



## HERBICIDAL MANAGEMENT OF WEEDS IN IRRIGATED GREEN GRAM (*VIGNA RADIATA* L. WILCZEK)

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(Date of Receiving : 29-08-2025; Date of Acceptance : 16-11-2025)

Uncontrolled weed growth significantly reduces the productivity of short-duration irrigated green gram, necessitating timely and effective weed management. This study aimed to identify a sustainable herbicidal strategy for green gram by evaluating the efficacy, phytotoxic effects, yield impact, and economic viability of different herbicide treatments. The study compared pre-emergence (PE) applications of Pendimethalin as well as early post-emergence (EPoE) applications (20 DAS) of Imazethapyr + quizalofop ethyl and Sodium acifluorfen + Clodinafop propargyl. Among the treatments, PE Pendimethalin followed by (f/b) and Sodium acifluorfen + Clodinafop propargyl achieved significantly higher weed control efficiencies of 87.9%, (30 DAS) and 83.6 (45 DAS). Although these herbicide combinations initially induced oxidative stress and minor phytotoxicity, the green gram plants fully recovered within 15 days of application. These treatments led to substantial yield improvements (33%) and higher net returns (48.5%) compared to the weedy check. Thus, Application of PE Pendimethalin followed by EPoE Sodium Acifluorfen + Clodinafop propargyl emerge as promising weed management strategies for irrigated green gram offering both agronomic and economic benefits.

**Keywords :** Greengram, Herbicidal weed management, Sodium acifluorfen + Clodinafop propargyl.

### ABSTRACT

The critical weed competition period in green gram occurs between 20–30 days after sowing (Singh *et al.*, 1991), and studies indicate that weed-induced yield losses can range from 30% to 85% if not managed in a timely manner (Singh *et al.*, 2015).

The increasing labor shortages, high wages, and unpredictable rainfall during the rainy season make manual weeding challenging. The timing of weeding operations largely depends on rainfall predictions, making herbicide application a more feasible and effective alternative. Herbicide-based weed control, which requires less labor, ensures efficient weed management over large areas in a short period. While selective herbicides target weeds while preserving the crop, their prolonged and indiscriminate use may lead to herbicide resistance. Additionally, despite their selectivity, herbicides can induce oxidative stress, hinder leaf expansion, reduce biomass accumulation (Cobb & Reade, 2010), and cause phytotoxic damage to crops (Deligios *et al.*, 2018). Risk assessment is particularly crucial for pulse crops, as herbicides can

### Introduction

Pulses are often referred to as the "poor man's meat" due to their affordability as a protein source in India, which ranks 105th on the Global Hunger Index (2024). By 2050, the country's pulse requirement is projected to reach 39 million tonnes (IIPR, 2024). Among various pulse crops, green gram (*Vigna radiata* (L.) Wilczek) is particularly favored for its taste and nutritional value. This warm-season crop is primarily cultivated as a *kharif* or summer crop under irrigated conditions and as a *rabi* crop under rainfed conditions in Tamil Nadu. In the monsoon-driven Cauvery Delta, short-duration green gram is occasionally grown as a rice fallow crop in well-drained upland areas by marginal rice farmers as an additional harvest. However, frequent weed growth following rainfall or irrigation, coupled with the crop's low competitive ability due to slow early growth, significantly hampers productivity. Additionally, the short growth cycle of existing varieties (~60 – 65 days) offers little scope for recovery from setbacks caused by late weed removal.

interfere with the symbiotic relationship between legumes and rhizobia (Anderson *et al.*, 2004).

Pre-emergence application of pendimethalin, along with early post-emergence application of imazethapyr and quizalofop-ethyl, has shown promising results in green gram cultivation (Singh *et al.*, 2015). However, a single herbicide often proves ineffective against diverse weed species with different physiological and morphological traits. In contrast, using either pre-mixed or tank-mixed herbicides with different modes of action can provide comprehensive weed control, prevent shifts in weed populations, and reduce herbicide resistance (Banerjee *et al.*, 2018). For instance, early post-emergence application of a ready-mix formulation containing Sodium Acifluorfen (16.5%) and Clodinafop-Propargyl (8%) has been effective in controlling both monocot and dicot weeds in black gram (Harithavardhini, 2016; Lakra, 2017). Clodinafop-Propargyl inhibits acetyl-CoA carboxylase to control grassy weeds, while Acifluorfen targets both grassy and broadleaf weeds by inhibiting protoporphyrinogen oxidase (Caverzan *et al.*, 2019). However, herbicide efficiency is influenced by climatic and soil conditions, with optimal application rates varying across crops. There is currently limited information on the impact of newly available herbicide formulations in the Indian market on the growth and productivity of green gram. Given these factors, the

present study aims to evaluate the effects of newly introduced herbicide formulations on the growth, yield, and economic viability of irrigated green gram.

## Materials and Methods

### Experimental Site

Field experiments were conducted at the Department of Pulses, Tamil Nadu Agricultural University, Coimbatore, to evaluate green gram cultivation during the *kharif* and *rabi* seasons of 2023 and 2024. The experimental site is geographically located at 11°1'N latitude, 76°55'E longitude, at an altitude of 434 meters above mean sea level. Throughout the study period, weekly maximum temperatures ranged between 30.0°C and 34.5°C, while minimum temperatures varied from 20.0°C to 26.0°C. The mean relative humidity fluctuated between 62% and 84%. The cumulative rainfall recorded during the experimental periods was 172 mm and 104 mm in the *kharif* and *rabi* seasons of 2023–24, respectively, and 286.4 mm and 50 mm during the corresponding seasons in 2024–25. The soil at the study site was classified as sandy loam, with a texture comprising 55.45% sand, 23.25% silt, and 9.40% clay. The soil had a pH of 8.6 and an electrical conductivity of 0.53 dS m<sup>-1</sup>. Organic carbon content was 0.39%, while available nutrients included 183.4 kg N ha<sup>-1</sup>, 17.6 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>, and 550.1 kg K<sub>2</sub>O ha<sup>-1</sup>.

