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Boron Seed Treatment via Film Coating and Pelleting Mitigates Deficiency Symptoms and Boosts Curd Yield in Cauliflower Under Boron-Deficient Management

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Highlights

What are the main findings?

- Film coating and pelleting at 0.5 g B kg^{-1} seed maintained seed germination at 86.0% and 92.5%, meeting commercial standards (≥80%), while higher doses caused severe phytotoxicity.
- Despite suppressed laboratory germination at higher doses, seedling emergence in substrate remained high (≥94%), likely due to irrigation diluting localized boron toxicity.
- Under boron-deficient management, seed treatments reduced deficiency symptoms and increased commercial yield by up to 145.7%.

What are the implications of the main findings?

- Boron seed treatment serves as a low-cost preventive strategy to safeguard cauliflower yield against soil deficiency or fertilizer management failures.
- Film coating at 0.5 g B kg^{-1} seed offers the seed industry a viable method for micronutrient delivery while preserving legal germination standards.

Abstract

Boron (B) is essential for cell wall integrity and meristematic activity in cauliflower (*Brassica oleracea* var. *botrytis*). Still, the narrow range between deficiency and toxicity limits soil and foliar fertilization, and seed-applied B remains unexplored. This study establishes the conceptual novelty of boron seed treatment as a targeted preventive tool to safeguard cauliflower performance under boron-deficient soil conditions or management failures. This study evaluated a boron–polyol complex applied via film coating and pelleting to cauliflower seeds, assessing physiological quality, seedling development, and agronomic performance across five B doses (0.5–4.5 g B kg^{-1}) in the laboratory and nursery, and two doses (0.5 and 4.5 g B kg^{-1}) under two fertilization managements in an 8 L pot greenhouse experiment. Boron sharply reduced laboratory germination as the dose increased, falling below the commercial minimum (80%) beyond 0.5 g B kg^{-1}, while seedling emergence in substrate remained largely unaffected (≥94% in most treatments). In the greenhouse trial, without soil or foliar boron, untreated seeds resulted in 62.5% of plants with deficiency symptoms and the lowest commercial yield (3.5 t ha^{-1}); boron seed treatment reduced

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deficiency incidence and increased commercial yield in three of the four seed treatments, with gains of up to 145.7% relative to untreated seeds. Film coating at 0.5 g B kg^{-1} seed is recommended as the only dose preserving seed physiological quality for commercialization while increasing commercial yield by 125.7%, showing promise as a low-cost strategy. Conceptually, this work proves that B seed treatment acts as a vital technological safety net in B-restricted areas, drastically reducing deficiency symptoms and protecting yield when standard soil or foliar supply is inadequate.

Keywords: *Brassica oleracea* var. *botrytis*; emergence; germination; micronutrient; yield

1. Introduction

The Brassicaceae family comprises a vast array of economically important vegetable species used for fresh or industrialized consumption, forage, medicinal, and ornamental purposes, among which cauliflower (*Brassica oleracea* var. *botrytis*) is a prominent herbaceous crop [1]. Cauliflower exhibits a high nutrient demand within a short growth cycle, requiring adequate availability of micronutrients—particularly boron and molybdenum—to ensure healthy and productive plants [2]. Micronutrient supply has traditionally been conducted via foliar applications, pre-seeding immersion in nutrient solutions, or direct soil application at sowing [3]. However, direct immersion of seeds in nutrient solutions can severely impair seed physiological quality [4]. To minimize direct chemical damage, seed enhancement technologies such as film coating and pelleting have been adopted.

Film coating was developed primarily to facilitate sowing operations and protect seeds in the soil under adverse conditions without altering their original shape or size [5], utilizing a polymer, plastic, and colorant mixture that can incorporate crop protection agents and nutrients [5]. Furthermore, nutrient delivery through seed coating strategically localizes mineral elements at the seed surface, enhancing nutrient availability during early seedling establishment and representing a precise, increasingly adopted strategy for field crops [6,7]. Conversely, pelleting modifies seed shape, size, and mass to facilitate manual or mechanized distribution, particularly for vegetable seeds with irregular surfaces, trichomes, thorns, or small size [8]. These seeds are coated with filler materials and a cementing agent (binder or adhesive). The filler should be dry, inert, and of fine granulometry, whereas the binder must ensure adhesion among coating components, be readily water-soluble, and exhibit no phytotoxic effects [9]. Along with these materials, other substances, such as nutrients, can also be incorporated. Because they form several layers of inert material, they can allow additives, such as nutrients, to stay away from seeds and can reduce the damage these additive substances can cause. It is important that the coating does not offer resistance to the protrusion of the root and the structure that will form the aerial part of the plant, and it should allow the passage of water and oxygen for the embryo to begin to develop naturally [10].

