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# Evaluation of bio-stimulant Phylgreen on performance of summer greengram

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## Abstract
The present study was undertaken to evaluate the efficacy of the bio-stimulant Phylgreen 200 (P-6084), developed by Tradecorp Rovensa India Pvt. Ltd., on the performance of summer greengram (*Vigna radiata* L.) under irrigated conditions. Conducted at the Agricultural Research Station, Amadalavalasa, during the summer of 2022, the experiment employed a randomized block design (RBD) with nine treatments and three replications. Treatments included different foliar doses (2.0, 2.5, 3.0, and 6.0 ml/l) of Phylgreen 200, applied at the first flowering and pod setting stages under both normal and drought-stressed conditions, with appropriate controls. The study recorded key biometric observations including plant height, number of flowers and pods per plant, 100-seed weight, and grain yield. Results revealed that all Phylgreen 200 treatments significantly outperformed controls with respect to growth and yield parameters. The application of Phylgreen 200 at 2.5 ml/l ($T_2$) and Drought Stress Phylgreen 200 at 2.5 ml/l ($T_6$) produced the highest grain yields (648.1 and 624.2 kg/ha, respectively) and return on investment (179.7% and 128.7%, respectively). Importantly, no phytotoxic symptoms were observed at any dosage, including the highest tested concentration of 6.0 ml/l. The results suggest that Phylgreen 200 enhances crop performance and economic returns in greengram cultivation, especially under drought stress. Further research is warranted to understand the physiological mechanisms underpinning the observed benefits.

**Keywords:** Greengram, *Vigna radiata*, Phylgreen 200, bio-stimulant, drought stress, yield

## Introduction
Green gram (*Vigna radiata* L.) is one of the most important conventional pulse crops cultivated in India. It has been reported that Green gram has been cultivated in India since ancient times. It is believed that Green gram is a native of India and Central Asia and grown in these regions since prehistoric times. It is widely cultivated throughout the Asia, including India, Pakistan, Bangladesh, Sri Lanka, Thailand, Laos, Cambodia, Vietnam, Indonesia, Malaysia, south China, and Formosa. The important states producing this pulse are Rajasthan, Madhya Pradesh, Karnataka, Maharashtra, Odisha, Bihar, Tamil Nadu and Andhra Pradesh.

Green gram is a protein rich staple food. It contains about 24 percent protein, which is almost three times that of cereals. It supplies protein requirement of vegetarian population of the country. It is consumed in the form of split pulse as well as whole pulse, which is an essential supplement of cereal based diet. It is particularly rich in Leucine, Phenylalanine, Lysine, Valine, Isoleucine, *etc*. In addition to being an important source of human food and animal feed, Green gram also plays an important role in sustaining soil fertility by improving soil physical properties and fixing atmospheric nitrogen. It is a drought resistant crop and suitable for dryland farming and predominantly used as an intercrop with other crops.

Green gram has an advantage over the other pulse crops owing to its high nutritive value and easy digestibility. The nutritive value of the seed is estimated to be 24.20% protein, 1.30% fat and 60.4% carbohydrates per 100 g of the seed. The seed also contains significant elements such as calcium (Ca) 118 mg and phosphorus (P) 340 mg per 100 g of seed(Imran *et al.*, 2015) [7]. Moreover, green gram possesses a significant position in our daily diet as well as an important component in our agricultural production system. In India, the low productivity of this crop is due to the unavailability of healthy seeds, high cost of seeds, pesticides, fertilizers and scarcity of labors at the time of peak agricultural operations as well as wide gap in adoption of recommended production technology.