**Table 1 :** Details of the treatment adopted in the greengram experiment

| Treatment  | Dose                         | Time of application   |
|--|------------------------------|-----------------------|
| T <sub>1</sub> - PE Pendimethalin 30 % EC + one Hand Weeding (HW) on 25DAS   | 1 kg a.i./ha                 | 3 DAS<br>HW on 25 DAS |
| T <sub>2</sub> - EPoE Imazethapyr 10% SL + Quizalofop ethyl 5% EC (tank mix) on 20 DAS                                     | 50 g a.i./ha + 50 g a.i./ ha | 20 DAS                |
| T <sub>3</sub> - EPoE Sodium acifluorfen 16.5% + Clodinafop-propargyl 8% EC (Ready mix) on 20 DAS                          | 175 g a.i /ha                | 20 DAS                |
| T <sub>4</sub> - PE Pendimethalin on 3 DAS followed by EPoE Sodium acifluorfen 16.5% + Clodinafop-propargyl 8% EC on 20DAS | 1 kg a.i./ha + 175 g a.i /ha | 3 DAS + 20 DAS        |
| T <sub>5</sub> - Hand weeding twice (20 and 40 DAS)  | -                            | 20 and 40 DAS         |
| T <sub>6</sub> - Weedy check   | -                            | -                     |

EC- Emulsifiable Concentrate; SL- Soluble Liquid; DAS- days after sowing

### Treatment Details

The experiment comprised six treatments involving pre-emergence (PE) and early post-emergence (EPoE) herbicides, as well as manual weeding (twice) and a weedy check (Table 1). The treatments were arranged in a randomized block design (RBD) with three replications, covering a gross experimental plot area of 5 m × 4 m. Herbicide applications were carried out using a knapsack sprayer

fitted with a flat-fan nozzle, delivering a spray volume of 500 L water per hectare.

### Sowing and Crop Management

The experimental field was thoroughly prepared using a tractor-drawn disc plough followed by an eleven-tyne cultivator to achieve fine pulverization. After harrowing, the land was leveled using a tractor-drawn leveler. Green gram (Cv. VBN 6) seeds were pre-treated with the recommended *Rhizobium* strain, supplied by the Department of Agricultural

Microbiology, TNAU, Coimbatore. Sowing was carried out in both seasons at a row-to-row spacing of 30 cm and a plant-to-plant spacing of 10 cm. A recommended basal fertilizer dose of 25 kg N, 50 kg P<sub>2</sub>O<sub>5</sub>, and 25 kg K<sub>2</sub>O was applied at sowing, along with an additional 20 kg N. The nutrient sources used were urea (N), single super phosphate (SSP) (P<sub>2</sub>O<sub>5</sub>), and muriate of potash (K<sub>2</sub>O). Additionally, the sulfur requirement of the crop was met through Single Super Phosphate (SSP). The crop was cultivated under irrigated conditions, adhering to the recommended agronomic practices outlined in the TNAU Crop Production Guide (2020).

### Observations on Weeds and Crops

Weed and crop observations were recorded at 30 and 45 days after sowing (DAS). In each green gram plot, an observation area of 50 cm in length (along the row) and 100 cm in width (across rows) was marked. The designated area included one central row per plot.

Weeds from the sampled area were identified species-wise and classified into three categories *viz.*, Grasses, Broad-leaved weeds, Sedges. Weed density for each category was determined by manual counting, followed by dry weight measurement. For dry weight assessment, weeds were first sun-dried for two days, then oven-dried at 70 ± 5°C for 48 hours before recording the final weight. The Weed Control Efficiency (WCE) was calculated using the following equation:

$$WCE \% = \frac{WDM_c - WDM_t}{WDM_c} \times 100$$

Where WDM<sub>c</sub> is the weed dry matter (g m<sup>-2</sup>) in control plot, i.e. the weedy check and WDM<sub>t</sub> is the weed dry matter (g m<sup>-2</sup>) in the treated plot. Similarly on 45 DAS, plant sampling was done for nodule count and leghaemoglobin estimation. Leghaemoglobin content was determined by using the method proposed by Wilson and Reisenauer (1963). Approximately 0.5 g of fresh nodule tissue was macerated in a 10 ml round-bottom centrifuge containing 3 ml Drabkin's solution. To facilitate the settling of larger tissue particles, the mixture was centrifuged at 10,000 rpm for 20 minutes. The absorbance of the clear supernatant was measured at 540 nm, using Drabkin's solution as a blank. The leghaemoglobin content (mg g<sup>-1</sup>) was then determined using a standard curve.

The chlorophyll content in green gram leaves was estimated following the method of Hiscox and Israelstam (1979). A 0.05 g leaf sample was extracted in 10 mL dimethyl sulfoxide (DMSO) and heated at 60°C for 3 hours in a hot air oven. After cooling to room temperature, the optical density of the

supernatant was recorded at 480 nm, 510 nm, and 652 nm using a UV-VIS spectrophotometer. The total chlorophyll and carotenoids were worked out using the following formulae as proposed by Arnon (1949)-

$$\text{Total chlorophyll (mg/g)} = \frac{D652 \times 100}{34.5} \times \frac{V}{1000 \times W}$$

$$\text{Carotenoids mg/g} = 7.6D480 - 1.49D510 \times \frac{V}{1000 \times W}$$

where D is the optical density at a given wavelength, V is the final volume of DMSO (ml), and W is the fresh weight of sample (g). The phytotoxic effects of herbicide treatments, such as chlorosis, white blotching, and bronzing of green gram leaves, were assessed at 7, 15, and 25 days after herbicide application (DAHA) using the rating scale suggested by Rao (2000). Crude protein content (%) was determined by multiplying the seed nitrogen content by 6.25 (Intl, 1995).

