Field and Greenhouse Evaluation of Crop Aid Plus for Clubroot Management

Project Report

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Introduction

Clubroot of canola, caused by *Plasmodiophora brassicae* Wor., is a major disease that threatens canola (*Brassica napus* L.) production in western Canada (Strelkov and Hwang 2014). Preliminarily tests suggest that the fertilizer product Crop Aid Plus (CAP) may supress clubroot on canola and enhance the growth potential of the crop (Darren Fischer, personal communication, Crop Aid Nutrition). The objectives of this project were to (1) evaluate CAP for its efficacy against clubroot, and (2) determine the optimal timing of application to improve plant growth under the disease pressure.

Materials and Methods

Fertilizer product and plant materials:

The CAP liquid fertilizer product was provided by Crop Aid Nutrition, Saskatoon, SK. Following the product instructions, the fertilizer was diluted with distilled water (for greenhouse) or tap water (for field) at 250 mL CAP per 50L water for application. Two canola hybrids '45H31' (clubroot susceptible) and 'CS2000' (moderately resistant) were employed in this study.

Field trials:

The field trials took place at two sites, separated by approximately 100 m, within the Henwood clubroot nursery (53° 38’ 50” N, 113° 22’ 30” W; and 53° 38’ 48” N, 113° 22’ 46” W). Two canola hybrids ‘45H31’ (clubroot susceptible) and ‘CS2000’ (moderately resistant) were seeded in a split-plot design with four replicates at each site. Approximately 0.7 g seeds were planted in each of four rows in 6 × 1.5 m plots using a push-seeder. Tap water or the CAP solution was applied to the plots using a backpack sprayer. The treatments included an untreated control (UTC) and CAP at 250 mL/ac applied 1 week before seeding (1WB), 1 week after seeding (1WA), or 3 weeks after seeding (3WA). An additional treatment with CAP at 250 mL/ac was applied to all the plots except for the UTC at 4 weeks after seeding. Twenty plants from each plot were sampled for clubroot symptom evaluation 8 weeks after seeding. Plants were rated on a 0 (no symptoms at all) to 3 (severe root galling), and the individual ratings were used to calculate a disease severity index (DSI) for each experimental unit with the formula: $\text{DSI}\ (\%) = \frac{\sum (n \times 0) + (n \times 1) + (n \times 2) + (n \times 3)}{N \times 3} \times 100\%$, where n = plant number in each rating group and N = total plant number in an experimental unit (Horiuchi and Hori 1980; Strelkov et al. 2006). Plant height, weight of the aboveground biomass and gall weight were also recorded for each treatment. The seeds harvested from each plot were weighed and the yield calculated.

Greenhouse trials:

Clubroot-free field soil and Sunshine Mix (Sungro Horticulture, Seba Beach, AB) potting mixture were mixed in a 1:1 volume to volume ratio, and then inoculated with *P. brassicae* to produce final resting spore concentrations (SCs) of 1×10^5 or 1×10^7 spores/g soil mixture. The inoculated soil mixtures were used to fill plastic tubs (43 x

28 x 17.8 cm). The same canola hybrids ('45H31' and 'CS2000') were included as in the field trials. Treatments including UTC, 1WB, 1WA, 3WA at 250 mL/ac were arranged in a completely randomized design for each cultivar and SC. Diluted fertilizer solution or distilled water was applied from the top of the tubs using a spray bottle. After four weeks of seeding, an additional application of CAP at 250 mL/ac was also applied to all the tubs except for the UTC, which was treated with water. Two rows of 12 seeds were sown per tub at 2.5-cm seed intervals and with 10-cm row spacing. Each experiment was arranged in a completely randomized design with four replicates, where one tub represented one experimental unit. After 7 weeks, 10 plants were sampled from each tub and evaluated for clubroot severity as above. Individual plant height, aboveground plant biomass and clubroot gall weight were measured. Each experiment was repeated independently once on separate benches in the greenhouse.