## Materials and Methods

The experiment was conducted at Agricultural Research Station (ARS), ANGRAU, Amadalavalasa, Srikakulam District (A.P.) during summer season, 2021-22 under irrigated situation on Sandy loam with clay base soils. Land preparation was done by ploughing twice with tractor drawn cultivator followed by levelling. Fine tilth obtained and applied recommended dose of fertilizers as basal. Recommended dose of fertilizers was applied as basal in last ploughing before sowing. The greengram variety IPM-2-14 was selected and treated with imidacloprid 600 FS @ 5 ml kg-1 seed and kept for overnight for good adherence. Treated seed collected @ 20 kg ha-1and the seed sown at spacing of 30 cm x 10 cm manually to a depth of 3-5 cm followed by sprinkler irrigation was given good germination. Pre-emergence application of pendimethalin was done within 24 hours of sowing. Thinning was done at 10-15 days after sowing (DAS) to maintain good plant stand. Manual hand weeding and hoeing was done at 15 and 30 DAS and maintained weed free situation up to harvesting stage. The crop was grown during summer season and received 227.2 mm rainfall during May and June, 2022 months. Supplementary irrigations (3) were provided with micro-sprinkler system during crop growth period. Bio-stimulant Phylgreen 200 was applied as foliar spray as per the protocol given by manufacturer. The experiment comprised of five doses of treatments viz., $T_1$-2.0 ml/L, $T_2$-2.5 ml/L, $T_3$-3.0 ml/L and $T_4$-Control at first flower appearance and pod setting and Drought stress Phylgreen 200 as $T_5$-2.0 ml/L, $T_6$-2.5 ml/L, $T_7$-3.0 ml/L and $T_8$-Control at after irrigation at first flower appearance and 7 days after first application and Phylgreen 200 as $T_9$-6.0 ml/l (Phytotoxicity treatment) at first flower appearance and pod setting. The trial was conducted in RBD design with three replications. Crop was harvested manually after attaining physiological maturity. After harvesting the crop, the crop dried for three days and transported to threshing floor for threshing and winnowing.

During the crop growth period (26thMarch, 2022 to 24$^{th}$ June, 2022), a total of 227.2 mm rainfall received during crop growth period and supplementary irrigations (3) were given during crop growth periodusing micro-sprinkler system. The initial growth of the crop was very good with minor incidence of sucking pest complex, Spodoptera at later stage of the crop (flowering and pod setting) and leaf spot at pod development stage. Plant protection measures were taken up time to time to maintain healthy crop. More rainfall received during the months of May and June, 2022.

### General information on trial

**1. Product: Phylgreen 200 (P-6084)**
**2. Crop:** GREENGRAM
**3. Season:** *Summer*
**4. Year:** 2022
**5. Location:** Agricultural Research Station (ANGRAU) Amadalavalasa, Srikakulam-532 186, A P.
**6. Variety:** IPM 2-14
**7. Soil type:** Sandy loam with clay base

### Details of the Experiment

1. Design: RBD
2. Number of treatments: 9
3. Replications: 3
4. Plot size: 3.0 m x 4.0 m,
5. Spacing: 30 cm x10 cm
6. Date of sowing: 28-3-2022
7. Stage of the crop at the time of spray (s): Details given below

**Table A:** Details of the products along with method and time of application

| S. No | Product name | Application method | Stage of the crop | | | |
|---|---|---|---|---|---|---|
| | | | I spray | Spray fluid (l/ha) | II spray | Spray fluid (l/ha) |
| 1 | Phylgreen 200<br>$T_1$-2.0 ml/L<br>$T_2$-2.5 ml/L<br>$T_3$-3.0 ml/L<br>$T_4$-Control | Foliar | With first flower appearance | 375 | Pod setting | 500 |
| 2 | Drought stress Phylgreen 200<br>$T_5$-2.0 ml/L<br>$T_6$-2.5 ml/L<br>$T_7$-3.0 ml/L<br>$T_8$-Control | Foliar | After irrigation at first flower appearance | 375 | 7 days after first application | 500 |
| 3 | Phylgreen 200<br>$T_9$-6.0 ml/l<br>(Phytotoxicity treatment) | Foliar | With first flower appearance | 375 | Pod setting | 500 |

8. Application method: Foliar spray
9. Type of spraying equipment & nozzle used: Knapsack sprayer with hollow cone nozzle
10. Quantity of spray volume used: As per the details given above
11. Frequency and/or interval of application: As per the details given above
12. Pests/diseases involved: Sucking pests, *Spodoptera*spp. & leaf spot

### Climate and weather conditions

During the crop growth period (26$^{th}$March, 2022 to 24$^{th}$June, 2022), a total of227.2 mm rainfall received during crop growth period and supplementary irrigations (3) were given during crop growth periodusing micro-sprinkler system to both Phylgreen 200 and Drought Stress Phylgreen 200 treatments. The initial growth of the crop was very good with minor incidence of sucking pest complex, Spodoptera at later stage of the crop (flowering and pod setting) and leaf spot at pod development stage. Plant protection measures were taken up time to time to maintain healthy crop. More rainfall received during the months of May and June, 2022.

