	g/100g	0.06
Glutamine	g/100g	0.33
Serine	g/100g	0.05

(Source: Karthikeyan and Shanmugam, 2016a)

**Table 2:** Vegetative growth and yield of onion applied with bio-stimulant Aquasap powder

Characters	Control	Treated plants	Yield increased in treated plants
Plant weight (g)	99.80±25.51	144.80±22.34	45.09
Plant length (cm)	44.57±6.37	58.84±5.04	32.03
No. of root (nos.)	41.07±16.39	52.47±12.48	27.76
Root length (cm)	6.19±1.21	9.09±2.20	46.85
No. of leaves (nos.)	10.46±2.55	12.40±3.03	18.55
Leaves length (cm)	33.18±5.12	48.21±4.95	45.29
Leaves weight (g)	22.47±5.75	43.27±7.71	92.57
Leaves thickness (mm)	1.54±0.39	2.93±0.93	90.26
Bulb weight (g)	94.80±17.11	124.80±19.94	31.65
Diameter of bulb (mm)	50.81±7.20	61.75±3.49	21.53
Diameter of neck (mm)	17.02±3.37	21.31±3.19	25.21
Yield per acre (kg)	5266	6933	31.65

**Table 3:** Effect of bio-stimulant Aquasap powder on quality of onion

Characters	Control		Treated plants	
	Day of harvest	After 30 days	Day of harvest	After 30 days
Total soluble solid (%)	8.52±1.73	15.37±1.85	9.74±0.14	15.79±1.64
Moisture content (%)	89.68±0.83	82.74±0.99	91.26±2.08	84.89±2.06
Ash content (%)	0.61±0.06	0.83±0.02	0.74±0.04	0.91±0.07
Organic content (%)	7.91±1.71	14.61±1.76	9.58±0.80	14.91±1.59
Pyruvic acid ( $\mu\text{mol g}^{-1}$ )	0.92±0.19	1.11±0.78	0.98±0.11	1.32±0.33
Physiological loss in 30d (%)	-	4.46±0.83	-	3.33±0.25
Rotting percentage in 30d (%)	-	0.98±0.24	-	0.85±0.25

method of Gordon E Anthon and Diane M Barrett (2003). Onions were sliced in half longitudinally in which the outer skin and ends were cut down. One half of the onion extract was to homogenate for 1 min in a Waring blender at a ratio of 1 ml of water per gram of onion. The homogenate was allowed to stand for 10 min at room temperature; it was then filtered through two layers of cheesecloth. This filtrate was transferred to a 1.5 ml centrifuge tube and centrifuged at 10,000 rpm for 10 min.

25 µl of the clarified onion filtrate was added to 1.0 ml of water in a test tube with a Drummond positive displacement pipette. Additionally 1.0 ml of 0.25 g<sup>-1</sup> DNPH in 1 M HCl was added and shaken well then the samples were incubated at 37°C in a water bath for 10 min and 1.0 ml of 1.5 M NaOH was added. Absorbance at 515 nm was taken and calculated the pyruvic acid level in onion samples against standard.

#### 2.4. Statistical analysis

Statistical analysis such as analysis of variance (ANOVA, SYSTAT version 7), correlation and regression were applied to analyze the data.

### 3. RESULT AND DISCUSSION

#### 3.1. Yield and Pyruvic acid content

Weight of control plant ranged from 80 to 153 g (ave. 99.8 g) and treated plant was 119 to 195 g (ave. 144.8 g) with 45.09% more yield than control plant and significant positive correlated with leaf weight ( $r= 0.585$ ;  $p=0.02$ ). The plant length ranged from 47.8 to 66.1 cm (ave 58.84 cm) and 32.2 to 57.5 cm (ave. 44.57 cm) in treated and control plants respectively i.e. 32.02% increased in treated plants and it correlated significantly with leaf length ( $r = 0.954$ ;  $p = 0.001$ ) (Table 2). The similar result was observed by Vijayanand et al. (2014) that extract of *Sargassum wightii* increased the total plant height (33%) and total fresh weight (155%) in cluster bean. Karthikeyan and Shanmugam (2015) observed that height of peanut and sunflower increased to 31.24% and 10.57% respectively when they were treated with bio-stimulant Aquasap derived from *K. alvarezii* through foliar application and similar response was also observed in some banana varieties (Karthikeyan and Shanmugam 2014).

