

## Enhancing bottle gourd growth and fruit characteristics through the stimulant effects of humic acid, seaweed extract, and salicylic acid

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### Abstract

Bottle gourd is a highly nutritious crop with great important as health staples. On the other hand, organic acids have been widely used to improve the quality and quantity of horticultural crops as an alternative to mineral fertilizers, which have shown negative effects on the environment and health. This study was conducted during the two successive seasons 2021 and 2022 at the Agricultural Experimental Farm, Faculty of Agriculture and Natural Resources, Aswan University, Aswan, Egypt. The experiment investigated three plant growth stimulators; humic acid (HA), seaweed extract (SW) and salicylic acid (SA). The study aimed to evaluate the effect of those stimulators on bottle gourd growth and fruit characteristics. The stimulators showed significant effects on growth characteristics such as plant height and number of leaves, but the effects were not clear for number of flowers and fruits. In the same line, the stimulators showed significant effects on fruit dry weight and its seeds dry weight, but not for fruit length and diameter. HA at high concentration (6 g/L) showed high values of both growth and fruit characteristics. Meanwhile SA showed high impact on growth characters at 0.6 g/L, and high dry weight of fruits and its seeds at low 0.2 g/L concentration. From the results of this study, it can be recommended to treat bottle gourd with HA at 6 g/L to ensure significant improvement in both plant growth and fruit characteristics.

**Keywords:** *Lagenaria siceraria*, biostimulators, algae extract, growth characters, fruit characters

### Introduction

Bottle gourd (*Lagenaria siceraria* L.) plant is driven from Cucurbitaceae family, and it is one among highly liked vegetables raised in South East Asia [1]. It is originated from Africa and nowadays is grown mainly throughout the tropics and subtropics of the world. China, Ukraine, Argentina and Turkey account for approximately 45% of the world production of bottle gourd. Bottle gourds are cultivated in many places, old and new soils, in the world and are of great importance as they are rich in edible and health staples [2].

Bottle gourd is a monoecious annual herbaceous crop exhibiting considerable variations in floral morphology and vegetative structure [3]. This species is a white-colored flowered gourd with highly cross-pollinated, and has enormous amount of variation for many economic traits. The fruits show immense variation in

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shape and size ranging from flattened and discoidal form, long club shaped and can be conical, round or club shape [4]. Bottle gourd is highly nutritious to humans, since it possess all major and micronutrients responsible for maintaining good health. One hundred gram of fruit may contain protein (2000 mg), carbohydrate (2500 mg) and traces of minerals like calcium (20 mg), iron (0.7 mg) and phosphorus (10 mg) [5].

Organic acids have been widely used to improve the quality and quantity of horticultural crops, and the presence of hormonal compounds have significant effects on improving the physical, chemical, and biological properties of soil and increasing the yield and quality of agricultural crops [6]. Thus, the use of natural fertilizers including humic acid, salicylic acid, and algae extract, without adverse environmental damage, can improve plant yield.

Humic acid (HA) is a water-soluble organic acid that is naturally present in soil organic matter. It is the main fraction of humate substances and the most active components of soil and compost organic matter. HA is a natural polymer that contains of H<sup>+</sup> positions related to acidic carboxyl of benzoic and phenolic agents [7]. It is known to contain carboxyl, alcoholic hydroxyl, ketone, quinoide and phenolic hydroxyl [8]. It can be recognized that HA has several beneficial effects on soil structure and soil microbial populations, in addition to plant growth stimulation, cell permeability, and nutrient uptake [9, 10]. HA can enhance nutrient availability and improve chemical, biological, and physical soil properties [11]. It contains abundant nutrients improves soil fertility and increase the availability of nutrients to plants and thus it influences plant growth and yield [12]. HA is a fertile source of many major nutrients including 6-8% hydrogen, 46-42% oxygen, 44-58% carbon and 4-5% nitrogen, as well as many other nutrients which encourage plant growth [13]. When HA applied to field, it is readily converted into available humic material which directly or indirectly influences plant development by enabling the plant to absorb more nutrients from the soil and making these nutrients in an easily available form [14].