**Table B:** Meteorological Data at ARS, Amadalavalasa during summer, 2022 (26th March-24th June)

| SW | Period | Temperature (°C) | | Morning R.H. (%) | Evening R.H. (%) | Rainfall (mm) | Wind speed (kmph) | Sun-shine (hrs) |
|---|---|---|---|---|---|---|---|---|
| | | Max. | Min. | | | | | |
| 13 | 26 Mar-01 Apr | 35.86 | 27.44 | 81.4 | 56.8 | 0 | 4.86 | 6.38 |
| 14 | 02-08 Apr | 35.56 | 25.99 | 73.00 | 55.14 | 0 | 5.30 | 5.77 |
| 15 | 09-15 Apr | 36.43 | 27.17 | 73.57 | 60.71 | 0 | 6.21 | 7.94 |
| 16 | 16-22 Apr | 37.30 | 25.70 | 80.00 | 57.43 | 0 | 5.76 | 6.96 |
| 17 | 23-29 Apr | 39.73 | 27.89 | 79.29 | 57.14 | 0 | 4.77 | 8.50 |
| 18 | 30Apr-06 May | 37.64 | 26.11 | 80.86 | 57.14 | 35.6 | 5.00 | 7.64 |
| 19 | 07-13 May | 34.84 | 26.06 | 84.86 | 67.29 | 50 | 5.64 | 5.77 |
| 20 | 14-20 May | 35.16 | 28.26 | 79.43 | 74.71 | 0 | 5.27 | 8.80 |
| 21 | 21-27 May | 33.79 | 26.60 | 80.86 | 81.14 | 50 | 6.09 | 8.04 |
| 22 | 28 May-03 June | 37.89 | 27.74 | 78.57 | 73.00 | 0 | 4.03 | 5.27 |
| 23 | 04-10 June | 36.10 | 26.24 | 83.00 | 71.57 | 65.2 | 4.81 | 3.39 |
| 24 | 11-17 June | 35.66 | 25.51 | 81.86 | 81.71 | 1.8 | 3.54 | 4.29 |
| 26 | 18-24 June | 35.36 | 26.74 | 81.00 | 69.2 | 24.6 | 5.7 | 6.06 |

**Land preparation**

Land prepared by ploughing twice with tractor drawn cultivator followed by levelling. Fine tilth obtained and applied recommended dose of fertilizers as basal.

**Application of manures & fertilizers**

Recommended dose of fertilizers was applied as basal in last ploughing before sowing. Line sowing was done at spacing of 30 cm x10 cm.

**Seed treatment**

Healthy and bold seed was selected and treated with imidacloprid 600 FS @ 5 ml $kg^{-1}$ seed and kept for overnight for good adherence.

**Sowing of the seeds**

Treated seed collected @ 20 kg $ha^{-1}$and theseed sown at spacing of 30 cm x 10 cm manually to a depth of 3-5 cm followed by sprinkler irrigation was given good germination. Pre-emergence application of pendimethalin was done within 24 hours of sowing.

**Thinning and Weeding**

Thinning was done at 10-15 days after sowing (DAS) to maintain good plant stand. Manual hand weeding and hoeing was don at 15 and 30 DAS and maintained weed free situation up to harvesting stage.

**Irrigation**

The crop was grown during summer *season* and received 227.2 mm rainfall during May and June, 2022 months. Supplementary irrigations (3) were provided with micro-sprinkler system at vegetative and first flower appearence stage.

**Application /Spraying**

Bio-stimulant wasapplied as foliar spray as per the protocol given by M/s.Tradecorp India Pvt.Ltd, Pune.

**Harvesting**

Crop was harvested manually after attaining physiological maturity. After harvesting the crop, the crop dried for three days and transported to threshing floor for threshing and winnowing.

**Methods of recording biometric observations**

**a) Growth attributes**

Data on flower count at full flowering stage andplant height (cm) at pod setting stagewere recorded by tagging 10 plants randomly in each plot.

**b) Yield attributes**

Data on yield attributes viz., number of pods per plant, test weight (g)/100 grain weight and seed yield (kg $ha^{-1}$) were recorded at the time of harvest.

**c) Methods of Statistical analysis**

The statistical analysis followed as per the procedures mentioned by Panse and Sukhatme (1978).

**Results**

**I. Bio-Stimulant, Phylgreen 200 (P-6084)**

The experiment comprised of nine treatments *viz*., Phylgreen treatment ($T_1$ to $T_4$), Drought Stress Phylgreen 200 treatments ($T_5$ to $T_8$) and Phytotoxicity treatment ($T_9$) and all the treatments imposed twice at first flower appearance stage and seven days after 1st spray/pod setting stage. The trial was conducted in RBD design with three replications.