Data analysis:

Data comparison to identify significant differences at $P < 0.05$ were subjected to Fisher's LSD test using the 'Agricolae' package of R 3.6.1 (de Mendiburu 2019; R Core Team 2019).

In June, July and August of 2020, abnormally high frequency and volume of precipitation occurred in the Edmonton area (Environment Canada, http://www.climate.weatheroffice.gc.ca). Two of four replicates in each site were flooded in the field, which resulted in unreliable data. Therefore, the other four un-flooded replicates from the two sites were pooled as one experiment in the data analysis.

Results

Field trials:

The CAP treatments significantly reduced DSI on both '45H31' and 'CS2000' (Figure 1). The treatments 1WA and 1WB had DSI scores of 29.5% and 27.2%, respectively on '45H31', which were not significantly different from each other, but were significantly lower than the UTC (42.5%) and the treatment 3WA (37.8%). Correspondingly, significantly lower clubroot gall weights were observed with these two treatments on '45H31' compared with UTC and 3WA (Table 1). On 'CS2000', the three CAP applications significantly reduced DSI to ~7.0% to 8.7% compared with the UTC (23.1%), but no significant differences were detected among the treatments (Figure 1). In contrast, all the treatments significantly increased plant biomass production and yield over the UTC on both cultivars (Figures 3 & 5). For '45H31', the application of CAP at 3WA resulted in the highest biomass among the treatments, but did not result in significantly higher yield. In the case of 'CS2000', biomass production for the 3WA treatment was not significantly different from the other treatments, but produced the highest yield.

Greenhouse trials:

Low inoculum level (1×10^5 SC)

A significant reduction in clubroot severity was observed in all the CAP treatments compared with the UTC for both canola cultivars at the 1×10^5 SC (Figure 2a). The treatment at 1WB reduced DSI by 31.3% on the susceptible '45H31' and by 25.7% on the moderately resistant 'CS2000'. The application 1WA also lowered the DSI by ~23% on both cultivars. No significant differences were observed on individual plant heights (Table 1). However, all of the CAP treatments significantly increased the individual

plant biomass, compared with the UTC, by 23.5% to 44.6% on '45H31' and 21.8% to 34.5% on 'CS2000' (Figure 4a). A significant reduction in clubroot gall weight per plant was observed on '45H31' but not on 'CS2000' (Table 2).

High inoculum level (1 × 10^7 SC)

At 1 × 10^7 SC, all the CAP treatments significantly reduced clubroot severity on both canola cultivars (Figure 2b). However, no significant differences were detected with respect to application timing. CAP treatments reduced DSI by less than 8% on the susceptible '45H31', and up to 16.4% on 'CS2000'. No significant differences were observed on individual plant heights for either cultivar, whereas 'CS2000' grew significantly taller than '45H31' (Table 1). The applications at 1WB and 3WA significantly increased individual plant biomass of '45H31', while biomass of 'CS2000' was significantly enhanced by 1WA and 3WA (Figure 4b). Significantly higher clubroot gall weight was observed on '45H31' with the treatment 3WA, compared with all other treatments (Table 2).

Discussion

In the greenhouse, the applications of CAP appeared able to reduce clubroot severity on canola as well as helping to maintain plant growth in the presence of clubroot. At the lower SC of 1 × 10^5 resting spores/g in soil, the treatment at 1WB reduced the DSI to < 50% of the untreated control on the susceptible '45H31', and to < 40% on the moderately resistant 'CS2000'. Although the 1WA application resulted in slightly higher average DSI than 1WB, there were no significant differences between them, indicating that both of these application timings may efficiently control clubroot under low disease pressure. In contrast, under a higher disease pressure of 1 × 10^7 SC, all the CAP treatments showed similar reductions in DSI on both cultivars, regardless of the application timings. However, with the DSIs of ~92% on the susceptible

cultivar and > 62% on the moderately resistant cultivar, the applications may not deliver enough clubroot control for the higher SC. While the 3WA application did not provide the highest disease reduction, it consistently resulted in significantly higher biomass than the UTC on both cultivars and under both SCs.