Seaweed extracts have now been one of the important strategies to ensure sustainable agriculture, especially in arid and semi-arid regions where soils are poor in organic nutrients [15, 16], as a mean 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 [17]. They have gained prominence as plant biostimulants in horticulture [18]. The beneficial effect of seaweed extract application is as a result of many components that may work synergistically at different concentrations [19]. They are available in soluble powder form or as liquid extracts and may be side-dressed near the root of the plant or applied as foliar sprays. Seaweeds, particularly the brown algae (Phaeophyceae), are a source of polysaccharides, sterols, N-containing compounds (e.g., betaines), macro- and micro-nutrients, and also growth-promoting substances such as auxins, cytokinins, gibberellins, and brassinosteroids. Seaweed extracts have recently been included as ‘plant biostimulants’, with wide applications in agricultural systems [20-22].

Salicylic acid (SA) is known to regulate biochemical, molecular, and physiological processes, such as nutrient uptake, transport, photosynthetic capacity, and membrane permeability under stress conditions [23]. However, the increase in fruit quality can be attributed to the cell division, growth enhancement and signal transduction ability [24]. SA is known to enhance endogenous proline contents, also acting as osmolarity, thus protecting complex II electron transport [25], and strengthening cell membrane three-dimensional structure [26] along with antioxidants enzymes, such as SOD, POD, and Rubisco.

The main objective of this study was to investigate the effect of humic acid, seaweeds and salicylic acid on growth and fruit characteristics of pumpkin plants grown under Aswan conditions.

## Materials and methods

### Plant material and experimental site

This field study was conducted during the two successive seasons of 2021 and 2022 at the Agricultural Experimental Farm, Faculty of Agriculture and Natural Resources, Aswan University, Aswan, Egypt.

Seeds of bottle gourd (*Lagenaria siceraria*) were obtained from the National Research Center, Cairo, Egypt. Compost was applied at a rate of 20 m<sup>3</sup> per feddan two weeks before sowing seeds. Plot size was 3×4 m and had two rows. At the mid-April for both seasons, the seeds were soaked in water for 24 hours and were sown directly into the soil at 60 cm in a row and about 1 m between rows. After 52 days, the plants were thinned to one plant per hill (10 plants per plot) i.e. 7000 plants per feddan.

In mid-April, it was the second season, and after 7 days, all the plants were out, and after about 15 days of planting, the plants were thinned, and the distance between one plant and the other and between the rows was the same as the first season.

### Experimental design

The experiment was laid out in randomized complete block in factorial design incorporated three replicates. Humic acid, salicylic acid, and seaweed extract were used. The total treatments were 10 treatments as followed; Control (water only), three levels of HA (3 g, 4.5 g, or 6 g per liter), three levels of SA (0.2 g, 0.4 g, or 0.6 g per liter) and three levels of seaweed extract (1 ml, 2 ml, or 3 ml per liter).

The treatments were applied where humic acid was applied to the soil and salicylic acid and seaweed extract were sprayed on the whole plant three times starting from 50 days after planting and repeated every 15 days for both seasons. A drip irrigation system was used for irrigation and all other agricultural practices were implemented as recommended during the two seasons. Each season lasted for six months.

### Data recorded

The plants were harvested at the end of October, and the following plant parameters were measured one day before treatment and every 15 days after each treatment. Measurements were taken and recorded four times on 30<sup>th</sup> May, 14<sup>th</sup> June, 29<sup>th</sup> June and 14<sup>th</sup> July in both seasons. The parameters were:

1. Plant height (cm): The length of the main stem from the soil surface to the top of the plant was measured using a multimeter
2. The number of leaves on the main stem.
3. Number of flowers: The number of leaves was measured from base to apex
4. The number of fruits
5. The length of the fruits
6. The diameter of the fruits
7. The dry weight of the fruits
8. Dry weight of seeds

### Statistical analysis

Data obtained were subjected to statistical analysis using “F” Test [27] and the means were compared using a least significant difference (L.S.D.) test according to [28]. Statistical analysis was performed using Microsoft Office 365 Excel program.

