International Journal of Agriculture and Plant Science
www.agriculturejournal.in
Online ISSN: 2664-7664
Received: 04-07-2026, Accepted: 04-08-2026, Published: 19-08-2026
Volume 8, Issue 3, 2026, Page No. 264-272

Stimulative effects of Yeast and Aloe extracts on growth of Fittonia

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DOI: https://doi.org/10.66856/ijaps.2026.8.3.8216

Abstract
This study evaluated the efficacy of yeast extract (YE) and Aloe vera gel (AVG) as eco-friendly bio stimulants for enhancing vegetative growth, leaf chemical composition, and biomass accumulation of Fittonia albivenis (Verschaffeltii Group), a commercially significant indoor foliage plant in two seasons (2024 and 2025). A Randomized Complete Block Design (RCBD) was employed with seven treatments across three replications: a distilled water control (C) and three concentrations each of YE (1%, 2%, 3%) and AVG (1%, 2%, 5%), applied as foliar sprays every two weeks over a 90-day period under controlled greenhouse conditions. Morphological parameters assessed included: leaf area (cm²), number of leaves per stem, stem diameter (mm), leaf fresh weight (g), and leaf dry weight (g). Chemical analysis comprised total chlorophyll content (mg g^{-1} FW) and leaf concentrations of nitrogen (N%), phosphorus (P%), and potassium (K%). Also, Catalase and Peroxidase enzymes were analyzed. Total phenolic and flavonoids were taken a place in chemical analysis. Results demonstrated that both bio stimulants conferred statistically significant improvements over the control across all measured parameters ($p \leq 0.05$). Yeast Extract at 2% (Y_2) yielded the highest values for leaf area (265.4 cm²), stem diameter (5.4 mm), leaf fresh weight (16.82 g), leaf dry weight (3.62 g), total chlorophyll (2.15 mg g^{-1} FW), and N, P, K concentrations. Aloe vera Gel at 2% (A_2) recorded competitive improvements across all parameters and is recommended as a sustainable alternative for commercial ornamental production. Both Y_2 and A_2 are proposed as optimal bio stimulant treatments for the commercial cultivation of Fittonia albivenis.

Keywords: *Fittonia albivenis*, yeast extract, *Aloe vera* gel, bio stimulants, foliar application, leaf area, stem diameter, chlorophyll, mineral nutrition, catalase, peroxidase, ornamental horticulture

Introduction

Fittonia albivenis (Lindl. ex-Veitch) Brummitt (Family: Acanthaceae), commonly known as the nerve plant, is among the most aesthetically distinguished ornamental foliage species in contemporary interior horticulture (Deng, M., *et al*. 2026) [12]. Its ornamental value derives principally from its distinctive reticulate venation patterns manifested in white, pink, or red vein networks superimposed upon a rich green laminar surface (Kang and Lopez 2026) [27]. The plant thrives under warm, humid, low-light conditions, rendering it particularly suitable for indoor cultivation (Koster and Sibley 2017) [29]. However, F. albivenis is notably sensitive to suboptimal moisture regimes, inadequate mineral nutrition, and elevated abiotic stress, which precipitate premature leaf senescence, diminished pigment integrity, and reduced biomass accumulation (Hao *et al*., 2025 [21] van Doorn & Woltering, 2008[47]).

In the context of sustainable intensification of ornamental crop production, the integration of plant bio stimulants has emerged as a scientifically validated strategy. Bio stimulants operationally defined as substances or microorganisms that, when applied to seeds, plants, or the rhizosphere, stimulate natural processes to enhance growth, nutrient use efficiency, and tolerance to abiotic stress constitute an increasingly prominent sector within global horticulture (Du Jardin, 2015[14]; Yakhin *et al*., 2017[51]).

Among the most extensively studied bio stimulants, yeast extract (YE) derived through the controlled autolysis of Saccharomyces cerevisiae represents a biochemically complex matrix containing essential amino acids, cytokinins, B-complex vitamins (B1, B2, B6, B12), nucleotides, and minerals (Mg, Fe, Zn, K, N, P). These constituents synergistically activate protein and nucleic acid biosynthesis, enhance chloroplast differentiation, stimulate apical meristematic cell division, and directly augment leaf mineral content (Mitchison *et al*., 2019) [36]. Currently, Aloe vera gel (AVG) obtained from the inner parenchyma of Aloe barbadense Miller constitutes a rich reservoir of acemannan polysaccharides, gibberellin like phytohormones, salicylic acid, phenolic antioxidants, and essential microelements that collectively enhance photosynthetic capacity and nutrient uptake (Hasan *et al*., 2026[22]; Liu, C., *et al*. 2019[32])

Despite the expanding body of literature on bio stimulant efficacy in field crops and vegetables, investigations focused specifically on sensitive indoor foliage plants particularly Fittonia albivenis with emphasis on leaf morphometrics, biomass quantification, and leaf mineral composition remain sparse. This study was therefore designed to: (1) quantify the dose dependent effects of YE and AVG on leaf area, leaf number per stem, stem diameter, and leaf fresh and dry weight; (2) assess their influence on chlorophyll content and leaf N, P, K composition; (3) identify optimal concentrations; and (4) provide a comparative assessment of their agronomic utility.