The CAP treatments also reduced clubroot severity under field conditions. However, clubroot disease levels under field conditions in this study were relatively low. The applications of 1WA and 1WB reduced DSI on both cultivars, but the treatment 3WA only significantly controlled the disease on the moderately resistant 'CS2000'. Therefore, the recommendation on spray timing would be 1WB or 1WA for clubroot control. The 3WA treatment increased yield by 14.6% and 23.0% and enhanced biomass by 72.8% and 34.9% respectively on '45H31' and 'CS2000'. Although it is not the optimal treatment for clubroot control, this application timing significantly enhanced canola growth. As CAP can be applied to the field multiple times during the growing season, an additional spray of the product at 3WA as well as the 1WA or 1WB application may result in better crop growth or yield.

The CAP product is a blend of lignin extract, kelp extract, and contains multiple micronutrients such as zinc, iron, manganese, and boron. It may not achieve complete disease control solely by one application. Nonetheless, compared with other chemical products for managing clubroot, a major advantage of CAP is that it is an environmentally friendly product originating from natural products and does not cause phytotoxicity (Hwang et al. 2011; Strelkov and Hwang 2014). It may be worthwhile to explore the use of CAP further as part of an integrated disease management system for canola production.

References

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Table 1. Average plant heights of the canola hybrids '45H31' and 'CS2000' in field and greenhouse trials at inoculum concentration of 1×10^5 and 1×10^7 resting spores/g soil. Numbers within a column followed the same letter are not significantly different at $P<0.05$.

Treatment*	Cultivar	Field	10^5	10^7
UTC	45H31	107.77a	68.10a	46.23b
	CS2000	111.10a	71.70a	70.55a
1WB	45H31	112.39a	73.88a	49.60b
	CS2000	114.88a	77.88a	75.75a
1WA	45H31	114.20a	73.46a	47.02b
	CS2000	112.03a	75.20a	75.10a
3WA	45H31	114.80a	79.13a	53.18b
	CS2000	108.18a	77.38a	74.26a

*UTC, untreated control; 1WB, CropAid (CAP) applied at 250 mL/ac 1 week before seeding; 1WA, CAP applied 1 week after seeding; 3WA, CAP applied 3 weeks after seeding.

Table 2. Gall weight per plant of the canola hybrids '45H31' and 'CS2000' in field and greenhouse trials at inoculum concentration of 1×10^5 and 1×10^7 resting spores/g soil. Numbers within a column followed the same letter are not significantly different at $P<0.05$.

Treatment*	Cultivar	Field	10^5	10^7
UTC	45H31	2.57a	3.25a	3.72b
	CS2000	1.05c	1.13c	1.39c
1WB	45H31	1.15bc	1.80b	4.41ab
	CS2000	0.48c	0.83c	1.31c
1WA	45H31	1.13c	1.80b	4.49b
	CS2000	0.51c	0.59c	1.03c
3WA	45H31	1.94ab	2.26b	5.30a
	CS2000	0.72c	0.84c	1.64c

*UTC, untreated control; 1WB, CropAid (CAP) applied at 250 mL/ac 1 week before seeding; 1WA, CAP applied 1 week after seeding; 3WA, CAP applied 3 weeks after seeding.