### Results and discussion

Analysis of variance showed that there were significant differences among treatments in term of plant height and number of leaves per plant in both seasons (Table 1). Increasing of HA, SW and SA concentrations led to significant increases in plant height in both seasons compared to control. The tallest plant was shown in plant treated with 6.0 g/L HA in the first season, and in plants treated with 0.6 g/L SA in the second season (Table 1). In the same line, number of leaves per plant increased with increasing HA, SW and SA compared with control. The highest number of leaves was shown in plants treated with 6.0 g/L HA and 0.6 g/L SA in the first season, and in plants treated with 0.6 g/L SA in the second season (Table 1). Number of flowers did not show significant differences among treatments in both seasons. While the differences in number of fruits was significant in the first season and not significant in the second season (Table 1).

**Table (1):** Effect of humic acid (HA), seaweed extract (SW) and salicylic acid (SA) on bottle gourd plant height as well as number of leaves, flowers and fruits during 2020-2021 and 2021-2022 seasons.

	Plant height (cm)		Number of leaves		Number of flowers		Number of fruits	
	1 <sup>st</sup> season	2 <sup>nd</sup> season						
Control	217.5	121.7	63.8	33.8	7.00	4.25	2.50	1.00
3.0 g/L HA	214.2	127.5	64.5	44.0	9.50	4.25	1.00	1.25
4.5 g/L HA	265.8	138.3	82.0	55.8	8.00	4.75	1.50	1.25
6.0 g/L HA	<b>294.1</b>	149.2	<b>114.3</b>	58.5	8.00	4.00	1.25	1.50
1 ml/L SW	240.8	125.8	94.5	50.0	6.00	4.50	1.25	1.25
2 ml/L SW	243.3	141.7	95.8	52.8	6.50	4.00	1.50	1.50
3 ml/L SW	247.5	174.2	97.5	73.8	6.75	4.25	1.75	2.00
0.2 g/L SA	223.3	144.2	80.5	86.8	7.25	5.00	2.25	1.75
0.4 g/L SA	254.2	162.5	97.5	99.3	6.75	4.50	1.50	1.75
0.6 g/L SA	268.4	<b>193.4</b>	<b>107.5</b>	<b>101.5</b>	6.50	4.75	1.00	1.75
LSD 0.05	<b>9.6</b>	<b>8.6</b>	<b>5.2</b>	<b>5.7</b>	<b>ns</b>	<b>ns</b>	<b>0.32</b>	<b>ns</b>

*ns* represents not significant differences

Analysis of variance showed that there were significant differences among treatments in term of fruit length in the first season but not in the second season, while fruit diameter was not differ significantly in the first season and differed significantly in the second season (Table 2). The tallest fruit was shown under control and decreased with increasing of HA, SW and SA concentrations compared to control. Meanwhile fruit dry weight and seeds dry weight per fruit differed significantly among treatments compared to control in both seasons (Table 2). The highest fruit dry weight was shown in plants treated with 6.0 g/L HA and 0.2 g/L SA in both seasons. While the highest seeds weight was shown in plants treated with 0.6 g/L SA in both seasons (Table 2).

**Table (2):** Effect of humic acid (HA), seaweed extract (SW) and salicylic acid (SA) on bottle gourd fruit length, diameter and dry weight height as seeds dry weight/fruit during 2020-2021 and 2021-2022 seasons.