### Superoxide Dismutase (SOD) Activity Assay

The superoxide dismutase (SOD) activity in green gram leaves was measured based on the inhibition of photoreduction of nitro blue tetrazolium (NBT), as described by Dhindsa *et al.* (1981). The enzyme was extracted by grinding 0.5 g of frozen leaf samples in 10 ml of 0.1 M potassium phosphate buffer (pH 7.5) containing 0.5 mM EDTA using a prechilled mortar and pestle. The resulting homogenate was filtered through cheesecloth, and the filtrate was centrifuged at 10,000 rpm for 10 minutes at 0°C in a refrigerated centrifuge. The reaction mixture (3 ml) contained: 75 mM NBT, 13 mM methionine, 2 mM riboflavin, 0.1 mM EDTA, 50 mM phosphate buffer (pH 7.8) and 50 ml enzyme extract. The reaction mixture was transferred to test tubes and exposed to fluorescent light (15 W) for 10 minutes. The reaction was terminated by turning off the light and wrapping the tubes in black cloth. Absorbance was measured at 560 nm, with a non-irradiated reaction mixture serving as the control. One unit of SOD activity was defined as the enzyme amount required to inhibit 50 per centage of NBT photoreduction. The leaf area index (LAI) was estimated using the area-weight relationship method (Radford, 1967).

### Yields and Economics

The net area considered for biological and seed yield estimation of green gram was 12.0 m<sup>2</sup>, with the final yield expressed in kg ha<sup>-1</sup>. Observations on yield attributes were recorded by randomly selecting five plants from the net plot for sampling. Harvest index was calculated by using the following formula

$$\text{Harvest index} = \frac{\text{Seed yield (kg/ha)}}{\text{Biological yield (kg/ha)}}$$

The input costs for field preparation, seeds, labor for sowing, fertilizer and irrigation application, interculture operations, harvesting, and threshing were aggregated to calculate the common cost of cultivation, irrespective of treatments. The cost of weed control measures was considered as the treatment-specific cost.

The gross returns (GR) were computed using the minimum support price (MSP) set by the Government of India for green gram seeds. The net returns (NR) were determined by subtracting the cost of cultivation from the gross returns. The benefit : cost (GR:C) was worked out by dividing GR with cost of cultivation.

### Microbiological Observations

Fresh soil samples (200–250 g) were collected from each plot at a 15 cm depth using an auger (5 cm diameter) at four time points: Initial (before sowing), 5 days after herbicide application (5 DAHA), 25 DAHA and 50 DAHA.

Soil was sampled from five locations between green gram rows. Microbial populations were enumerated using the serial dilution technique and agar/pour plate method (Pramer & Schmidt, 1964). The colony-forming units (cfu) of fungi, total bacteria, and actinomycetes were counted using the media *viz.*, Martin's rose bengal streptomycin agar medium for fungi, Thornton's agar medium for total bacteria, Jensen's agar medium for actinomycetes. Following serial dilution and plating, microbial incubation was carried out at 30°C. Bacterial populations were scored within 3 days, while actinomycetes and fungi were scored within 5–7 days (Das *et al.*, 2010).

### Statistical Analysis

Data on crop and weed parameters were analyzed using analysis of variance (ANOVA) for a randomized block design (RBD) following the methods of Gomez & Gomez (1984). Weed density data were subjected to square root transformation  $\sqrt{x+0.5}$  as suggested by Panse & Sukhatme (1978). ANOVA results showed non-significant variation ( $p > 0.05$ ) across

experimental years, treatments, and year-treatment interactions. Hence, the average data from both years were reported. Duncan's multiple range test (DMRT) was used to compare treatment means at  $p \leq 0.05$ , provided that the initial F-test was significant at  $p \leq 0.05$ . Pearson's correlation coefficient ( $n=10$ ) was computed to evaluate associations between different parameters using SPSS (Version 25). Graphs were generated using Microsoft Excel (Version 2016).

## Results

### Weed Growth and Weed Control Efficiency in Green Gram

The predominant weed flora in the experimental field comprised: Grasses *viz.*, Bermuda grass (*Cynodon dactylon*), Egyptian crowfoot grass (*Dactyloctenium aegyptium*), Peacock plume grass (*Chloris barbata*), and Goose grass (*Eleusine indica*). Broad-leaf weeds (BLW): Horse purslane (*Trianthema portulacastrum*), Pigweed (*Amaranthus viridis*), Asthma herb (*Euphorbia hirta*), and Alligator weed (*Alternanthera philoxeroides*). Sedges: Purple nutsedge (*Cyperus rotundus*). Among these, broad-leaf weeds exhibited the highest density, surpassing grasses and sedges throughout the study (Table 2). Hand weeding at 20 and 40 DAS achieved the highest weed control efficiency (WCE) of 92.8–95.1%, significantly reducing weed density and dry matter accumulation at both 30 and 45 DAS (Tables 2 and 3). Herbicidal treatments effectively suppressed weeds compared to the weedy check.

Among herbicides, early post-emergence (EPoE) application of Na Acifluorfen + Clodinafop propargyl effectively reduced broad-leaf and grass populations at 30 and 45 DAS (Table 2). However, no significant difference in grass dry matter was observed between Na Acifluorfen + Clodinafop propargyl and other treatments (Table 3). Lowest weed biomass ( $12.3 \text{ g m}^{-2}$ ) was recorded in plots treated with Na Acifluorfen + Clodinafop propargyl, followed closely by hand weeding twice (no significant difference between them).