Boron is an important micronutrient that can be applied through seed treatment, whose best-established function is maintaining cell wall structure through borate cross-linking of pectic polysaccharides, particularly rhamnogalacturonan II in the middle lamella [11,12]. In the case of boron omission, root stretching may halt due to the lack of division of meristematic cells, since this micronutrient is essential for normal cell division and cell wall integrity. It is essential for cell growth, especially in young tissues such as buds, apical meristems, and reproductive organs, which have not yet matured [13,14]. Under boron deficiency, cauliflower develops brown heart, involving dark punctuations in the inflorescence, a hollow and darkened medulla, and corticosis necrosis in the main ribs of the leaf [15,16]. Soil applications of boron sources (boric acid and tetrahydrate sodium

octaborate) in the cauliflower 'Piracicaba Precoce' demonstrated that boric acid provides superior plant fresh and dry mass accumulate, regardless of dose [17].

Also for cauliflower, Trani et al. [18] recommend top-dressing fertilization with boron, plus foliar spraying with boric acid and ammonium molybdate three times during the cycle. They also recommend applying 40 t ha^{-1} of organic compost, which can be a source of nutrients, including boron. However, the farmer does not always have organic compost in such quantity. Besides this, the application of boron in small quantities is not always done because this micronutrient has a very narrow range between the minimum necessary and the maximum allowed to not cause toxicity [19], and, if applied in excess or unevenly in the field, it can cause other damage.

In view of the importance of boron for the proper development of cauliflower and the advantages of seed treatment as a targeted technological safety net in boron-restricted areas or management failure scenarios, this study aimed to assess the effect of boron treatment in cauliflower seeds, through film coating and pelleting, on the physiological quality of seeds and on the development and production of cauliflower plants with and without boron fertilization. It was hypothesized that a low boron dose applied to the seed would act as a preventive tool to reduce deficiency symptoms and increase commercial yield under boron-deficient field management, without compromising seed germination below the commercial threshold.

2. Materials and Methods

2.1. Experimental Location and Growth Conditions

Seed treatments were performed at Sakata Seed Sudamerica®, located in Bragança Paulista, Brazil. Seed germination and vigor tests were conducted at the Vegetable Seed Laboratory of the Department of Crop Production, whereas seedling production was carried out in the nursery of the Department of Forestry Science, Soil and Environment. Both facilities belong to the School of Agricultural Sciences (FCA-UNESP), Botucatu Campus. The nursery greenhouse was an arch-type structure with a ceiling height of 3.0 m, covered with light-diffusing polyethylene film and lateral sides fitted with 50% monofilament shade netting. Seedling trays were placed on metal benches positioned 50 cm above the ground and irrigated by inverted microsprinklers delivering an average water depth of 5 L m^{-2}.

Seeds of a summer cauliflower hybrid produced in Limarí Province, Chile, were supplied by Sakata Seed Sudamerica (the hybrid name is withheld at the supplier's request for commercial confidentiality). The boron source was a boron–polyol complex donated by Núcleo de Pesquisas Aplicadas Ltd.a. (Jaboticabal, SP, Brazil). The boron source was supplied as a true solution containing 6.5% (w/w) water-soluble B, equivalent to 85.8 g L^{-1} (w/v), with a density of 1.32 g mL^{-1}. The formulation contained boron in the form of boron esters complexed with polyols.

Two independent experiments were conducted: one to evaluate seed physiological quality and seedling development under laboratory and nursery conditions, and another to assess the agronomic performance of cauliflower grown in pots under greenhouse conditions.

For the laboratory and nursery experiments, thirteen treatments were evaluated in a 2 × 5 + 3 factorial arrangement, consisting of two seed treatment technologies (film coating and pelleting), five B doses (0.5, 1.5, 2.5, 3.5, and 4.5 g B kg^{-1} seed), and three control treatments. The control treatments consisted of each seed treatment technology without nutrient application, corresponding to 0 g B kg^{-1} seed, and untreated seeds. Four replications were used, with a completely randomized design for the laboratory tests and a randomized block design for the nursery tests.