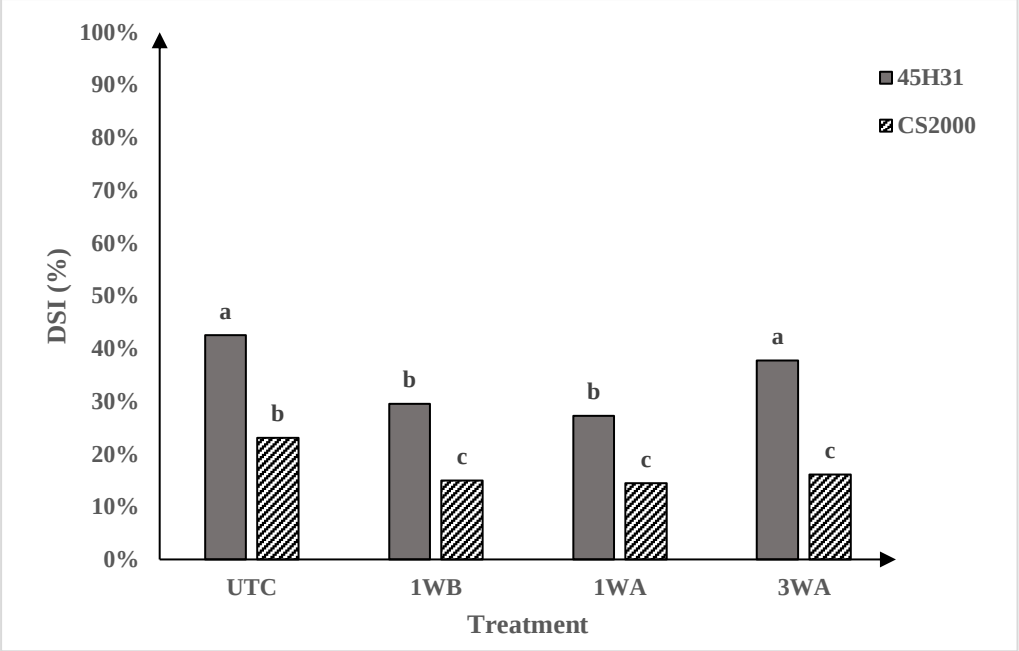


Figure 1. Clubroot disease severity index (DSI) on canola hybrids '45H31' and 'CS2000' under field conditions and various treatment regimes. UTC, untreated control; 1WB, CropAid (CAP) applied at 250 mL/ac 1 week before seeding; 1WA, CAP applied 1 week after seeding; 3WA, CAP applied 3 weeks after seeding. Bars capped with the same letter are not significantly different at $P<0.05$.