**Table 2 :** Effect of weed management on grass, broad-leaved, sedges, and total weed (nos.  $\text{m}^{-2}$ ) in green gram (Pooled data of two years)

| Treatments  | 30 DAS         |                |               | 45 DAS        |               |               | Total          |                |
|---|----------------|----------------|---------------|---------------|---------------|---------------|----------------|----------------|
|   | Grasses        | BLW            | Sedges        | Grasses       | BLW           | Sedges        | 30 DAS         | 45 DAS         |
| T <sub>1</sub> - PE Pendimethalin + one HW                        | 1.5bc<br>(1.8) | 1.8cd<br>(2.8) | 1.4d<br>(1.5) | 1.9b<br>(3.3) | 2.8b<br>(7.6) | 2.5b<br>(6.0) | 2.6d<br>(6.1)  | 4.2c<br>(16.9) |
| T <sub>2</sub> - EPoE Imazethapyr + Quizalofop ethyl              | 1.6b<br>(2.0)  | 2.0c<br>(3.5)  | 2.4b<br>(5.5) | 2.0b<br>(3.4) | 2.8b<br>(7.5) | 2.6b<br>(6.5) | 3.4b<br>(11.0) | 4.2b<br>(17.4) |
| T <sub>3</sub> - EPoE Sodium acifluorfen + Clodinafop - propargyl | 1.7b<br>(2.4)  | 2.3b<br>(5.0)  | 2.3b<br>(4.8) | 2.0b<br>(3.7) | 2.7b<br>(7.0) | 2.6b<br>(6.2) | 3.6b<br>(12.2) | 4.2b<br>(16.9) |

|   |                |                |               |                |                |                |                |                |
|---|----------------|----------------|---------------|----------------|----------------|----------------|----------------|----------------|
| T <sub>4</sub> - PE Pendimethalin /b EPoE Sodium acifluorfen + Clodinafop-propargyl | 1.3c<br>(1.2)  | 1.9c<br>(3.0)  | 2.0c<br>(3.7) | 1.6c<br>(2.4)  | 2.6bc<br>(6.4) | 2.4bc<br>(5.3) | 2.9c<br>(7.9)  | 3.8d<br>(14.1) |
| T <sub>5</sub> - Hand weeding twice   | 1.1cd<br>(0.8) | 1.6e<br>(2.0)  | 1.2d<br>(1.0) | 1.0d<br>(0.5)  | 1.9d<br>(3.1)  | 1.1d<br>(0.8)  | 2.1e<br>(3.8)  | 2.2e<br>(4.4)  |
| T <sub>6</sub> - Weedy check  | 4.1a<br>(16.0) | 4.9a<br>(24.0) | 2.9a<br>(8.0) | 4.9a<br>(23.2) | 6.2a<br>(37.4) | 3.7a<br>(13.5) | 7.0a<br>(48.0) | 8.6a<br>(74.1) |

- Square-root transformation of weed density  $\sqrt{(x+0.5)}$  was done before statistical analysis, and original weed density values are provided in parentheses.
- Means with different grouping letter in a column are significantly different at  $p \leq 0.05$  by Duncan's multiple range test.

**Table 3 :** Effect of weed management on dry matter of grass, broad-leaved, sedges, and total weed ( $\text{g m}^{-2}$ ) and weed control efficiency (WCE %) in green gram (Pooled data of two years)

| Treatments  | 30 DAS         |                |               | 45 DAS         |                |                | Total          |                | WCE (%) |        |
|---|----------------|----------------|---------------|----------------|----------------|----------------|----------------|----------------|---------|--------|
|   | Grasses        | BLW            | Sedges        | Grasses        | BLW            | Sedges         | 30 DAS         | 45 DAS         | 30 DAS  | 45 DAS |
| T <sub>1</sub> - PE Pendimethalin + one HW  | 1.7c<br>(2.5)  | 2.4d<br>(4.5)  | 1.3e<br>(1.0) | 2.5c<br>(5.9)  | 2.7b<br>(7.0)  | 3.0c<br>(8.3)  | 3.0e<br>(8.0)  | 4.2d<br>(21.2) | 89.7    | 78.3   |
| T <sub>2</sub> - EPoE Imazethapyr + Quizalofop ethyl                                | 1.8c<br>(2.7)  | 2.6c<br>(6.1)  | 2.3b<br>(4.6) | 2.5c<br>(5.9)  | 2.6c<br>(6.3)  | 3.0c<br>(8.7)  | 3.7c<br>(13.4) | 4.6c<br>(20.9) | 82.8    | 78.7   |
| T <sub>3</sub> - EPoE Sodium acifluorfen + Clodinafop -propargyl                    | 2.0b<br>(3.5)  | 3.0b<br>(8.3)  | 2.0c<br>(4.0) | 2.6b<br>(6.1)  | 2.5d<br>(5.9)  | 3.1b<br>(9.1)  | 4.0b<br>(15.8) | 4.8b<br>(21.1) | 79.7    | 78.4   |
| T <sub>4</sub> - PE Pendimethalin /b EPoE Sodium acifluorfen + Clodinafop-propargyl | 1.4d<br>(1.5)  | 2.3e<br>(4.7)  | 1.9d<br>(3.2) | 2.1d<br>(3.8)  | 2.5d<br>(5.5)  | 2.7d<br>(6.8)  | 3.2d<br>(9.4)  | 4.1e<br>(16.1) | 87.9    | 83.6   |
| T <sub>5</sub> - Hand weeding twice   | 1.3d<br>(1.1)  | 2.0f<br>(3.6)  | 1.2f<br>(0.9) | 1.2e<br>(0.9)  | 1.8e<br>(2.8)  | 1.3e<br>(1.1)  | 2.5f<br>(5.6)  | 2.3f<br>(4.8)  | 92.8    | 95.1   |
| T <sub>6</sub> - Weedy check  | 5.0a<br>(24.3) | 6.9a<br>(46.8) | 2.7a<br>(6.9) | 6.8a<br>(45.2) | 5.7a<br>(32.2) | 4.6a<br>(20.5) | 8.9a<br>(78.0) | 9.9a<br>(97.9) | -       | -      |

- Square-root transformation of weed drymatter  $\sqrt{(x+0.5)}$  was done before statistical analysis, and original weed density values are provided in parentheses.
- Means with different grouping letter in a column are significantly different at  $p \leq 0.05$  by Duncan's multiple range test.