Plant length increased to 48.21% in maize by the application of extract of seaweed *Laurencia obtusa* and *Jania rubens* (Safinaz and Ragaa, 2013). Similar kind of results were recorded

in green chillies, turnips and pineapple when treated with extracts of *Hypnea musciformis*, *Spathoglossum asperum*, *Stoechospermum marginatum* and *Sargassum* by Dhargalkar and Untawale (1983), in *Cajanus cajan* (Mohan et al., 1994), ragi, maize and kambu (Rajkumar Immanuel and Subramanian, 1999).

Average root lengths in control and treated plants were 6.19 cm (4.7 to 9.2 cm) and 9.09 cm (7.2 to 16.6 cm) respectively ( $r= 0.530$ ;  $p= 0.05$ ) i.e., root length increased to 46.85% when compared with control one. The no. of root ranged from 11 to 65 nos. (ave. 41.07 nos.) and 31 to 80 nos. (ave. 52.47 nos.) in control and treated plants respectively with 27.76% increased in treated plants (Table 2). The similar result was observed in *Lycopersicon esculentum* treated with extract of *Sargassum johnstonii* by Kumari et al. (2011). Gigantic growth and more increase in the fresh and dry mass of root than the control was examined in Marigold seedlings when treated with seaweed concentrate (Aldworth and van Staden, 1987).

The no. of leaves per plant ranged from 8 to 17 nos. (ave. 10.46 nos.) and 8 to 18 nos. (ave. 12.4 nos.) in control and treated plants respectively with 18.55% increase in treated one. Average leaves length in treated plants and untreated were 48.21 cm (39.3 to 56.5 cm) and 33.18 cm (28 to 42.8 cm) respectively with 45.29% increased in treated plants. Leaves weight ranged from 14 to 37 g (ave. 22.47 g) and 30 to 56 g (ave.43.27 g) in untreated and treated plants respectively with 92.57% increased in treated plants. Average leaves thickness in control and treated plants were 1.54 mm (1.1 to 2.6 mm) and 2.93 mm (1.2 to 4.9 mm) respectively with 90.26% increase in treated plants (Table 2). Similar kind of results were recorded in *Amaranthus caudatus* when treated with extract of seaweed *Padina tetrastromatica* it enhanced number of leaves (56.8%) and biomass (Kumareswari and Maria, 2015), similar as same observations were made by Sunarpi et al. (2010) on rice plant applied with extract of seaweed *Sargassum*. *Helianthus annuus* treated with 2.5% extract of *S. wightii* made remarkable influence on shoot length, root length, leaf length, leaf breadth, fresh weight, dry weight and number of leaves (Akila and Jeyadoss, 2010).

The weight of bulb ranged from 62 to 134 g with

an average of 94.80 g control plants and it was 100 to 170 g (ave. 124.8 g) in treated plants i.e. 31.65% increase in plants treated bio-stimulant Aquasap powder. Average of bulb diameter in control and treated plants were 50.81 mm (36.9 to 68.1 mm) and 61.75 mm (56.9 to 66.9 mm) respectively with 21.53% increase in treated plants. Diameter of bulb neck ranged from 11.2 to 22.6 mm (ave. 17.02 mm) and 15.2 to 27.8 mm (ave. 21.31 mm) in control and treated plants respectively with 25.21% increased in treated (Table 2) (Fig. 1). Yield of bulb weight 94.80 kg and 124.80 kg were observed in control and treated plants respectively and yield was 31.65% more over control (Table 2). Eswaran et al. (2009) had observed that 22% more bulb yield was in onion treated *Eucheuma* seaweed powder. Dogra and Mandradia (2012) also recorded weight gain and quality improvement in onion treated with extract of seaweed *Ascophyllum nodosum*. Yield increase of 57% was recorded in soybean when treated with 15% aqueous extract of fresh *K. alvarezii* through foliar application (Rathore et al., 2009). In the present investigation, total solid, moisture content, ash content, organic matter, physiological loss and rotting percentage for 30 days were estimated (Table 3). Out of the entire quality parameters total soluble solid, ash content, organic matter were obtained higher value when compare with the value at the day of harvest. Moisture content had decreased after 30 days when compared to the 1<sup>st</sup> day of the harvesting onion. After 30 days physiological loss and rotting percentage were found low in treated plants as compared to control plants, but those values are not statistically significant (Table 3).