In addition, F. albivenis is an economically vital tropical foliage ornamentally valued globally for its vibrant foliar vein coloration and compact growth habit. Native to the

understory canopy of South American rainforests, F. albivenis has evolved specialized light harvesting super complexes to efficiently utilize far-red and low light spectra. However, this shade adapted metabolic machinery renders the species particularly susceptible to oxidative stress when exposed to environmental fluctuations common in commercial greenhouse production, such as sudden shifts in light intensity, temperature extremes, or altered relative humidity (Hasanuzzaman *et al*., 2020) [23].

Under such abiotic perturbations, the overproduction of reactive oxygen species (ROS) most notably hydrogen peroxide (H_2O_2), superoxide radicals (O_2^-), and hydroxyl radicals (OH) exceed basal cellular quenching capacities (Sachdev, *et al*., (2021) [41]. Unchecked

ROS accumulation induces lipid peroxidation of photosynthetic membranes, degrades leaf pigments, and initiates foliar chlorosis or necrosis, severely compromising the aesthetic quality and commercial value of the crop (Sakaki, *et al*., 1983) [43].

To mitigate oxidative damage and preserve metabolic homeostasis, plants rely on an integrated defense network comprising both enzymatic antioxidants and secondary metabolites (Ahmad, *et al*., 2010) [2]. Among the enzymatic defense mechanisms, catalase (CAT) and peroxidase (POD) serve as critical primary lines of defense by regulating intracellular H O concentrations (Anjum, *et al*., 2016) [5] Catalase directly dismutates bulk cellular H_2O_2 into water (H_2O) and molecular oxygen (O) without requiring chemical reductants:

$$2H\,O \xrightarrow{CAT} 2H\,O + O$$

Functioning primarily in peroxisomes during photorespiratory fluxes (Anjum *et al*., 2016) [5]. Concurrently, peroxidases detoxify excess H O across cytosolic, vacuolar, and cell wall compartments using diverse electron donors, while simultaneously participating in cell wall reinforcement, lignification, and wound healing responses (Cleverson 2024) [9].

Furthermore, the coordinated activity of enzymatic antioxidants like CAT and POD is essential for maintaining redox balance in shade adapted ornamentals during acute acclimation periods in protected cultivation environments.

Materials and Methods

Experimental Conditions

The experiment was conducted at a controlled environment greenhouse facility during the 2024 - 2025 growing seasons at Sabahia Research Station, Alexandria, Egypt. Environmental parameters were maintained within physiological optima for F. albivenis: temperature 24 ± 2°C, relative humidity 75–80%, and photosynthetic photon flux density (PPFD) of 120 µmol m^{-2} s^{-1} under filtered indirect illumination with a 14/10-hour light/dark photoperiod. All conditions were monitored at 4-hour intervals using calibrated digital dataloggers (accuracy: ±0.2°C; ±2% RH).

Plant Material and Propagation

Uniform, 12-week-old rooted cuttings of Fittonia albivenis were obtained from a certified local commercial nursery. All cuttings exhibited homogeneous dimensions (8–10 cm shoot length, 4–6 leaf pairs) and were free of visible pathogens or mechanical damage. Plants were transplanted into 15 cm diameter plastic pots containing a sterilized substrate of peat moss and horticultural perlite at a 2:1 volumetric ratio (v/v). Each experimental unit comprised one plant per pot. Plants were irrigated uniformly with half strength Hoagland nutrient solution twice weekly throughout the experimental period.

Preparation of Bio stimulant Solutions

Yeast extract (YE) was prepared from locally available commercial food grade dried yeast (Saccharomyces cerevisiae, purity >95%) dissolved in distilled water at three concentrations: 10 g L^{-1} (1%, Y_1), 20 g L^{-1} (2%, Y_2), and 30 g L^{-1} (3%, Y_3). Aloe vera gel (AVG) was sourced from freshly harvested, mature leaves (3–5 years old) of Aloe barbadense Miller, from which the inner parenchymatous gel was mechanically extracted, homogenized using a sterile blender, and immediately diluted to three concentrations: 10 mL L^{-1} (1%, A_1), 20 mL L^{-1} (2%, A_2), and 50 mL L^{-1} (5%, A_3). All solutions were freshly prepared within one hour of each application to prevent oxidative degradation of bioactive constituents.

Experimental Design and Treatments

Seven treatments were arranged in a Randomized Complete Block Design (RCBD) with three replicates (blocks), yielding 21 experimental units. Treatments were as follows:

1. **Control (C):** Foliar application of distilled water
2. **Y_1:** Yeast Extract at 1% (10 g L^{-1})
3. **Y_2:** Yeast Extract at 2% (20 g L^{-1}) primary YE treatment
4. **Y_3:** Yeast Extract at 3% (30 g L^{-1})
5. **A_1:** Aloe vera Gel at 1% (10 mL L^{-1})
6. **A_2:** Aloe vera Gel at 2% (20 mL L^{-1}) primary AVG treatment
7. **A_3:** Aloe vera Gel at 5% (50 mL L^{-1})

All bio stimulant solutions were applied as foliar sprays at run off point using hand operated pressurized sprayers. Applications were performed every two weeks (every 15 ± 1 day) in the early morning hours (7–9 Am) to maximize stomatal aperture and minimize phytotoxic risks. A total of six foliar applications were made over the 90-day experimental period (January–April, 2024 and January–April, 2025).