For the greenhouse experiment, ten treatments were evaluated, comprising a 2 × 2 × 2 factorial arrangement plus two additional controls. The factorial arrangement consisted

of two seed treatment technologies (film coating and pelleting), two boron doses (0.5 and 4.5 g B kg^{-1} seed), and two fertilization managements: the recommended fertilization for cauliflower production according to Trani et al. [18] and fertilization without boron. The two additional controls consisted of untreated seeds under each fertilization management. The experiment was arranged in a randomized complete block design with six replicates. Each experimental plot consisted of four plants, with each plant grown in a separate 8 L pot. The four pots constituted a single experimental unit for statistical analysis. The two boron doses used in the greenhouse experiment (0.5 and 4.5 g B kg^{-1} seed) corresponded to the lowest and highest doses evaluated in the preceding laboratory and nursery experiments. These doses were selected to contrast the dose that maintained commercial seed germination with the dose that produced the strongest numerical response while still resulting in adequate seedling emergence.

Two fertilization managements were evaluated. Management 1 consisted of the application of castor bean cake at 4 t ha^{-1}, nitrogen fertilization with urea at 60 kg N ha^{-1}, phosphorus fertilization with single superphosphate at 520 kg P_2O_5 ha^{-1}, potassium fertilization with potassium chloride at 180 kg K_2O ha^{-1}, and boron fertilization with boric acid at 2.7 kg B ha^{-1}. Foliar fertilization with B and Mo was also applied at 15, 35, and 55 days after transplanting (DAT), using a solution containing 1 g L^{-1} boric acid and 1 g L^{-1} sodium molybdate. Management 2 consisted of no organic fertilizer, basal NPK fertilization without boron, and foliar fertilization with only Mo, using a solution containing 1 g L^{-1} sodium molybdate applied at 15, 35, and 55 DAT.

The chemical composition of the castor bean cake was as follows: N = 4.34%; P_2O_5 = 1.22%; K_2O = 1.09%; Ca = 1.12%; Mg = 0.60%; S = 0.32%; Na = 685 mg kg^{-1}; B = 36 mg kg^{-1}; Cu = 19 mg kg^{-1}; Fe = 2870 mg kg^{-1}; Mn = 102 mg kg^{-1}; Zn = 96 mg kg^{-1}; C/N ratio = 10/1; humidity = 7% and pH = 6.3.

Management 1 was designed to represent the complete standard commercial fertilization practice for cauliflower in the region (incorporating organic amendment, NPK, and complete B supply). Management 2 was established as a B-deficient baseline (omitting both inorganic B sources and organic fertilizer) to evaluate whether seed-applied boron could compensate for B omission. Organic fertilizer was intentionally omitted from Management 2 because castor bean cake naturally contains boron (36 mg B kg^{-1}), which would otherwise introduce a substantial organic background supply of B and prevent the establishment of a true B-limiting control.

2.2. Seed Treatment Technologies and Boron Doses

Seed treatments were performed at Sakata Seed Sudamerica®. Seeds were divided into lots according to the experimental treatments, and treatment solutions containing the respective boron doses were prepared and uniformly mixed. Film coating was performed using the company's prototype film-coating equipment, whereas pelleting was carried out in a rotary pelleting machine. For pelleted seeds, boron doses were calculated based on the mass of naked seeds before pelleting and incorporated into the pellet during the pelleting process. Following film coating or pelleting, the seeds were dried at 35 °C for 1 h. Seed moisture content was monitored during drying and adjusted to 5–6% for film-coated seeds and 2.54% for pelleted seeds before storage. Finally, the treated seeds were stored in a dry chamber at 20 °C and 40% relative humidity until use.

Seedlings were produced in the nursery of the Department of Forestry Science, Soil and Environment. After transplanting, the plants were grown at the Experimental Farm of São Manuel. Both facilities belong to the School of Agricultural Sciences (FCA-UNESP), Botucatu Campus.

Seeds were sown in 162-cell trays filled with Carolina Soil® substrate, and the seedlings were transplanted 25 days after sowing (DAS). After transplanting, the plants were grown individually in 8 L pots in a greenhouse covered with high-density polyethylene film (150 µm) and fitted with anti-aphid screens on the sides.