**Table 1:** Phytotoxicity symptoms-Crop injury (Days after spray)

| Treatments | 1 DAS | 3 DAS | 5 DAS | 7 DAS | 10 DAS | 1 DAS | 3 DAS | 5 DAS | 7 DAS | 10 DAS |
|---|---|---|---|---|---|---|---|---|---|---|
| $T_1$-Phylgreen 200 @ 2.0 ml/l | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| $T_2$-Phylgreen 200 @ 2.5 ml/l | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| $T_3$-Phylgreen 200 @ 3.0 ml/l | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| $T_4$-Control | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| $T_5$-Drought Stress-Phylgreen 200 @ 2.0 ml/l | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| $T_6$-Drought Stress-Phylgreen 200 @ 2.5 ml/l | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| $T_7$-Drought Stress-Phylgreen 200 @ 3.0 ml/l | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| $T_8$-Drought Stress-Control | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| $T_9$-Drought Stress-Phylgreen 200 @ 6.0 ml/l | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |

The data revealed that the no phytotoxic symptoms like epinasty, hyponasty, yellowing, necrosis, leaf injury, vein clearing, stunting etc., were noticed at 1, 3, 5, 7, and 10 days after imposition of the treatments (Table 1).

**Table 2:** Growth and yield of Greengram as influenced by bio-stimulant Phylgreen 200 (P-6084) during summer, 2022

| Treatments | Plant height at harvest (cm) | Flowers/plant | Pods/plant | 100 grain weight (g) | Grain Yield (kg/ha) |
|---|---|---|---|---|---|
| $T_1$-Phylgreen 200 @ 2.0 ml/l | 42.10 | 47.00 | 29.23 | 4.22 | 586.7 |
| $T_2$-Phylgreen 200 @ 2.5 ml/l | 42.67 | 47.27 | 30.20 | 4.32 | 648.1 |
| $T_3$-Phylgreen 200 @ 3.0 ml/l | 40.80 | 45.37 | 28.90 | 4.15 | 574.7 |
| $T_4$-Control | 38.33 | 37.87 | 24.30 | 3.93 | 461.9 |
| $T_5$-Drought Stress-Phylgreen 200 @ 2.0 ml/l | 40.00 | 44.70 | 28.20 | 4.16 | 560.6 |
| $T_6$-Drought Stress-Phylgreen 200 @ 2.5 ml/l | 42.10 | 45.87 | 28.87 | 4.26 | 624.2 |
| $T_7$-Drought Stress-Phylgreen 200 @ 3.0 ml/l | 39.53 | 43.53 | 28.37 | 4.16 | 553.9 |
| $T_8$-Drought Stress-Control | 39.20 | 36.50 | 23.87 | 4.09 | 471.9 |
| $T_9$-Drought Stress-Phylgreen 200 @ 6.0 ml/l | 41.93 | 42.77 | 26.10 | 4.13 | 559.7 |
| F test | Sig | Sig. | Sig. | Sig | Sig. |
| SEM + | 0.55 | 0.87 | 1.27 | 0.06 | 22.61 |
| CD (0.05) | 1.66 | 2.62 | 3.81 | 0.17 | 67.77 |
| CV (%) | 2.36 | 3.48 | 7.98 | 2.43 | 6.99 |

All the growth and yield attributes significantly influenced by foliar application ofPhylgreen treatment ($T_1$ to $T_4$), Drought Stress Phylgreen 200 treatments ($T_5$ to $T_8$) and Phytotoxicity treatment ($T_9$) when compared to respective treatments (water spray only) (Table 2). All the treatments were significantly superior over the control with respect to growth parameters and grain yield. Grain yield in different dosages of Phylgreen 200 treatments was more compared to respective dosages of Drought Stress Phylgreen 200 treatments. Among the treatments, significantly higher grain yield of 648.1and 624.2 kg/ha was registered with foliar spray of Phylgreen 200 @ 2.5 ml/land Drought Stress Phylgreen 200 @ 2.5 ml/l, respectively. The grain yield was dose dependent, grain yield increased with increase in dose of Phylgreen 200 up to 2.5 ml/l in both Phylgreen 200 and Drought Stress Phylgreen treatments. Beyond the level of 3.0 ml/l, the grain yield was reduced. Flowers/plant at full flowering stage, plant height at pod setting stage, pod per plant at harvestand grain weight were significantly influenced by foliar application of Phylgreen treatment ($T_1$ to $T_4$), Drought Stress Phylgreen 200 treatments ($T_5$ to $T_8$) (Table 2).