Collected Data and Analytical Methods

Morphological Parameters

At the conclusion of the experimental period (Day 90), the following morphological measurements were recorded: (1) Leaf area (cm^2) determined using a portable leaf area meter (LI-COR LI-3100C, USA); (2) Number of leaves per stem counted manually on a representative main stem per plant; (3) Stem diameter (mm) measured at the midinternode position of the main stem using a digital vernier caliper (accuracy ± 0.01 mm); (4) Leaf fresh weight (g) determined immediately after harvest by analytical balance; (5) Leaf dry weight (g) determined gravimetrically after oven drying leaf material at 70°C for 72 hours to constant mass.

Chemical Analysis

Total chlorophyll content (mg g^{-1} FW) was determined spectrophotometrically following the (Porra *et al*.,1989) [38] protocol, using 80% acetone as the extractant (absorbance measured at 645 nm and 663 nm). For mineral analysis, freshly dried and ground leaf samples were subjected to dry ashing at 500°C followed by dissolution in dilute HCl (1 N). Nitrogen (N%) was determined by the micro Kjeldahl method (Bradstreet,2002) [8]. Phosphorus Content (P%): Phosphorus was determined spectrophotometrically using the ascorbic acid method at a wavelength of 882 nm according to the procedure described by Murphy and Riley (1962) [37]. Potassium (K%) was determined by flame photometry (Cooper,1963) [10]. All chemical analyses were performed in triplicate on fresh dried, ground leaf samples collected from each experimental unit. On the other side, Catalase (CAT) Activity, Peroxidase (POD) Activity, Total Phenolic Content (TPC) and Total Flavonoid Content (TFC) were analyzed as follows:

Enzymatic Antioxidant Assays

To evaluate the plant's enzymatic defense response under the applied treatments, crude enzyme extracts were prepared from fresh leaf tissue at 4 °C.

- **Catalase (CAT) Activity:** CAT activity was determined spectrophotometrically by monitoring the decomposition of hydrogen peroxide (H_2O_2) as a decrease in absorbance at 240 nm. Activity was expressed as Umg^{-1} protein (Hadwan *et al*., 2024) [18].
- **Peroxidase (POD) Activity:** POD activity was measured by recording the guaiacol oxidation in the presence of H O through the increase in absorbance at 470 nm. Activity was expressed as Umg^{-1} protein (Liu *et al*., 2014) [31].

Quantification of Non-Enzymatic Antioxidant Metabolites

Secondary metabolites were extracted from dried tissue samples using 80% (v/v) aqueous methanol:

Total Phenolic Content (TPC): TPC was quantified using the Folin Ciocalteu colorimetric reagent at an absorbance of 765 nm. Results were expressed as gallic acid equivalents (mg GAEV g^{-1} DW) derived from a standard calibration curve (Kupina *et al*., 2017) [30].

Total Flavonoid Content (TFC): TFC was determined via the aluminum chloride ($AlCl_3$) assay at an absorbance of 510 nm. Concentrations were expressed as quercetin equivalents (mg QE g^{-1}) (Shrestha 2022) [44].

Statistical Analysis

All data were subjected to analysis of variance (ANOVA) using SPSS Statistics version 29.0 (IBM Corp., USA). Mean separations were performed using Duncan's Multiple Range Test (DMRT) at probability level $p \leq 0.05$. Results are expressed as means of three replicates. Different letters within each column indicate statistically significant differences between treatment means ($p \leq 0.05$).

Results

Effects on Morphological Characteristics

The analysis of variance (ANOVA) indicated that foliar applications of both Yeast Extract (YE) and Aloe vera Gel (AVG) exerted statistically significant improvements ($P \leq 0.05$) across all evaluated morphological parameters of Fittonia albivenis relative to the untreated control in both consecutive seasons 2024 and 2025 (Table 1). This finding in agreement with established bio stimulant research demonstrates that exogenous applications of organic extracts enhance vegetative architecture and vegetative mass in tropical indoor foliage plants (Hamouda *et al*.,2012[20]; Cristofano *et al*., 2021[11]).

Table 1: Effect of Yeast Extract and Aloe vera Gel Foliar Applications on Morphological Traits of Fittonia albivenis across Two Consecutive Seasons

Treatment	Leaf Area (cm^2)		No. Leaves / Stem		Stem Diameter (mm)		Fresh Weight (g)		Dry Weight (g)	
	2024	2025	2024	2025	2024	2025	2024	2025	2024	2025
Control	185.3 c	189.1 e	8.4 c	8.76d	3.2c	3.4c	8.25g	8.41e	2.25c	2.21d
Yeast 1%	230.4 b	219.6 c	10.2b	11.36bc	4.4ab	4.2b	11.43e	12.2c	2.85b	2.91b
Yeast 2%	265.4 a	270.2 a	13.8a	14.1a	5.4a	5.5a	16.82a	17.15a	3.62a	3.71a
Yeast 3%	240.2 b	254.3 b	13.0a	12.4ab	4.8ab	4.96a	14.31c	14.62b	3.36a	3.18b
Aloe 1%	202.4 b	206.8 d	9.6b	9.8cd	4.0b	3.96b	10.14f	10.36d	2.58b	2.64c
Aloe 2%	248.4ab	260.3ab	12.5a	12.8a	5.1ab	5.2a	15.64b	16.95a	3.35a	3.60a
Aloe 5%	225.6 b	229.8 c	10.9b	11.2bc	4.4ab	4.5b	12.53d	12.8c	2.92b	2.98b
LSD ($P \leq 0.05$)	19.04	11.68	1.14	1.55	0.75	0.466	0.639	1.02	0.288	0.25

Note: Values represent means of three replicates. Different letters within each column (for a given season) indicate statistically significant differences between treatment means (Duncan's Multiple Range Test, $P \leq 0.05$). LSD = Least Significant Difference.