The soil used was classified as a sandy Dystrophic Red Oxisol. The soil chemical properties before the experiment were as follows: pH ($CaCl_2$) = 4.3; organic matter = 2 g dm^{-3}; P_{resin} = 2 mg dm^{-3}; H + Al = 27 mmolc dm^{-3}; K = 0.3 mmolc dm^{-3}; Ca = 11 mmolc dm^{-3}; Mg = 7 mmolc dm^{-3}; B = 0.21 mg dm^{-3}; Mo = 1.0 mg dm^{-3}; sum of bases = 18 mmolc dm^{-3}; CEC = 45 mmolc dm^{-3}; and base saturation (V) = 40%. Lime was applied to increase the base saturation to 80%, as recommended for cauliflower [18]. Organic and inorganic fertilizers were incorporated into the soil according to the respective treatments before the soil was placed into the 8 L pots. The pots were spaced 0.5 m apart within rows and 1.0 m between rows. Drip irrigation was used to maintain soil moisture close to field capacity. Top-dressing fertilization was applied via fertigation on eight occasions at 7-day intervals, using an NPK solution according to Trani et al. [18], starting at 15 days after transplantation (DAT).

2.3. Characteristics Evaluated

Seed physiological quality was evaluated based on thousand-seed or thousand-pellet mass, pellet hardness, first germination count, germination percentage, and germination speed index (GSI). Thousand-seed and thousand-pellet mass were determined according to the Rules for Seed Testing [20]. Pellet hardness was measured using a pellet hardness tester (durometer) and is expressed in kgf. Germination was evaluated using four replicates of 50 seeds placed in Gerbox containers containing moistened blotter paper and incubated at 20 °C (Figure 1A,B). Normal seedlings were counted 10 days after sowing according to the Rules for Seed Testing [20]. The first germination count was performed five days after sowing, and the GSI was calculated according to Maguire [21].

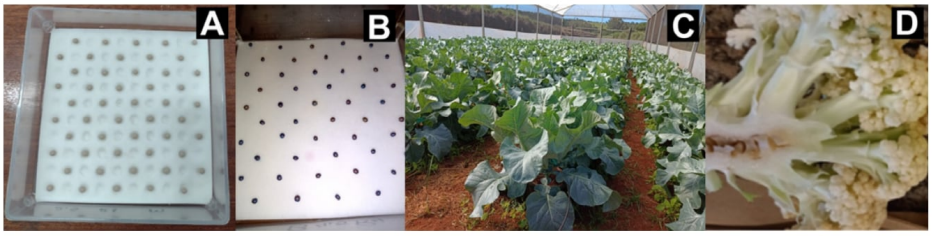


Figure 1. (**A**) Pelleted seeds placed in Gerbox containers; (**B**) film-coated seeds placed in Gerbox containers; (**C**) greenhouse development of the plants in the pots; (**D**) plants showing boron deficiency symptoms (brown rot).

Seedling quality was assessed based on emergence percentage, emergence speed index (ESI), seedling height, and fresh and dry shoot and root biomass. Emergence was evaluated using four replicates of 25 seeds sown in polypropylene trays filled with Carolina Soil® substrate. Normal seedlings were counted 10 days after sowing. The ESI was calculated according to Maguire [21]. Seedling height was measured in ten randomly selected seedlings per replicate at 25 days after sowing. Fresh shoot and root biomass were determined immediately after sampling, whereas dry shoot and root biomass were determined after drying the material in a forced-air oven at 80 °C for 72 h.

For the greenhouse experiment (Figure 1C), the following variables were evaluated: boron concentration in seedlings at transplanting, crop cycle duration, number of leaves, fresh and dry biomass of leaves, stems, and inflorescences, inflorescence diameter, incidence of plants showing boron deficiency symptoms (brown rot) (Figure 1D), total yield, and

commercial yield. Fresh biomass was determined at harvest, whereas dry biomass was obtained after drying the plant material in a forced-air oven at 65 °C until constant weight. Inflorescence diameter was measured at harvest. Total yield was estimated by multiplying the mean fresh inflorescence mass by a plant population of 20,000 plants ha^{-1}. Commercial yield was calculated based only on marketable inflorescences free of visual defects and brown rot symptoms.

At transplanting, the boron concentration in seedlings was determined by sampling ten randomly selected seedlings per replicate. The shoot tissue from the ten seedlings within each replicate was pooled to form a single composite sample per biological replicate (n = 4 independent replicates per treatment). Samples were dried in a forced-air oven at 65 °C until constant weight, ground, and analyzed for boron concentration using the dry-ashing procedure described by Malavolta et al. [19].