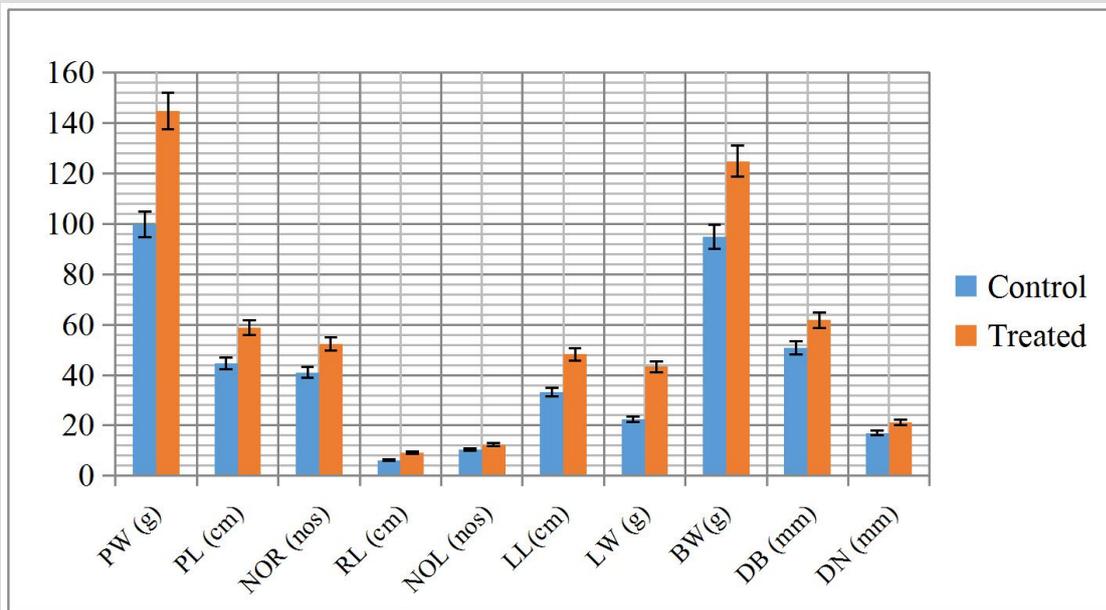
The pyruvate concentration in onion bulbs ranges from 1.6  $\mu\text{mol g}^{-1}$  FW to 13  $\mu\text{mol g}^{-1}$  FW in mild onion (Randle, 1992). In the present investigation, it was 0.92  $\mu\text{mol g}^{-1}$  and 0.98  $\mu\text{mol g}^{-1}$  in control and treated plants respectively and after 30 days, it found increased 1.11  $\mu\text{mol g}^{-1}$  and 1.32  $\mu\text{mol g}^{-1}$  in control and treated plants which could be due to loss in moisture (Table 3). Similar kind of observation was made by De et al. (2013) in onion treated with biozyme, a seaweed based bio-stimulant.