The vegetative growth parameters of the studied plants, including leaf area, number of leaves per stem, stem diameter, fresh weight, and dry weight, were significantly affected ($p \leq 0.05$) by foliar applications of active dry yeast extract and Aloe vera extract during the two consecutive growing seasons (2024 and 2025) compared to the untreated control (Table 1).

Leaf Area and Number of Leaves per Stem

Application of both natural extracts led to a marked expansion in leaf surface area and an increase in the number of leaves per stem. The highest significant values for leaf area were recorded under the treatment of Yeast 2%, achieving 265.4cm^2 and 270.2cm^2 in the first and second seasons, respectively, closely followed by Aloe 2% (248.4$cm^2$and 260.3cm^2). Similarly, the number of leaves per stem reached its maximum value with Yeast 2% (13.8 and 14.1) and Aloe 2% (12.5 and 12.8) showing no statistical difference between them in both seasons. Conversely, increasing the concentration beyond 2% (Yeast 3% or Aloe 5%) led to a slight decline in both parameters, although they remained significantly superior to the control,

which registered the lowest values (185.3cm^{2}and 189.1cm^2 for leaf area, and 8.4 and 8.76 for leaf number in 2024 and 2025, respectively). Stem Diameter

Stem diameter showed a similar trend to leaf parameters. Foliar spraying with Yeast 2% and Aloe 2% yielded the thickest stems, recording values of 5.4 mm and 5.5 mm for Yeast 2%, and 5.1 mm and 5.2 mm for Aloe 2% in 2024 and 2025, respectively. The control plants exhibited the minimum stem diameter 3.2 mm and 3.4 mm).

Fresh and Dry Biomass Accumulation

On the other side, biomass production responded positively to the tested biometric stimuli. The maximum fresh weight was recorded in plants treated with Yeast 2% (16.82 g in 2024 and 17.15 g in 2025), followed closely by Aloe 2% (15.64 g in 2024 and 16.95 g in 2025, with no significant difference between them in the second season). Furthermore, the dry weight exhibited a parallel response, peaking at 3.62 g and 3.71 g under Yeast 2%, and 3.35g ,3.60g under Aloe 2% across the two experimental seasons. On the other hand, untreated plants produced the lowest biomass accumulation (fresh weight 8.25 g and 8.41 g; dry weight 2.25 g and 2.21 g).

Table 2: Effect of Yeast Extract and Aloe vera Gel Foliar Applications on Leaf Chemical Composition and Macro-Nutrients of Fittonia albivenis across Two Consecutive Seasons

Treatment	Total Chlorophyll (mg g FW)		N Content (%)		P Content (%)		K Content (%)	
	2024	2025	2024	2025	2024	2025	2024	2025
Control	1.25d	1.28e	1.85d	1.89c	0.23f	0.25c	1.42e	1.45e
Yeast 1%	1.77bc	1.69c	2.34bc	2.61b	0.31d	0.36b	1.85c	1.91c
Yeast 2%	2.15a	2.20a	3.12a	3.17a	0.45a	0.47a	2.48a	2.52a
Yeast 3%	1.89ab	1.92b	2.92a	2.80ab	0.39b	0.38ab	2.15b	2.19b
Aloe 1%	1.51c	1.54d	2.15c	2.19c	0.27e	0.28c	1.68d	1.71d
Aloe 2%	2.06a	2.11a	3.04a	3.06a	0.43a	0.46a	2.41a	2.47a
Aloe 5%	1.73bc	1.76c	2.48b	2.52b	0.34c	0.35b	1.95c	1.99c
LSD ($P \leq 0.05$)	0.22	0.101	0.201	0.312	0.028	0.058	0.108	0.113

Note: Values are means of three replicates. FW = fresh weight. Different letters within each column (for a given season) indicate statistically significant differences (Duncan's Multiple Range Test, $P \leq 0.05$). N = nitrogen; P = phosphorus; K = potassium. LSD = Least Significant Difference. Means within the same column followed by different letters are significantly different according to LSD test at $p \leq 0.05$

Chemical Composition

Foliar applications of active dry yeast extract and Aloe vera gel extract significantly ($p \leq 0.05$) enhanced the chemical constituents of the leaves, including total chlorophyll content, as well as nitrogen (N), phosphorus (P), and potassium (K) percentages, compared to the untreated control during both the 2024 and 2025 growing seasons (Table 2).

Total Chlorophyll Content

Photosynthetic pigment accumulation showed a marked increase in response to both bio stimulants. The maximum total chlorophyll content was recorded in plants treated with Yeast 2% (2.15 mg/g FW in 2024 and 2.20 mg/g FW in 2025) and Aloe 2% (2.06 mg/g FW in 2024 and 2.11 mg/g FW in 2025), with no statistically significant differences observed between them in either season. Increasing the application dose beyond 2% (Yeast 3% or Aloe 5%) led to a moderate decline in chlorophyll values (1.89- 1.92 mg/g for Yeast 3% and 1.73- 1.76 mg/g for Aloe 5%), though they remained significantly superior to the control, which consistently exhibited the lowest values (1.25 mg/g in 2024 and 1.28 mg/g in 2025).