2.4. Statistical Analysis

Data from the laboratory and nursery experiments were subjected to analysis of variance (ANOVA). When significant effects were detected, means for seed treatment technologies were compared using Tukey's test ($p < 0.05$), and regression analyses were performed for boron dose using Sisvar software (version 5.0). Data from the greenhouse experiment were also subjected to ANOVA, and means for seed treatment technologies, boron doses, and fertilization managements were compared using Tukey's test ($p < 0.05$) in Sisvar software.

In both experiments, treatment means were also compared with the respective control treatments using Dunnett's test ($p < 0.05$). For the laboratory and nursery experiments, treatments were compared with untreated seeds and with the corresponding seed treatment technology without boron application. For the greenhouse experiment, treatments were compared with the corresponding controls consisting of untreated seeds under each fertilization management. Dunnett's tests were performed using Assistat software (version 7.7) for comparisons.

3. Results and Discussion

3.1. Seed Quality and Seedling Development

3.1.1. Physical Properties of Coated Seeds

The comparison of the thousand-seed mass between the technologies shows a large increase (almost six times) in the weight of the pelleted seeds in relation to that of the film-coated ones (Table 1), which was already expected, as an increase in size and mass is the main objective of pelleting [5]. This characteristic made it easier to handle the seeds and to sow them.

Table 1. Averages of a thousand-seed mass (TSM), first germination count (FGC), germination (GERM) and germination speed index (GSI) of cauliflower seeds treated with film coating and pelleting.

	TSM	FGC	GERM	GSI
	g	%		
Film coating	5.43 b	5.70 a	29.00 a	1.97 a
Pelleting	29.45 a	0.30 a	23.80 b	1.58 a

Averages followed by the same letter in the columns do not differ by the Tukey test ($p < 0.05$).

For the film-coated seeds, a linear increase was observed in the mass of a thousand seeds the higher the dose of B (Figure 2A). This can be explained by the viscosity of the B source product (boron–polyol), which may have contributed to the greater adherence of the product used to film coat the seeds as higher doses of the product were used. In the

case of pelleting, there was a tendency for the values to decrease and then remain stable as the doses tested increased. In grain sorghum, coating seeds with boron at doses of 2.5 to 12.5 g B kg^{-1} seed increased seed size [22].