#### Nodulation efficiency, leaf area index and phytotoxicity in green gram

The nodulation efficiency in terms of numbers (nos.) of nodules and leghaemoglobin content was significantly influenced by weed control practices (Table 4). Hand weeding twice recorded the highest number of nodules (39) and leghaemoglobin content ( $1.68 \text{ mg g}^{-1}$ ) whereas, the weedy check had the lowest nodulation efficiency.

Herbicidal treatments irrespective of mode of application had poor nodulation efficiency than hand weeding. Nevertheless, among the herbicides, Na Acifluorfen + Clodinafop propargyl produced significantly higher nodules (31) and leghaemoglobin ( $1.59 \text{ mg g}^{-1}$ ). The treatments which had EPoE applied herbicides recorded significantly higher nodule numbers and leghaemoglobin than PE herbicide applied one. Na Acifluorfen + Clodinafop propargyl recorded significantly higher nodulation efficiency than Imazethapyr + Quizalofop ethyl. At seven days

after herbicide application (DAHA), PE applied herbicides did not cause any crop injury, whereas the EPoE applied herbicides showed phytotoxicity to the extent of 5 % (Table 4). Among the EPoE herbicides, Na Acifluorfen + Clodinafop propargyl had comparatively higher toxic effect (4 - 5%) on green gram resulting in significant reduction in leaf area index (LAI) on 30 DAS (Table 4). The extent of leaf area reduction in these two treatments ranged from 7 to 15% over PE herbicidal management. However, Na Acifluorfen + Clodinafop propargyl recorded significantly higher LAI compared to weedy check with the magnitude of increment ranging between 13.0 and 18.0 % on 30 DAS. Acifluorfen Na + Clodinafop propargyl showed only 1.0% phytotoxicity on 15 DAHA. However, no phytotoxic effects were observed on 25 DAHA. The percent increment in LAI from 30 DAS to 45 DAS was highest (34.5%) in hand weeding twice closely followed by Na Acifluorfen + Clodinafop propargyl (31.9%) and Pendimethalin + HW on 25 DAS (29.9%).

**Table 4 :** Effect of weed management on nodule numbers plant<sup>-1</sup>, leghaemoglobin content (mg g<sup>-1</sup>), phytotoxicity (%), Leaf Area Index in green gram (Pooled data of two years)

| Treatments  | 45 DAS                         |                          | Phytotoxicity (%) |         |         | LAI    |        |
|---|--------------------------------|--------------------------|-------------------|---------|---------|--------|--------|
|   | Nodule Nos plant <sup>-1</sup> | LC (mg g <sup>-1</sup> ) | 7 DAHA            | 15 DAHA | 25 DAHA | 30 DAS | 45 DAS |
| T <sub>1</sub> - PE Pendimethalin + one HW  | 16d                            | 1.32d                    | 0                 | 0       | 0       | 2.41b  | 3.13b  |
| T <sub>2</sub> - EPoE Imazethapyr + Quizalofop ethyl                                | 20c                            | 1.48c                    | 2                 | 0       | 0       | 2.27c  | 2.89c  |
| T <sub>3</sub> - EPoE Sodium acifluorfen + Clodinafop -propargyl                    | 31b                            | 1.59b                    | 4                 | 0       | 0       | 2.16d  | 2.83c  |
| T <sub>4</sub> - PE Pendimethalin /b EPoE Sodium acifluorfen + Clodinafop-propargyl | 17d                            | 1.33d                    | 5                 | 1       | 0       | 2.12e  | 2.74d  |
| T <sub>5</sub> - Hand weeding twice   | 39a                            | 1.68a                    | 0                 | 0       | 0       | 2.67a  | 3.59a  |
| T <sub>6</sub> - Weedy check  | 12f                            | 1.34d                    | 0                 | 0       | 0       | 1.96e  | 2.18e  |

LC - Leghaemoglobin content, LAI -Leaf Area Index

### Total Chlorophyll and Carotenoid Contents, and SOD Activity in Green Gram Leaves

EPoE herbicides caused a significant reduction in chlorophyll concentration (22.9–33.3%) on 30 DAS (10 DAHA) compared to hand weeding (Table 5). The Na Acifluorfen + Clodinafop propargyl treatment, both alone and in combination with PE pendimethalin, recorded the greatest decline in chlorophyll content (0.90–0.98 mg g<sup>-1</sup>). However, by 45 DAS, Na Acifluorfen + Clodinafop propargyl showed a significant recovery, recording the highest chlorophyll content (1.89 mg g<sup>-1</sup>) among all herbicides, indicating that the crop recovered from initial phytotoxic effects. Similarly, Na Acifluorfen + Clodinafop propargyl also significantly reduced carotenoid content initially (0.437–0.453 mg g<sup>-1</sup>), with recovery observed by 45

DAS (0.483–0.497 mg g<sup>-1</sup>). Hand weeding recorded the highest chlorophyll (1.35 and 1.99 mg g<sup>-1</sup>) and carotenoid contents (0.482 and 0.506 mg g<sup>-1</sup>) at both observation stages. Regarding Superoxide Dismutase (SOD) activity, Na Acifluorfen + Clodinafop propargyl, Imazethapyr + Quizalofop ethyl, and the weedy check exhibited significantly higher SOD activity at 30 DAS, with no significant differences between them (Table 5). The increase in SOD activity ranged from 29% to 38% over hand weeding. However, at 45 DAS, SOD activity in Na Acifluorfen + Clodinafop propargyl and hand weeding showed no significant variation, while all EPoE herbicides recorded higher SOD activity initially, with the trend reversing later.