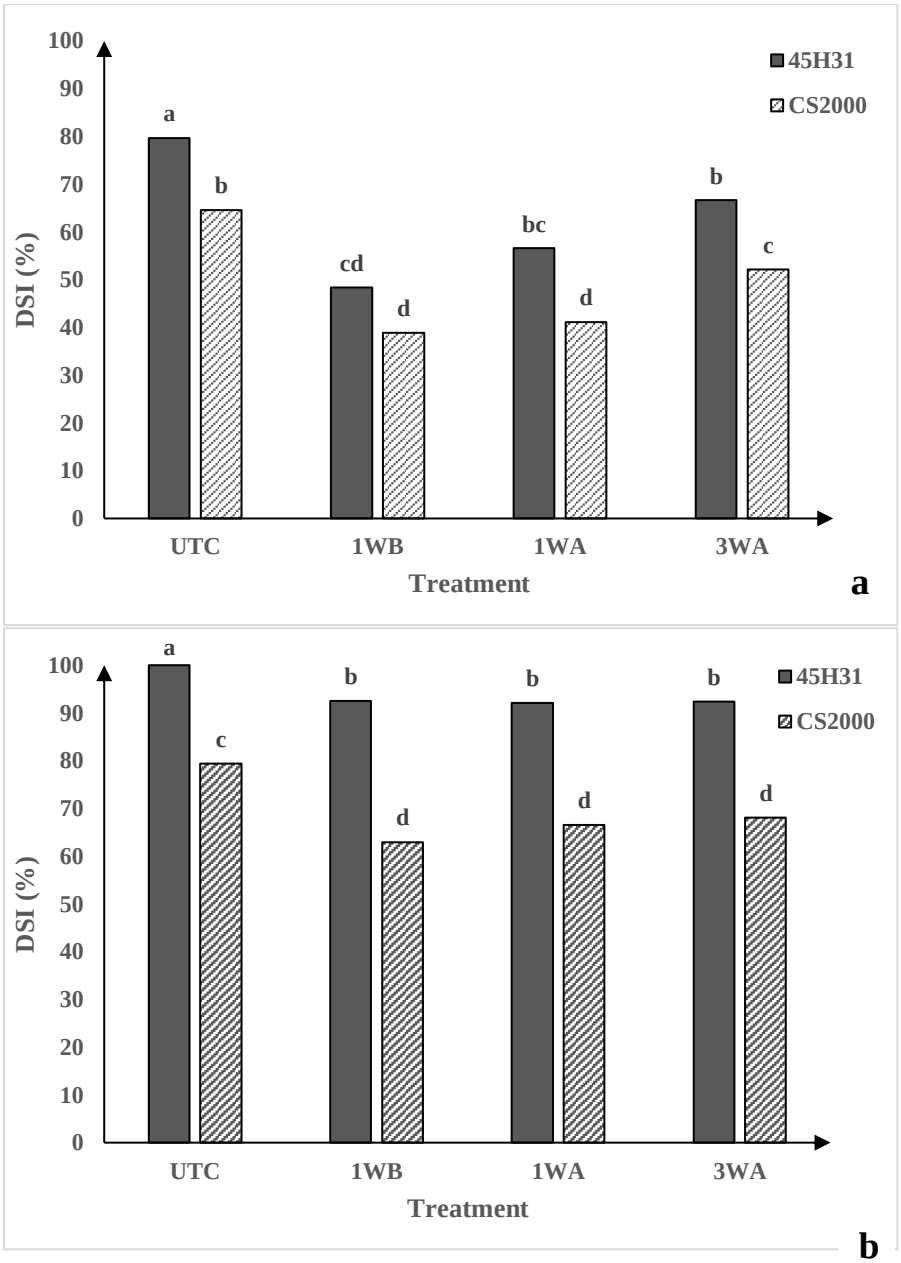


Figure 2. Clubroot disease severity index (DSI) on the canola hybrids '45H31' and 'CS2000' in the presence of 1×10^5 (a) or 1×10^7 (b) resting spores/g soil mix under greenhouse conditions and various treatment regimes. UTC, untreated control; 1WB, CropAid (CAP) applied at 250 mL/ac 1 week before seeding; 1WA, CAP applied 1 week after seeding; 3WA, CAP applied 3 weeks after seeding. Bars capped with the same letter are not significantly different at $P<0.05$.