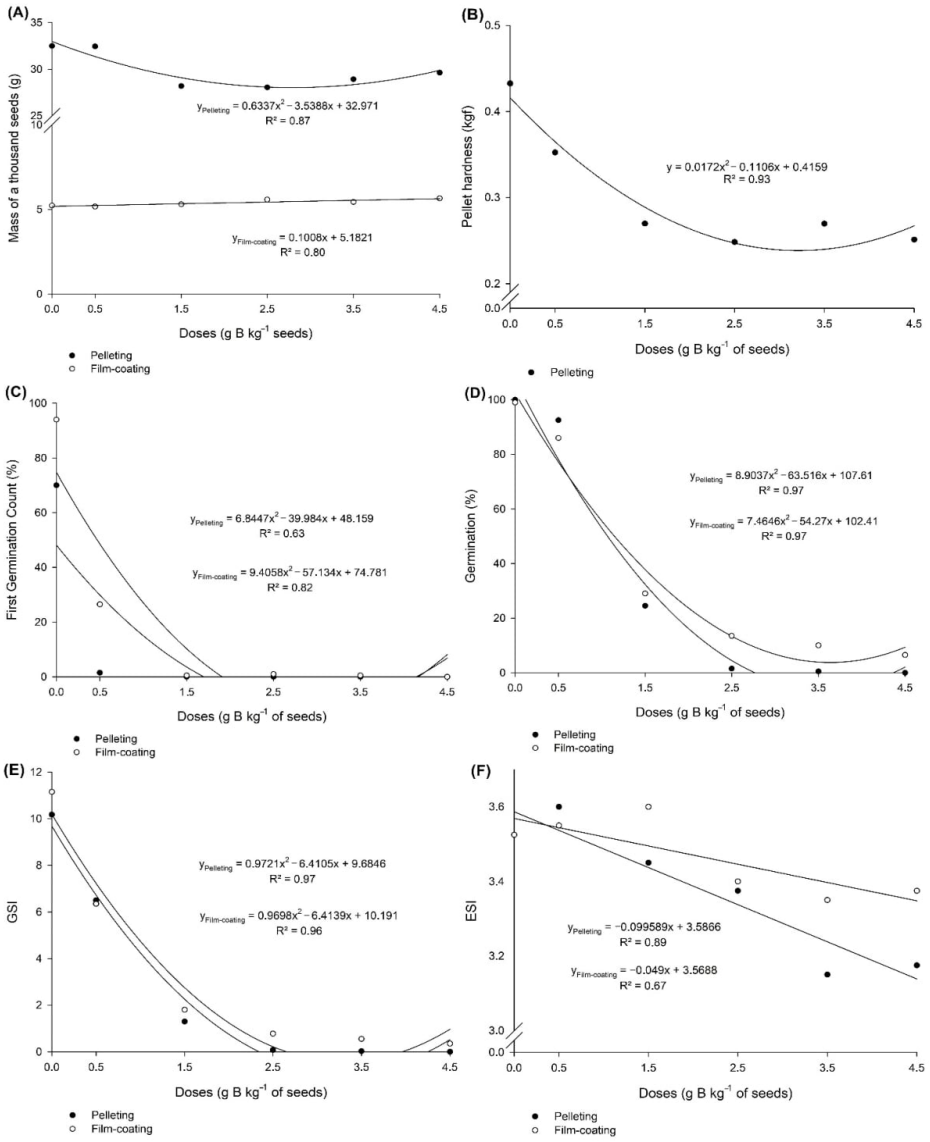


Figure 2. (**A**) Mass of a thousand seeds; (**B**) pellet hardness; (**C**) first germination count; (**D**) germination; (**E**) germination speed index; (**F**) emergence speed index of film coated and pelletized cauliflower seeds according to boron doses.

Comparing the boron treatments with the standard technologies, that is, without the application of this nutrient, the mass of a thousand seeds was similar for film coating with the two lowest doses and for pelleting with only the lowest dose of boron (Table 2). Compared to the control treatment, that is, untreated seeds, the mass was similar to that of standard film coating and film coating with boron at the two lowest doses, and lower than all the pelleted seed treatments. As discussed above, with pelleting, there is a large increase in the size and mass of the seeds, which is characteristic of this treatment, while with film coating, the increase in the mass of a thousand seeds is due to the viscosity of the product used; the higher the dose, the greater the adherence of the polymer used in the coating, increasing the mass of the seeds, resulting in greater mass compared to the control only at the higher doses.

Table 2. Comparison of boron film coating and pelleting of cauliflower seeds with film-coated and pelletized seeds without nutrient (boron) and with the control (untreated seeds) for the characteristics of thousand-seed mass (TSM), pellet hardness (HARD), first germination count (FGC), germination (GERM), germination speed index (GSI), emergence (EMERG), emergence speed index (ESI), seedling shoot fresh matter mass (SFM), seedling root fresh matter mass (RFM), seedling shoot dry matter mass (SDM), seedling root dry matter mass (RDM) and seedling height (HEIG).