**Table 5 :** Effect of weed management on total chlorophyll (mg g<sup>-1</sup>) and carotenoid contents (mg g<sup>-1</sup>) and superoxide dismutase activity (unit mg<sup>-1</sup> protein h<sup>-1</sup>) in green gram leaves (Pooled data of two years)

| Treatments  | Total Chlorophyll |        | Carotenoid |         | SOD     |        |
|---|-------------------|--------|------------|---------|---------|--------|
|   | 30 DAS            | 45 DAS | 30 DAS     | 45 DAS  | 30 DAS  | 45 DAS |
| T <sub>1</sub> - PE Pendimethalin + one HW  | 1.13b             | 1.62d  | 0.468ab    | 0.471ab | 25.54c  | 48.6a  |
| T <sub>2</sub> - EPoE Imazethapyr + Quizalofop ethyl                                | 1.04c             | 1.73c  | 0.472b     | 0.483bc | 28.94bc | 44.2b  |
| T <sub>3</sub> - EPoE Sodium acifluorfen + Clodinafop -propargyl                    | 0.98d             | 1.89b  | 0.453c     | 0.497d  | 30.56ab | 37.7cd |
| T <sub>4</sub> - PE Pendimethalin /b EPoE Sodium acifluorfen + Clodinafop-propargyl | 0.90e             | 1.75c  | 0.437e     | 0.483bc | 32.86a  | 38.9c  |
| T <sub>5</sub> - Hand weeding twice   | 1.35a             | 1.99a  | 0.482a     | 0.506a  | 23.65d  | 36.2cd |
| T <sub>6</sub> - Weedy check  | 0.95de            | 1.50e  | 0.448d     | 0.453e  | 32.39a  | 50.2a  |

### Yield Attributes, Yield, and Crude Protein Content in Green Gram

Among yield attributes, hand weeding recorded the highest number of pods per plant (22) and seeds per pod (12), while EPoE application of Imazethapyr + Quizalofop ethyl had the highest 100-seed weight (3.65 g) (Table 6). No significant variation in pods per plant was observed between hand weeding and Pendimethalin + Na Acifluorfen + Clodinafop propargyl, both of which had 20 pods per plant. Hand weeding resulted in the highest yield (805 kg ha<sup>-1</sup>),

with no significant yield variation between PE + EPoE herbicide applications and hand weeding. Application of Pendimethalin followed by Na Acifluorfen + Clodinafop propargyl achieved the highest yield (757 kg ha<sup>-1</sup>) among the herbicide treatments, 11–16% higher than EPoE herbicide-only treatments. Crude protein content varied between 21.7% and 24.1%, with Na Acifluorfen + Clodinafop propargyl (regardless of combination with PE Pendimethalin) recording the highest levels (23.7–24.1%). This was statistically similar to hand weeding.

**Table 6 :** Effect of weed management on yield attributes, seed yield, and crude protein content in greengram (Pooled data of two years)

| Treatments  | Pods plant <sup>-1</sup> | Seeds pod <sup>-1</sup> | 100 seed weight (g) | Seed Yield (kg ha <sup>-1</sup> ) | Harvest Index | Crude Protein % |
|---|--------------------------|-------------------------|---------------------|-----------------------------------|---------------|-----------------|
| T <sub>1</sub> - PE Pendimethalin + one HW  | 17c                      | 11b                     | 3.52b               | 732b                              | 0.34a         | 23.0ab          |
| T <sub>2</sub> - EPoE Imazethapyr + Quizalofop ethyl                                | 16d                      | 10c                     | 3.65a               | 654d                              | 0.33ab        | 22.5b           |
| T <sub>3</sub> - EPoE Sodium acifluorfen + Clodinafop -propargyl                    | 15d                      | 12a                     | 3.43cd              | 683c                              | 0.31bc        | 23.7a           |
| T <sub>4</sub> - PE Pendimethalin /b EPoE Sodium acifluorfen + Clodinafop-propargyl | 20ab                     | 11b                     | 3.39d               | 757ab                             | 0.30cd        | 24.1a           |
| T <sub>5</sub> - Hand weeding twice   | 22a                      | 12a                     | 3.45bc              | 805a                              | 0.32b         | 23.8a           |
| T <sub>6</sub> - Weedy check  | 10e                      | 8d                      | 3.03e               | 569e                              | 0.28e         | 21.7c           |

### Soil Microbial Population

At the time of sowing, no significant variation in the soil microbial population was observed across treatments (Fig. 1a-c). However, by 5 DAHA, a substantial decrease in bacterial, actinomycetes, and fungal populations was noted, irrespective of herbicide treatments. Pendimethalin (PE) application resulted in the lowest microbial population. By 25 DAHA, a marginal increase in microbial population was noted, followed by a steep rise at 50 DAHA, with EPoE herbicides showing a significantly higher microbial build-up at 50 DAHA than PE herbicides. Despite non-significant variation in bacterial and actinomycetes populations among the EPoE herbicides, Na Aciflofen + Clodinafop propargyl recorded the lowest fungal population. Notably, the microbial population at 50

DAHA was substantially higher than at sowing in all herbicidal treatments.