Leaf Nitrogen Content (N %)

Leaf nitrogen status followed a similar upward trend. Application of Yeast 2% yielded the highest leaf N content (3.12% and 3.17%), closely accompanied by Aloe 2% (3.04% and 3.06% in both seasons respectively) and Yeast 3% (2.92% and 2.80%), with non-significant variations among these top-performing treatments. In contrast, untreated control plants registered the lowest nitrogen accumulation (1.85% in 2024 and 1.89% in 2025).

Leaf Phosphorus Content (P%)

Phosphorus percentage responded prominently to bio stimulant spraying. Plants receiving Yeast 2% (0.45% and 0.47%) and Aloe 2% (0.43% and 0.46%) attained the highest significant P percentages across both seasons, forming a statistically superior group. Intermediate values were obtained from Yeast 3% (0.39% and 0.38%) and Aloe 5% (0.34 - 0.35%), whereas the control treatment showed the lowest P concentrations (0.23% and 25% in both seasons respectively).

Leaf Potassium Content (K %)

Potassium accumulation in leaf tissue was substantially elevated by the bio stimulant treatments. The peak values of leaf K percentage were achieved by Yeast 2% (2.48% in 2024 and 2.52% in 2025) and Aloe 2% (2.41% in 2024 and 2.47% in 2025), without significant differences between them. The untreated plants exhibited the lowest potassium values (1.42% and 1.45%).

Enzymatic Antioxidant Defense Response

Table 3: Effect of Yeast Extract (YE) and Aloe vera Gel (AVG) foliar applications on the enzymatic antioxidant activities and total secondary metabolite content of Fittonia albivenis for Consecutive Seasons

Treatment	CAT Activity (Ug^{-1} FW)		POD Activity (Ug^{-1} FW)		Total Phenolics (mg GAE g^{-1} DW)		Total Flavonoids (mg QE g^{-1} DW)	
	2024	2025	2024	2025	2024	2025	2024	2025
Control	14.20 c	14.10 e	1.85 d	1.87 d	16.50 e	16.80 e	9.20 e	9.50 f
Yeast 1%	25.26 b	22.06 c	3.37 bc	3.42 b	28.43 c	26.23 c	15.4 c	15.86 d

Yeast 2%	33.2 a	32.9 a	4.35 a	4.37a	42.10 a	43.13 a	23.66 a	24.10 a
Yeast 3%	26.4 b	26.13 b	3.6 bc	3.6 b	34.54 b	34.50 b	19.1 b	19.43 b
Aloe 1%	18.3 c	18.3 d	3.2 c	2.45 c	21.30 d	21.35 d	13.0 d	13.4 e
Aloe 2%	32.5 a	32.03 a	4.07 ab	4.27 a	40.16 a	41.93 a	22.5 a	23.23 a
Aloe 5%	23.9 b	23.8 c	2.45 d	3.2 b	31.96 bc	32.50 b	17.63 b	17.83 c
LSD ($P \leq 0.05$)	4.65	1.73	0.604	0.584	3.87	2.47	1.66	1.56

Means within the same column followed by different letters are significantly different at $p \leq 0.05$according to Duncan's Multiple Range Test (DMRT).

Abbreviations: FW= Fresh Weight; DW= Dry Weight; GAE= Gallic Acid Equivalent; QE= Quercetin Equivalent; CAT= Catalase; POD= Peroxidase.

Antioxidant Enzymatic Activity and Secondary Metabolites

The activity of primary enzymatic antioxidants (CAT and POD) exhibited significant variation ($P \leq 0.05$) across treatments compared to the Control plants. Foliar application of active dry yeast extract and Aloe vera gel extract significantly ($p \leq 0.05$) enhanced the antioxidant enzyme activities specifically catalase (CAT) and peroxidase (POD) as well as the accumulation of secondary metabolites (total phenolics and total flavonoids) in plant tissues during both the growing seasons compared to the untreated control (Table 3).

Antioxidant Enzyme Activities (CAT and POD)

The enzymatic defense system exhibited a pronounced positive response to bio stimulant sprays:

Catalase (CAT) Activity: The highest CAT activity was recorded in plants treated with Yeast 2% (33.2 and 32.9 both seasons respectively) and Aloe 2% (32.50 in 2024 and 32.03 in 2025), forming a statistically superior group with no significant difference between them. Increasing the concentration to Yeast 3% (26.40 and 26.13) or Aloe 5% (23.90 and 23.80) caused a noticeable decrease in CAT activity, though they remained significantly higher than the control, which consistently exhibited the lowest enzyme activity (14.20 and 14.10).