**Table 3:** Return of investment (ROI) for the bio-stimulant, Phylgreen 200 (P-6084)

| Treatments | Grain Yield (kg/ha) | Yield Increment (kg/ha) | Additional returns (Rs./ha) | Additional cost (Rs./ha) | Net Returns (Rs./ha) | R O I (%) |
|---|---|---|---|---|---|---|
| $T_1$-Phylgreen 200 @ 2.0 ml/l | 586.7 | 124.8 | 9079.2 | 4275.00 | 4804.2 | 112.4 |
| $T_2$-Phylgreen 200 @ 2.5 ml/l | 648.1 | 186.2 | 13546.1 | 4843.75 | 8702.4 | 179.7 |
| $T_3$-Phylgreen 200 @ 3.0 ml/l | 574.7 | 112.8 | 8206.2 | 5412.50 | 2793.7 | 51.6 |
| $T_4$-Control | 461.9 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |
| $T_5$-Drought Stress-Phylgreen 200 @ 2.0 ml/l | 560.6 | 88.7 | 6453.0 | 4275.00 | 2178.0 | 50.9 |
| $T_6$-Drought Stress-Phylgreen 200 @ 2.5 ml/l | 624.2 | 152.3 | 11079.8 | 4843.75 | 6236.1 | 128.7 |
| $T_7$-Drought Stress-Phylgreen 200 @ 3.0 ml/l | 553.9 | 82.0 | 5965.5 | 5412.50 | 553.0 | 10.2 |
| $T_8$-Drought Stress-Control | 471.9 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |
| $T_9$-Drought Stress-Phylgreen 200 @ 6.0 ml/l | 559.7 | 87.8 | 6387.5 | 8825.00 | -2437.5 | -27.6 |
| F test | Sig. | | | | | |
| SEM± | 22.61 | | | | | |
| CD(0.05) | 67.77 | | | | | |
| CV (%) | 6.99 | | | | | |

Price of Greengram: Rs.7275 /-per quintal; Additional cost (chemical cost + spraying charges)

The return on investment (ROI) worked out and superior advantage of more net returns of Rs.8702.4/-$ha^{-1}$ (ROI 179.7%) and 6236.1/-$ha^{-1}$ (ROI 128.7%) realized at foliar spray of Phylgreen 200 @ 2.5 ml/l and Drought Stress Phylgreen 200 @ 2.5 ml/l, respectivelytwice at first flower appearance stage and seven days after 1$^{st}$ spray/pod setting stage than the other levels of foliar nutrition of the bio-stimulant Phylgreen 200 (Table 3).

## Conclusions

The Bio-stimulant, Phylgreen 200 (P-6084) supplied by the M/s.TradecorpRovensa India Pvt. Ltd., Pune was evaluated for its performancein greengram and observed that bio-stimulant, Phylgreen 200 (P-6084) and Drought Stress Phygreen 200at all the doses *i.e.* 2.0, 2.5, 3.0 and higher dose of6.0 ml per litre of water were found superior over control with respect to growth parameters and grain yield. Among different treatments, Phylgreen 200 (P-6084) @ 2.5 ml/l and Drought Stress Phygreen 200 @ 2.5 ml/l were found to be best in recording more grain yield and high Return on Investment (ROI). Moreover, the bio-stimulant, Phylgreen 200 (P-6084) has no phytotoxicity on greengram cropeven at higher dose (6 ml/l).

Drought is a prevalent stress factor especially in arid and semiarid areas and can affect different aspects of plant growth, development, and metabolism. Drought is a multidimensional stress factor, and hence its effects on plants are complex (Dubey and Pessarakli, 2002) [2] Its effects on plants can occur on a molecular level up to a whole-plant level. There are several reasons for drought in nature, including low rainfall, salinity, high temperature, and high intensity of light, among others (Abbas, 2013).

The water deficit often decreases the number of photons captured by the leaves because withered leaves are at a more acute angle to the sun's rays. Changes in the absorption characteristics of the leaves occur due to the shrinkage of the cells (Sharma *et al.*, 2014) [4] However, changes in chloroplasts and thylakoid during light capture and energy transfer centers are relatively small under water deficit conditions (Reddy *et al.*, 2004) [3]

The composition of biostimulants should present a variety of organic materials such as humic substances, seaweed extracts, organic matter, and amino acids in order to improve stress tolerance (Sharma and Dubey, 2005) [5]. The literature on biostimulants have been reporting an increase in enzyme activities involved in antioxidant functions, especially under stress conditions.

Investigations on the role of biostimulants in the physiological mode of action in plants subjected to drought stress should be continued, since considerable researches remain to be completed to gain a clearer understanding of how these products increase the physiological health of plants under water stress.

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