### Economic Analysis

In terms of cost of production, weedy check had the lowest and hand weeding the highest (Table 7). Despite hand weeding yielding the highest gross return (GR) of Rs. 69,161 ha<sup>-1</sup>, it resulted in a substantially lower net return (NR) and benefit-cost ratio (BCR) compared to other herbicidal treatments. The high cost of manual labor made hand weeding an expensive option, offsetting its yield advantage. EPoE application of Na Aciflofen + Clodinafop propargyl, either alone or with PE Pendimethalin, exhibited substantially higher net returns (Rs. 34,781–39,203 ha<sup>-1</sup>) and BCR (2.42–2.48) compared to other weed management practices.

**Table 7 :** Effect of weed management on economics of green gram (Pooled data of two years)

| Treatments  | COP (Rs. ha <sup>-1</sup> ) | GR (Rs. ha <sup>-1</sup> ) | NR (Rs. ha <sup>-1</sup> ) | BCR  | Additional income (Rs. ha <sup>-1</sup> ) |
|---|-----------------------------|----------------------------|----------------------------|------|---|
| T <sub>1</sub> - PE Pendimethalin + one HW  | 28850                       | 66197                      | 37347                      | 2.29 | 9946                                      |
| T <sub>2</sub> - EPoE Imazethapyr + Quizalofop ethyl                                | 25000                       | 56811                      | 31811                      | 2.27 | 4410                                      |
| T <sub>3</sub> - EPoE Sodium acifluorfen + Clodinafop -propargyl                    | 24500                       | 59281                      | 34781                      | 2.42 | 7380                                      |
| T <sub>4</sub> - PE Pendimethalin /b EPoE Sodium acifluorfen + Clodinafop-propargyl | 26500                       | 65703                      | 39203                      | 2.48 | 11802                                     |
| T <sub>5</sub> - Hand weeding twice   | 32000                       | 69161                      | 37890                      | 2.18 | 10490                                     |
| T <sub>6</sub> - Weedy check  | 23000                       | 49401                      | 26401                      | 2.15 | 9946                                      |

COP – Cost of Production, GR- Gross return, NR- Net return, BCR- Benefit Cost Ratio

### Discussion

#### Effect of herbicidal management on weed growth in greengram

In this study, variations in Weed Control Efficiency (WCE) among herbicides were attributed to differences in chemical structure and mode of action. While none of the herbicides achieved a WCE statistically equivalent to hand weeding, Na Acifluorfen + Clodinafop propargyl exhibited significantly higher WCE than other herbicides across both observation periods. The phytotoxic effects of Clodinafop propargyl and Acifluorfen manifest within one to three weeks (Noshadi *et al.*, 2017) and three to

seven days (Extoxnet, 1993) post-application, respectively. Additionally, Acifluorfen efficacy is enhanced under humid and high-temperature conditions (Extoxnet, 1993), making this herbicide combination particularly suitable for the hot and humid monsoons of India. The experimental green gram field was dominated by broad-leaved weeds, followed by grassy weeds. Since Clodinafop propargyl effectively targets grasses by inhibiting acetyl-CoA carboxylase, and Acifluorfen controls both grasses and broad-leaved weeds by inhibiting protoporphyrinogen oxidase (Harithavardhini, 2016; Caverzan *et al.*, 2019), this ready-mix formulation offers broad-spectrum weed control in green gram. Previous studies by

Harithavardhini (2016) and Lakra (2017) also found that a combined application of Clodinafop propargyl (8% EC) and Acifluorfen Na (16% EC) was highly effective in reducing weed density and growth of both dicot and monocot weeds in black gram.

#### **Effect of herbicidal management on carotenoids and chlorophyll contents and SOD activity in green gram leaves**

Cobb and Reade (2010) reported that despite selectivity mechanisms, herbicides may reduce leaf area index and biomass accumulation in crops. Additionally, herbicidal management can induce oxidative stress in non-target crops due to the generation of reactive oxygen species (ROS).

In the present study, both EPoE herbicides (ready-mix and tank-mix) exhibited significantly higher SOD activity initially compared to PE herbicides, indicating herbicide-induced oxidative stress. Previous studies have also reported increased antioxidant enzyme activity (SOD, catalase, and peroxidase) in green gram (Batish *et al.*, 2006) and black gram (Bajpai & Srivastava, 2015) in response to herbicide-induced oxidative stress. SOD (superoxide dismutase) plays a crucial role in scavenging reactive superoxide anions, converting them into oxygen and hydrogen peroxide (Sharma *et al.*, 2012). Carotenoids, a potent non-enzymatic antioxidant, may also help mitigate oxidative stress by quenching singlet oxygen, a highly reactive ROS, thereby preventing lipid peroxidation and membrane damage (Das & Sarkar, 2018; Pallet & Young, 1993). On 30 DAS, Na Acifluorfen + Clodinafop propargyl and Imazethapyr + Quizalofop ethyl recorded significantly lower carotenoid content than hand weeding, possibly leading to ROS-induced chlorophyll degradation (Kim *et al.*, 2004). Both Imazethapyr and Acifluorfen are known to reduce chlorophyll content and biomass accumulation, while Clodinafop and Quizalofop ethyl induce oxidative stress in crops (Lukatkin *et al.*, 2013). However, by 45 DAS, a decline in SOD activity along with a significant increase in carotenoid and chlorophyll contents suggests that the oxidative stress subsided, with no lasting physiological damage as evidenced by satisfactory crop yield. Beyond herbicidal effects, weed interference itself can trigger antioxidant defense mechanisms in crops, leading to substantial energy and yield costs (Caverzan *et al.*, 2019). This trend was observed in the current study, where PE-applied herbicides exhibited higher SOD activity during later crop growth stages, possibly due to increased weed competition and their lower weed control efficiency. The weedy check consistently recorded the highest SOD activity, but with poor yield performance,

indicating that weed-induced oxidative stress incurs significant energy costs.