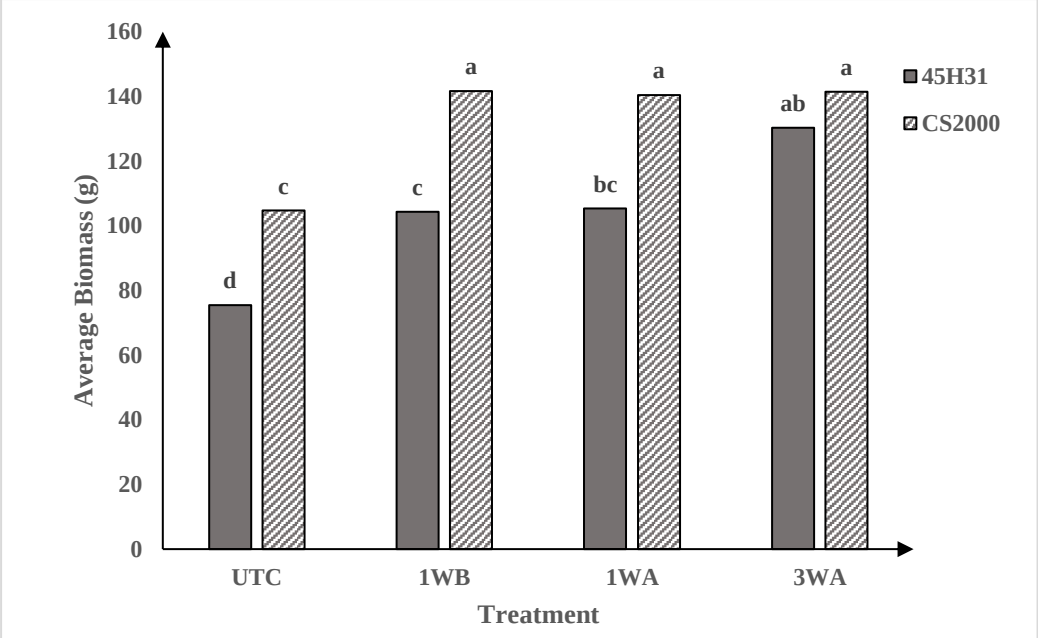
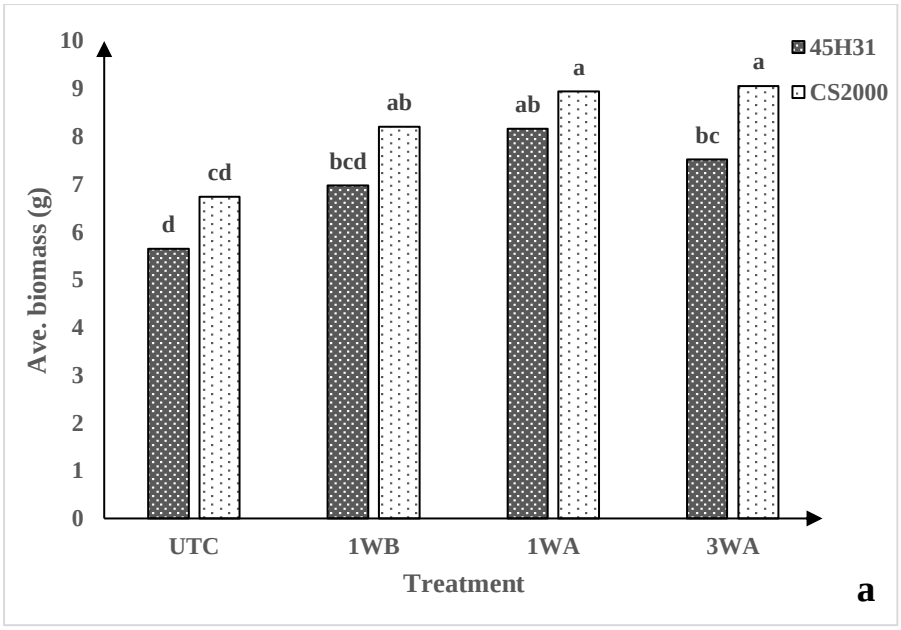


Figure 3. Average biomass per plant of the canola hybrids '45H31' and 'CS2000' under field conditions and various treatment regimes. UTC, untreated control; 1WB, CropAid (CAP) applied at 250 mL/ac 1 week before seeding; 1WA, CAP applied 1 week after seeding; 3WA, CAP applied 3 weeks after seeding. Bars capped with the same letter are not significantly different at $P<0.05$.