Peroxidase (POD) Activity: POD activity followed a similar trend. Peak values were attained under Yeast 2% (4.35 in 2024 and 4.37 in 2025) and Aloe 2% (4.07 in 2024 and 4.27 in 2025), with no statistically significant differences between them. The untreated plants showed the minimum POD activity (1.85 and 1.87 in both seasons respectively). These results are shown in Fig. (1)

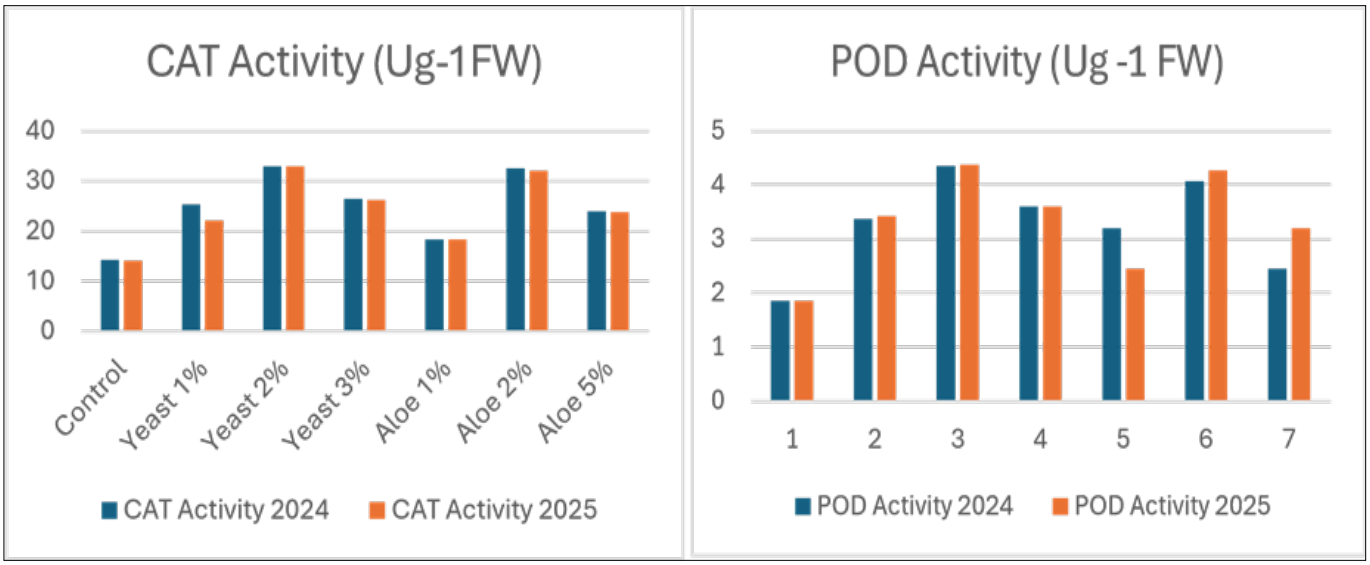

Fig 1: Effect of Yeast Extract (YE) and Aloe vera Gel (AVG) foliar applications on the enzymatic antioxidant activities Fittonia albivenis for Consecutive Seasons.

Non-Enzymatic Antioxidants (Total Phenolics and Total Flavonoids)

Secondary metabolites were significantly boosted across all treated plants relative to the control:

Total Phenolics: Foliar application of Yeast 2% and Aloe 2% markedly increased the accumulation of phenolic compounds, reaching maximum values of 42.10 mg/g and 40.16 mg/g in 2024, and 43.13 mg/g and 41.93 mg/g in 2025, respectively. Intermediate levels were observed with Yeast 3% (34.54 and 34.50 mg/g) and Aloe 5% (31.96-32.50 mg/g), whereas untreated control plants recorded the lowest phenolic content (16.50 mg/g and 16.80 mg/g).

Total Flavonoids: Flavonoid content peaked under Yeast 2% (23.66 mg/g in 2024 and 24.10 mg/g in 2025) and Aloe 2% (22.50 mg/g in 2024 and 23.23 mg/g in 2025), showing statistical parity between the two treatments. The control treatment consistently recorded the lowest flavonoid levels (9.20 mg/g and 9.50 mg/g). These results showed in Fig. (2)