Onion pungency depends on the presence of organosulphur compounds which can be manufactured from the enzymatic reaction

in which S-alk(en)yl-L-cysteine S-oxide decomposition occurs (Lancaster et al., 2000). This decomposition will be led to the formation of flavour, such as methyl cysteine sulphoxide, 1-propenyl cysteine sulphoxide and propyl cysteine sulphoxide, which are well insulated in onion cells. When onion bulbs are cut down immediately the cells are disrupted and these organosulphur compounds are released. When the cell membranes are broken those compounds then suddenly come into contact with the alliinase enzyme (Lancaster and Collin, 1981). The compounds which are synthesised by alliinase are pyruvate, ammonia and sulphenic acid, this is what lead to the pungency of onions. Nowadays the demand of the consumer has been turned towards onion cultivars with low pungency that was called as mild cultivars. The factors such as sulphur-based fertilization, the environment, and the genotype of the cultivars lead to the accumulation of organosulphur compounds in onions (Yoo et al., 2006). However, sulphur-based fertilization and onion pungency are not always positively correlated, and negative results have been reported (Randle and Bussard, 1993). The pyruvic acid content was determined by environmental growing conditions, storage and clones (Yoo et al., 2006). However the genetic background is one of the most important factors because different cultivars have different capabilities to uptake of sulphur content and consumption in the biosynthesis pathway which results to the flavour (Randle et al., 1994).

### 3.2. Role of potash in onion plants

*K. alvarezii* extract contains micro, macro nutrients and growth hormones (Karthikeyan and Shanmugam 2016 a,b). Aquasap powder, an extract of *K. alvarezii* is rich of potash (Table 1). Fig 2 shows the bulb of control plants were comparatively softer than one treated with Aquasap powder and average layer per bulb were  $8.9 \pm 0.88$  and  $10.4 \pm 0.85$  nos. in control and treated respectively. The potash is involved in protein synthesis and play role for the opening and closing of stomata (Wikipedia). Potash plays an important role in not only photosynthesis but also to enhance the translocation of assimilates, to maintain water balance which will be manage to grow in drought condition, to produce and transporting the carbohydrates and sugar and protein and also promoting the enzyme



**Fig. 1:** Growth and yield of onion applied with Bio-stimulant Aquasap powder

PW- Plant weight (g), PL- Plant length (cm), NOR- No. of root (nos.), RL- Root length (cm), NOL- No. of leaves (nos.), LL- Leaves length (cm), LW- Leaves weight (g), BW- Bulb weight (g), DB- Diameter of bulb (mm), DN - Diameter of neck (mm).



**Fig 2:** Texture of onion bulb of control and treated plants

activities (Marschner, 2012). It also plays an essential role for better shelf-life and quality of the crop produce. Even though onion requires high amount of potash content than that of nitrogen during bulb formation but over uptake of potassium lead to low yield. Deficiency of potash will lead to slow growth, leaves are erect, bulbs will be so soft and thin and it will be clearly identified by the appearance of brown tips and slight yellowish in older leaves and lead. The appropriate application of potassium to onion will lead to high growth and quality (Deshpande et al., 2013). The maximum plant height (51.6 cm), number of leaves per plant (9.89), bulb diameter (5.93), average bulb weight (64.89 g), leaf width (1.33 cm) and yield (22.91 t ha<sup>-1</sup>) were observed with the application of 120 kg K<sub>2</sub>O ha<sup>-1</sup> as described by (Shah Saud et al., 2013). Islam et al., (2008) had reported that significant improvement in different growth parameters and yield of onion in response to different application methods and levels of potassium.

#### 4. CONCLUSION

Seaweed is considered to be well balanced, harmless and natural source with a high degree of bio availability of trace elements. It can be concluded from present investigation study that onion had responded well to 0.2% Aquasap powder (brand name of AquAgri for bio-stimulant of seaweed *K. alvarezii*) with an average yield of about 31.65% of onion with much improved quality. Present findings encourage the application of such seaweed based bio-stimulant higher crop yield with improved quality in bellary onion.

#### ACKNOWLEDGEMENT

The authors are very grateful to Mr. Abhiram Seth, MD, Mr. Arun Patnaik, CEO and Mr. Tanmaye Seth of AquAgri Processing Private Limited, India for their constant encouragement and guidance to carry out present investigation. The authors also wish to thank staff of AquAgri Processing Private Limited for their support during the study.

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