#### **Effect of herbicidal management on soil microbial population**

An unintended consequence of herbicidal management is its impact on soil microbial ecology. In the present study, herbicide application significantly reduced microbial populations immediately after treatment (5 DAHA) (Fig. 1(a)–(c)), with PE herbicides exhibiting a stronger inhibitory effect than EPoE herbicides. This difference may be attributed to the greater foliage development by the time of EPoE application (20 DAS), which likely acted as a barrier, limiting herbicide-soil contact and thereby reducing its adverse effects on soil microbial communities. Previous studies have reported that herbicidal formulations and application doses significantly influence soil microbial populations (Banerjee *et al.*, 2018), with inhibitory effects being most pronounced immediately after herbicide application. Since herbicides contain biologically active compounds, their initial high concentration in soil can negatively impact microbial growth and activity. However, this deleterious effect tends to diminish over time due to microbial adaptation or herbicide degradation (Singh *et al.*, 2018). This trend was also observed in the present study, as evidenced by a gradual increase in microbial populations beyond 25 DAHA. Interestingly, by 50 DAHA, the soil microbial population surpassed the initial levels across all treatments. This suggests that soil microbes play a crucial role in herbicide degradation, potentially using them as a carbon source, which reduces herbicide-induced toxicity and enhances soil microbial diversity (Vandana *et al.*, 2012). This increase in microbial activity can, in turn, have positive effects on soil health and nutrient availability, contributing to improved crop performance.

#### **Effect of herbicidal management on yield attributes, yield, protein content, and profitability in green gram**

Weed infestation significantly affects crop growth and productivity by intensifying competition for essential resources. The substantial increase in green gram productivity following Na Acifluorfen + Clodinafop propargyl treatment was likely due to enhanced crop competitiveness, as reflected in its high WCE and LAI (Tables 3 and 4). This treatment effectively reduced crop-weed competition, allowing for better resource utilization, which ultimately led to higher yields. The resulting high BCR further underscores the economic viability of this weed management strategy (Table 7). A correlation study

revealed that WCE at 30 DAS was significantly and positively correlated with LAI at 60 DAS, leghaemoglobin content at 45 DAS, yield components, and final yield (Table 8). This suggests that effective weed suppression within the first 30 DAS is critical for optimizing crop performance and maximizing yield. Singh *et al.* (1991) similarly reported that a weed-free period of 30 days post-sowing increased green gram yield by 25–30% compared to crops that experienced weed competition during this period. Furthermore, LAI and nodulation efficiency exhibited a significant positive influence on yield and crude protein content (Table 8), highlighting the indirect role of weed management in enhancing final yield by facilitating legume-rhizobium symbiosis and canopy development. Efficient weed control minimizes competition for water, light, and nutrients, which in turn enhances nodulation, leading to nitrogen-rich seed production. Although a larger leaf area may theoretically reduce net assimilation rate by diluting the photosynthetic apparatus per unit area, a dense canopy provides a smothering effect against weeds, improving crop competitiveness. This effect is particularly beneficial during the monsoon season, when frequent rainfall triggers new weed flushes. A dense canopy can restrict weed seed germination by limiting light penetration (Fenner, 1980). Moreover, in indeterminate crops like green gram, continued leaf area expansion during the terminal stages can provide sufficient photosynthetic area, facilitating source-to-sink partitioning and ensuring that late-formed flowers successfully develop into pods.

Notably, despite significantly lower pod and seed counts per plant, Imazethapyr (40 g a.i.  $\text{ha}^{-1}$ ) produced a statistically comparable yield to that of hand weeding and Acifluorfen Na + Clodinafop propargyl (187.5 g a.i.  $\text{ha}^{-1}$ ). This could be attributed to its significantly higher 100-seed weight (3.31 g) (Table 6). Previous studies (Rohman *et al.*, 2003) have demonstrated that all major yield-attributing traits positively and directly impact green gram yield, a trend also observed in the current study (Table 8). This suggests that higher seed weight in Imazethapyr-treated plants compensated for the lower number of pods and seeds per pod, ensuring satisfactory yield levels. The inhibitory effect of Imazethapyr on protein synthesis, likely due to its interference with acetohydroxy acid synthase (AHAS) activity (Scarpone *et al.*, 1997), may have altered starch deposition patterns, leading to a higher starch-to-protein ratio in seeds. This raises concerns regarding its long-term sustainability, especially given India's national focus on both yield enhancement and protein-rich food production to combat protein-energy malnutrition. While yield intensification is a priority,

herbicidal interventions that compromise protein content may not align with long-term nutritional goals.

## Conclusion

Herbicultural management offers a cost-effective and practical weed control strategy for irrigated green gram, especially in regions experiencing frequent rainfall and high soil moisture, where timely manual weeding is often impractical due to labor constraints and rising costs. Among the herbicidal treatments, pre-emergence (PE) application of Pendimethalin (1 kg a.i.  $\text{ha}^{-1}$  on 3 DAS), followed by early post-emergence (EPoE) application of Na Acifluorfen + Clodinafop propargyl (187.5 g a.i.  $\text{ha}^{-1}$  on 20 DAS), demonstrated superior weed suppression, higher green gram yield, better quality, and greater profitability. Although the initial phytotoxic effects of these ready-mix herbicides caused minor setbacks in crop growth, the plants recovered fully within 15 days after herbicide application (DAHA), resuming normal development. Hence, the integrated approach of PE Pendimethalin followed by EPoE Na Acifluorfen + Clodinafop propargyl emerges as a sustainable, economically viable, and ecologically safer weed management option for irrigated green gram. Moreover, effective herbicultural management reduces the weed seed bank by inhibiting weed seed germination, limiting vigor, and preventing seed production, thereby offering a long-term weed control advantage. This cost-effective strategy holds significant potential to expand green gram cultivation in weed-infested regions, providing higher monetary returns for marginal farmers in Tamil Nadu.

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