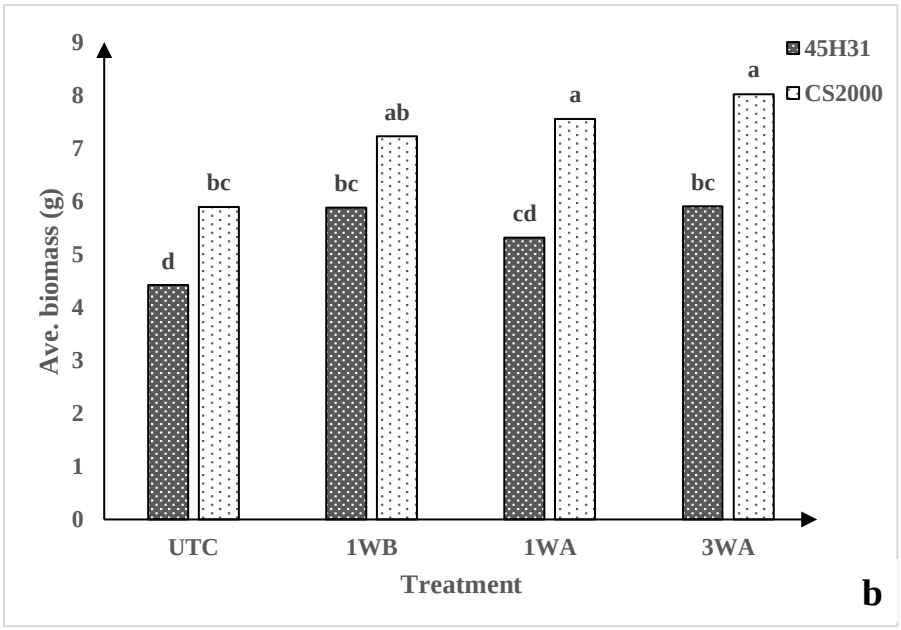


Figure 4. Average biomass per plant of the canola hybrids '45H31' and 'CS2000' in the presence of 1×10^5 (a) or 1×10^7 (b) resting spores/g soil mix under greenhouse conditions and various treatment regimes. UTC, untreated control; 1WB, CropAid (CAP) applied at 250 mL/ac 1 week before seeding; 1WA, CAP applied 1 week after seeding; 3WA, CAP applied 3 weeks after seeding. Bars capped with the same letter are not significantly different at $P<0.05$.

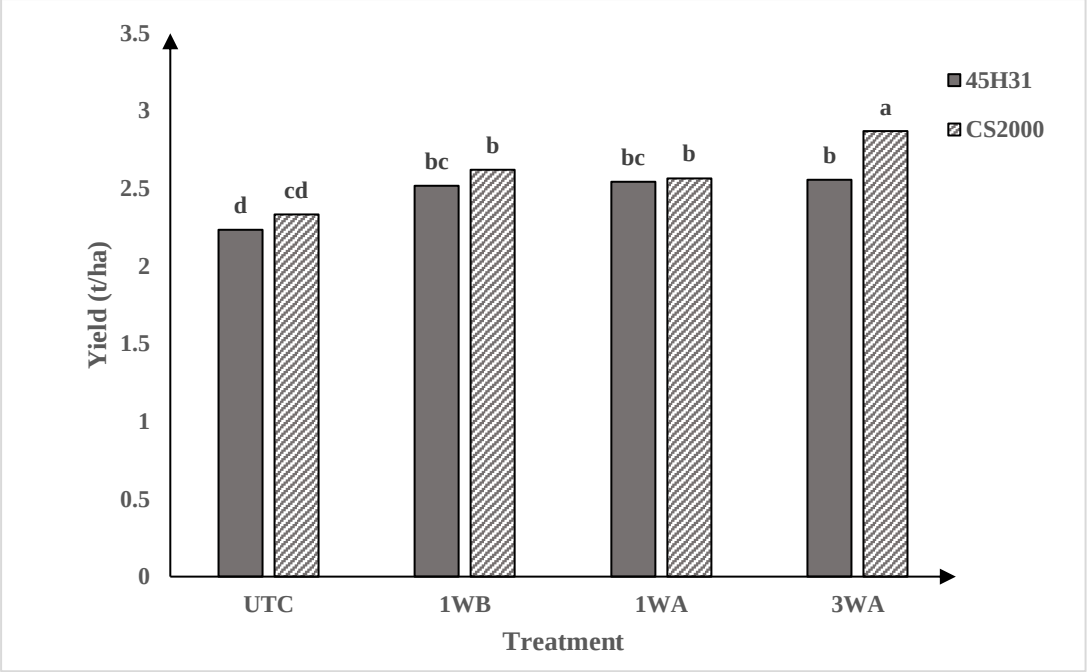


Figure 5. Yield of the canola hybrids '45H31' and 'CS2000' under field conditions and various treatment regimes. UTC, untreated control; 1WB, CropAid (CAP) applied at 250 mL/ac 1 week before seeding; 1WA, CAP applied 1 week after seeding; 3WA, CAP applied 3 weeks after seeding. Bars capped with the same letter are not significantly different at $P<0.05$.