Treatment	Dose	TSM	HARD	FGC	GERM	GSI	EMERG	ESI	SFM	RFM	SDM	RDM	HEIG
	g B kg^{-1} Seed	g	kgf	%			%			mg			cm
Film B	0.5	5.16 [a]*	-	26.5	86.0 [a]*	6.35	99.0 [a]*	3.54 [a]*	448.21 [a]*	50.31 [a]*	52.04 [a]*	10.99 [a]*	7.63 [a]*
Film B	1.5	5.30 [a]*	-	0.5	29.0	1.78	100.0 [a]*	3.57 [a]*	451.50 [a]*	47.76 [a]*	49.94 [a]*	9.02 [a]*	7.51 [a]*
Film B	2.5	5.58	-	1.0	13.5	0.80	96.0 [a]*	3.41 [a]*	455.53 [a]*	58.74 [a]*	50.67 [a]*	9.54 [a]*	7.59 [a]*
Film B	3.5	5.44	-	0.5	10.0	0.57	94.0 [a]*	3.35 [a]*	458.39 [a]*	65.67 [a]*	54.28 [a]*	9.42 [a]*	7.63 [a]*
Film B	4.5	5.65	-	0.0	6.5	0.36	96.0 [a]*	3.41 [a]*	428.01 [a]*	56.71 [a]*	49.52 [a]*	9.32 [a]*	7.41 [a]*
Film without nutrient	-	5.22 [a]*	-	94.0 [a]*	99.0 [a]*	11.13 [a]*	98.0 [a]*	3.50 [a]*	435.71 [a]*	47.28 [a]*	62.79 [a]*	11.60 [a]*	7.41 [a]*
Pellet B	0.5	32.44 [b]	0.35 [b]	1.50	92.50 [b]*	6.48	100.0 [b]*	3.57 [b]*	407.83 [b]*	74.33 [b]*	45.51 [b]*	8.88 [b]*	6.50 [b]*
Pellet B	1.5	28.19	0.27	0.0	24.50	1.30	97.0 [b]*	3.45 [b]*	416.94 [b]*	61.67 [b]*	44.18 [b]*	7.88 [b]	6.81 [b]*
Pellet B	2.5	28.05	0.25	0.0	1.50	0.09	98.0 [b]*	3.38 [b]*	378.10 [b]*	58.83 [b]*	39.82 [b]*	8.02 [b]*	6.93 [b]*
Pellet B	3.5	28.92	0.27	0.0	0.50	0.03	96.0 [b]*	3.15	348.47 [b]*	37.84 [b]*	35.86 [b]*	6.80	6.40 [b]*
Pellet B	4.5	29.63	0.25	0.0	0.00	0.00	98.0 [b]*	3.18[b]	395.14 [b]*	56.50 [b]*	39.00 [b]*	6.90	6.76 [b]*
Pellet without nutrient	-	32.49 [b]	0.43 [b]	70.0 [b]*	100.0 [b]*	10.18 [b]*	99.0 [b]*	3.53 [b]*	390.41 [b]*	71.82 [b]*	56.63 [b]*	12.56 [b]*	6.64 [b]*
Untreated seed	-	4.93 *	-	81.5 *	98.5 *	10.24 *	100.0 *	3.57 *	349.60 *	49.31 *	46.85 *	16.26 *	6.55 *

Film = Film coating. Pellet = Pelleting. [a] Means statistically equal to the film coating without nutrients by Dunnett's test at 5% probability. [b] Means statistically equal to the pelleting without nutrients by Dunnett's test at 5% probability. * Means statistically equal to the control (untreated seed) by Dunnett's test at 5% probability.

Film coating without nutrients resulted in a 5.9% increase in the mass of a thousand seeds compared to untreated seeds, while the highest dose of boron resulted in a 14.6% increase. On the other hand, comparing the film-coated seeds with the highest dose of boron with the film-coated seeds without nutrients, the increase was 8.2% (Table 2). Although significant, these differences are not large enough to facilitate sowing, as is the case with pelleting. However, simple film coating, despite not resulting in large increases in seed size, has advantages in that it makes it easier to see the seeds (due to their color) and makes it easier for the seeds to flow during sowing [23].

Pelleting technology also allows analysis of the pellet hardness characteristic. In this case, there was a reduction in hardness up to a dose of approximately 2.5 g B kg^{-1} seed, stabilizing thereafter (Figure 2B). Only the lowest dose (0.5 g B kg^{-1} seed) did not show pellets with lower hardness than standard pelleting, without nutrients (Table 2). Hölbig et al. [24] described that pellets must have the physical properties required to prevent breakage during transportation, handling and sowing. However, when moistened after sowing, they should disintegrate easily so that the pellet does not constitute a physical barrier that prevents or hinders germination. Therefore, it is important to balance the strength of the pellets so that they protect the seeds during handling and transportation, without impairing water absorption.

The pellet must be tough enough to protect the seeds, but not so hard that it hinders the development of the seedlings. Thus, only the lowest dose (0.5 g B kg^{-1} seed) did not reduce the hardness of the pellet compared to the standard pelleting used by the company (Table 2). However, even in the treatments with the lowest hardness values, direct visual observation during all seed handling, counting, and sowing stages confirmed that the pellets were still hard enough not to crumble and, thus, protect the seeds.

3.1.2. Germination and Seed Vigor

The boron treatment showed low values for all the characteristics related to germination and seed vigor measured in a controlled environment (BOD incubator) for both treatment technologies. For germination, pelleting performed worse than film coating (Table 1). Pelleting can hinder germination, probably due to the physical barrier imposed by the pellet on the protrusion of the radicle [23,25], even more so when the seed is placed on blotting paper, without soaking occurring as quickly as with untreated seeds.