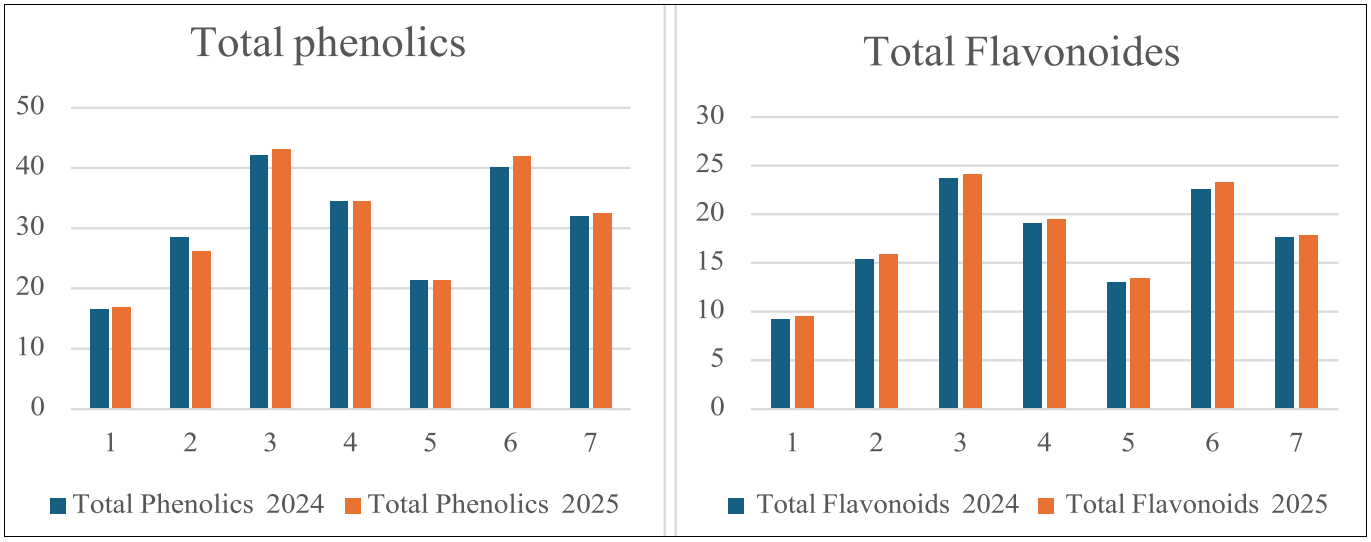

Fig 2: Effect of Yeast Extract (YE) and Aloe vera Gel (AVG) foliar applications on the total secondary metabolite content of Fittonia albivenis for Consecutive Seasons.

Discussion

The significant improvement observed in vegetative growth traits (leaf area, leaf count, stem diameter, and overall biomass) following the application of yeast and Aloe vera extracts highlights their role as effective natural bio stimulants.

Mechanism of Yeast Extract Action

The superiority of Yeast 2% in enhancing vegetative parameters can be attributed to its rich content of essential nutrients, vitamins (particularly Vitamin B-complex), phytohormones specifically cytokinins and auxins and free amino acids (Mir *et al*.,2025) [35] Cytokinins play a vital role in stimulating cell division (cytokinesis) and cell enlargement, leading to an increased number of leaves and expanded leaf area (Wu *et al*., 2021) [48]. Furthermore, yeast extract serves as a rich source of biological nutrients that boost chlorophyll synthesis and enhance photosynthetic efficiency, which directly translates into greater accumulation of carbohydrates and biomass (fresh and dry weight) (Youssef *et al*., 2022) [52]

Mechanism of Aloe vera Extract Action

Similarly, the pronounced positive response to Aloe 2% spraying is linked to the biostimulator components inherent to Aloe vera gel. Aloe vera extract is loaded with natural bioactive compounds, including acemannan (polysaccharides), essential micro- and macro-elements (K,Ca,Mg,Zn), and antioxidant vitamins (A, C, and E) (Bahgat *et al*., 2023) [7]. It also contains endogenous phytohormones, such as gibberellins and auxins, which stimulate cell elongation and apical meristematic activity (Manokari *et al*.,2023) [34]. Furthermore, Alo vera contains amino acids (Abdullah 2024) [1]. The application of Aloe vera extract enhances stomatal conductance and water retention efficiency within leaf tissues, thereby encouraging healthy vascular tissue development (reflected in increased stem diameter) and dry matter production (Ali *et al*.,2026) [4].

Interestingly, increasing the treatment concentrations beyond 2% (to 3% for Yeast and 5% for Aloe) resulted in a relative reduction in growth performance compared to the optimal 2% concentration. This trend is a well-documented physiological response: excessive levels of exogenously applied bio-extracts or phytohormone like substances can induce an osmotic imbalance or hormonal feedback inhibition, suppressing cell elongation and photosynthetic rate (Hamouda *et al*., 2012) [20]. Thus, 2% concentration for both Yeast and Aloe vera represents the physiological optimum threshold for maximizing plant vegetative growth without triggering adverse feedback effects.

Mechanism of Aloe vera Extract Action on Chemical Composition

The physiological response to Aloe vera 2% spraying which matched Yeast 2% in efficacy reflects the bio-functional constituents present in Aloe vera gel:

1. **Antioxidant Protection of Photosynthetic Pigments:** Aloe vera gel contains high concentrations of antioxidant molecules (ascorbic acid, α-tocopherol, aloin, and flavonoids) and enzymes such as superoxide dismutase (Hamman, 2008;[19] Surjushe *et al*., 2008[45]). These antioxidants scavenge reactive oxygen species (ROS) within the chloroplasts, preserving light harvesting complex proteins and thylakoid integrity (Hęś *et al*., 2019) [24].

2. **Nutrient Uptake and Membrane Permeability:** Acemannan (polysaccharide) and saponins present in Aloe vera act as natural surfactant agents that reduce the surface tension of spray droplets, enhancing foliar absorption and membrane permeability to macro-elements (N,P,K) (Wulandini *et al*.,2019) [50]. Furthermore, Aloe vera gel contains intrinsic mineral elements (K,Ca,Mg) that directly contribute to leaf nutrient reserves (Kamble, *et al*. 2022) [26].

The 2% Optimum, like vegetative traits, increasing the dosage beyond 2% (Yeast 3% or Aloe 5%) led to a moderate decline in total chlorophyll and N, P, K percentages compared to the optimal 2% concentration. This trend can be explained by:

Hormonal Feedback & Ethylene Induction: High doses of yeast supply excessive auxin levels, triggering ACC synthase activity and subsequent ethylene synthesis, which induces premature chlorophyll breakdown and stomatal closure (Ljung, 2013[33]; Taiz *et al*., 2015[46]).