	Fruit length (mm)		Fruit diameter (mm)		Fruit dry weight (g)		Seeds dry weight (g)	
	1 <sup>st</sup> season	2 <sup>nd</sup> season						
Control	<b>37.5</b>	<b>41.7</b>	80.8	81.1	62.5	63.3	18.6	12.8
3.0 g/L HA	32.5	33.3	77.4	82.4	65.0	66.7	25.0	28.7
4.5 g/L HA	27.0	32.3	83.1	86.5	72.5	76.7	31.0	42.7
6.0 g/L HA	24.2	28.3	90.6	97.0	<b>85.0</b>	<b>103.3</b>	46.8	57.9
1 ml/L SW	33.1	35.0	88.6	103.0	72.5	73.3	20.8	20.7
2 ml/L SW	27.3	30.0	78.8	89.2	60.8	76.7	37.8	40.0
3 ml/L SW	27.5	30.0	85.4	84.9	53.8	76.7	46.3	56.0
0.2 g/L SA	31.7	36.7	85.7	93.4	<b>81.7</b>	<b>105.0</b>	20.5	19.7
0.4 g/L SA	25.4	33.3	78.8	83.5	63.8	96.7	26.3	29.3
0.6 g/L SA	26.9	28.3	77.3	79.8	47.1	50.0	70.3	91.3
LSD 0.05	<b>2.7</b>	<b>ns</b>	<b>ns</b>	<b>4.6</b>	<b>5.0</b>	<b>5.4</b>	<b>3.3</b>	<b>4.3</b>

*ns* represents not significant differences

Natural and synthetic growth stimulators play an important role in promoting growth and development of crops. For decades, such plant growth stimulators have been used extensively in crop production, whether by soil

application or foliar spraying. Plant stimulators have positive effects not only on plant growth and nutrition, but also on abiotic and biotic stress tolerance [29]. For example, salicylic acid as a synthetic stimulator regulates several functions in plant including plant resistance to environmental stresses such as drought and salinity [30] and chilling [31].

HA is a promising natural resource to be used as an alternative for fertilizers to increase crop production [32]. It contains many elements which improve the soil fertility and increase the availability of nutrients and consequently increase plant growth and yield by acting on mechanisms involved, cell respiration, photosynthesis, water and nutrient uptake, and enzyme activities [33, 34]. Many researchers reported that HA increase fruit yield, number of fruit per plant, fruit length of squash [35, 36] and of cucumber [37-39].

Humic substances have been shown to stimulate shoot and root growth and nutrient uptake of vegetable crops [12, 40, 41]. When increasing HA, it works on the absorption of minerals and nutrients (nitrogen, potassium, and phosphorus) necessary for plant growth. In addition, it serves as a source of plant growth regulators, carbohydrates, amino acids, and vitamins [42]. It is also found that HA increased the level of endogenous substances; cytokinins, gibberellins, and auxins. In addition, it can also be used as a carrier of nutrients and growth regulators. Thus, increased uptake of nutrients enhanced the vegetative growth of the plant, stimulated plant growth hormones and increased cell division [9].

On the other hand, seaweed is classified as humic substance that had positive effects in improving crop growth and quality due to its containing specific amounts of active plant growth substances such as auxins, cytokinins and their derivatives [43]. In addition, seaweed extracts include polysaccharides laminarin, alginates and carrageenans and their breakdown products as well as micro and macro nutrients [20, 21].

Numerous studies indicated that the application of different seaweed extracts improved yields of vegetables, fruit, and field crops under normal and a variety of stress conditions through enhancing root vigor, plant growth, and development, plant hardiness, shoot/root biomass, and leaf numbers [44-46]. Furthermore, the application of different commercial seaweed extract types on vegetables and medicinal shrubs has been reported to improve tolerance to a wide range of abiotic stresses, including drought [47-49], and nutrient deficiency [50]. Seaweed extracts have been shown to be effective in improving stress resistance in many other crops, such as spinach [48], maize [51, 52], sweet orange, cucumber [22], and zucchini squash [53].

## Conclusion

From the results of this study, it was concluded that the best treatment to improve plant growth characteristics is HA at 6 g/L and SA at 0.6 g/L. On the other hand, the best treatment to improve fruit characteristics was HA at 6 g/L and SA at 0.2 g/L. Therefore, this study suggests treating bottle gourd with HA at 6 g/L to ensure significant improvement in both plant growth and fruit characteristics.

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