Leaf Surface Osmotic Load: High concentrations of Aloe vera gel (5%) form a dense viscous layer on the leaf surface, creating a temporary osmotic barrier or physically restricting stomatal gas exchange (CO uptake) and transpiration, thereby downregulating active nutrient translocation (Farooq *et al*., 2009) [16]

On the other hand, the elevation of antioxidant enzymatic activities (CAT and POD) and non-enzymatic phytochemical compounds (phenolics and flavonoids) in response to Yeast and Aloe vera extracts highlights the bio-elicitor function of these natural bio stimulants in activating secondary metabolism and cellular redox homeostasis.

Mechanism of Action Yeast Extract

The superior performance of Yeast Extract at 2% in upregulating antioxidant machinery and phenolic metabolism can be attributed to several biochemical pathways:

Elicitation via Cell Wall Components: Yeast extract contains active cell wall polysaccharides, such as β-glucans and chitin/chitosan micro fragments, which serve as natural pathogen associated molecular patterns (PAMPs) or elicitors (Kogan, G., & Kocher, A. 2007) [28]. When recognized by plasma membrane bound pattern recognition receptors (PRRs), these molecules activate signaling cascades involving calcium flux (Ca) and mitogen activated protein kinases (MAPK), ultimately upregulating genes encoding phenylpropanoid enzymes like phenylalanine ammonia-lyase (PAL) (Dixon & Paiva, 1995[13]; Apel & Hirt, 2004[6]). PAL is the key enzyme responsible for synthesizing phenolic acids and flavonoids.

Regulation of Reactive Oxygen Species (ROS) and Enzymatic Protection: Yeast extract provides vital micronutrients (Fe,Zn,Mn,Cu) which serve as essential cofactors for antioxidant enzymes. Specifically, catalase (CAT) requires heme iron (Fe), while peroxidases (POD) rely on iron/manganese centers (Grant *et al*., 1998[17]; Ribeiro *et al*., 2015[40]). By maintaining ROS scavenging efficiency (dismutation H O into H O and O), CAT and POD protect chloroplast and cellular membranes from lipid peroxidation, as reflected in overall cell viability (Qi *et al*., 2024) [39].

Mechanism of Action: Aloe vera Extract as an Elicitor

The pronounced impact of Aloe vera extract at 2% matching Yeast 2% across all evaluated parameters stems from its rich phytochemical matrix:

Direct Contribution and Enzymatic Priming: Aloe vera gel contains endogenous phenolic compounds (aloin, aloe-emodin) and flavonoids, as well as organic acids, vitamins (A, C, E), and trace elements (Hęś *et al*.,2019[24]; Elaıbı *et al*., 2023[15]). Foliar absorption of these bioactive molecules directly enriches the tissue pool of total phenolics and flavonoids.

Elicitation by Acemannan: Acemannan (polysaccharide) in Aloe vera triggers eicosanoid and phenylpropanoid biosynthetic pathways in plant tissues, leading to increased activity of antioxidant enzymes (CAT and POD) (Kahramanoğlu *et al*., 2019[25]; Wu *et al*., 2024[49]). Increased phenolic and flavonoid accumulation provide defense against photo oxidative stress and strengthens cell wall architecture through lignification mediated by POD (Aida *et al*., 2022) [3].

Increasing extract concentrations to 3% (Yeast) or 5% (Aloe) triggered a relative decline in enzyme activities and secondary metabolite accumulation compared to the 2% concentration (Ziemlewska, *et al*., 2025) [53]

Saturating Signal and Metabolic Feedback: High levels of elicitor molecules can over-stimulate defense pathways, triggering negative feedback loops or metabolic fatigue (Hasan *et al*., 2026) [22].

Enzyme Inactivation/Inhibition: excessively high ROS turnover or substrate accumulation can lead to oxidative self-inactivation of CAT and POD enzymes.

Osmotic Load: A dense coating from high-concentration Aloe vera (5%) can restrict stomatal diffusion, causing transient cellular stress that diverts metabolic energy away from secondary metabolite synthesis toward basic survival pathways (Saini *et al*.,
2025) [42].

Comparative Analysis

Evaluating performance across both experimental cycles demonstrates that 2% Yeast Extract (Y) provides the most consistent response for maximizing leaf expansion, stem thickness, biomass assembly, and mineral accumulation in Fittonia albivenis. 2% Aloe vera Gel (A) offers a closely comparable, eco-friendly organic alternative, exhibiting strong efficacy across all evaluated traits.

These complementary modes of action cytokinin driven meristematic activation and amino acid assisted nitrogen assimilation (YE) versus polysaccharide mediated cell turgidity, antioxidant protection, and micro-element uptake (AVG) highlight the potential for dual bio stimulant or rotational foliar application programs in commercial greenhouse production.

Recommendation

Based on these findings, a foliar application of 2% Yeast Extract or 2% Aloe vera gel is strongly recommended as an optimal, eco-friendly bio stimulant strategy to maximize vegetative growth, nutrient uptake, and biochemical defenses in Fittonia albivenis. Commercial growers should avoid exceeding this 2% threshold to prevent osmotic imbalance, ethylene-induced chlorophyll breakdown, and metabolic feedback inhibition. Furthermore, future research should explore rotational or combined application regimes of both extracts to optimize commercial greenhouse production.

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