The film coating and pelleting of cauliflower seeds with the addition of B considerably reduced the first germination count, germination and GSI of the seeds as the dose increased (Figure 2C–E). Pessoa, Luchese and Luchese [26] found that corn seeds treated with boric acid diluted in distilled water at doses equal to or greater than 0.20 g kg^{-1} reduced and delayed germination, with germination being zero at a dose of 4 g B kg^{-1} seed. In the present study, germination values were lower than 2.0% with pelleting, starting at a dose of 2.5 g B kg^{-1} seed, and being zero at 4.5 g B kg^{-1} seed.

For all doses of boron, the values obtained for the first germination count and GSI were lower than those for the standard treatments (film coating and pelleting without nutrients) and control (untreated seeds), while for germination, only the lowest dose (0.5 B kg^{-1} seed) did not differ from these treatments (Table 2). Verma and Mehta [27] found that pelleting chili seeds with boric acid did not harm seed germination. Rosal et al. [28], on the other hand, when pelleting corn seeds with increasing doses of boron (0, 0.05, 0.10, 0.15 and 0.20 g B kg^{-1} seed), observed that seeds treated with boron doses equal to or greater than 0.15 g B kg^{-1} seed showed reduced germination. In this study, cauliflower seeds proved to be sensitive to boron treatment, with a great loss of physiological quality (germination and vigor).

It is not uncommon to find reports in the literature about the small limit between the adequate and toxic doses for this micronutrient [13,14,29,30]. At high concentrations, boron disrupts antioxidant defense systems and impairs membrane integrity, contributing to embryo damage [30,31]. In addition, the viscosity of the product may have contributed to the decrease in seed germination and vigor by hindering the absorption of water and oxygen, which is essential for respiration. An additional, non-mutually exclusive possibility is that the polyol carrier itself exerted a dose-dependent osmotic effect on imbibition, independent of boron toxicity per se; the present experimental design, which did not include a polyol-only treatment without boron, does not allow these two effects to be distinguished.

Boron used in the form of boron esters with polyols is extremely viscous, not very fluid and has little ability to dissolve in the final solution used to treat the seeds (Figure 3A,B). Throughout the process, it was possible to notice that at the end of the film coating process, the treatment left traces of the product (Figure 3C) and drying took longer (Figure 3D). These properties intensified as the dose increased. Although in the pelleting technique there were no visual differences between the pellets in the treatments, there is a chance that the product, being more viscous, is more difficult to mix with the other components of the treatment and/or is more adherent to the seeds. Silva, Santos and Nascimento [32] state that solutions with high viscosity hinder gas exchange between the seed and the environment outside the pellet, as well as affect the drainage of water retained in the pellet pores, and can damage or delay germination. It is also important to note that the use of solid boron sources, such as boric acid, was physically unfeasible for seed treatment because achieving the target experimental doses would require an excessively high product volume that could not be uniformly diluted or physically adhered to small cauliflower seeds. Consequently, the liquid formulation was the only viable option, where its higher fluidity and lower required volume served as a key strategy to ensure uniform coating while mitigating deleterious effects on germination and vigor.

The boron–polyol complex used here behaves differently from inorganic boron sources such as boric acid. Polyol complexation reduces boron ion activity and modifies transport kinetics [33,34], which may partly explain why the phytotoxic effects observed in laboratory germination were not reproduced in seedling emergence under substrate conditions. Similar polyol-complexed boron sources have been shown to perform comparably to conventional sources in agronomic efficiency [35].

Unlike most micronutrients, whose primary roles are associated with enzymatic activation, boron performs a predominantly structural function in plants by forming borate diester bridges that cross-link rhamnogalacturonan II (RG-II) molecules within the primary cell wall, which determines wall porosity, rigidity, and the capacity for cell expansion [11,12]. This unique role explains why even relatively small deviations in boron availability may rapidly shift from nutritional sufficiency to toxicity. Borate bridges linking RG-II molecules are dynamically formed and dissociated within the cell wall rather than representing permanent structures, suggesting that transient localized excesses of boron during seed imbibition may interfere with cell wall remodeling during radicle protrusion in rapidly growing embryonic tissues [11,29,36].

Because the boron–polyol product was applied directly onto the seed coat, the radicle and embryonic axis were exposed to a transiently high, highly localized boron concentration during the first hours of imbibition, precisely when cell wall plasticity in the meristem is most critical, in line with the toxicity responses reported for mustard and rice seedlings exposed to excess boron [31,37].

Because a polyol-only treatment without boron was not included, carrier-induced osmotic or physical effects cannot be strictly separated from boron ion toxicity.

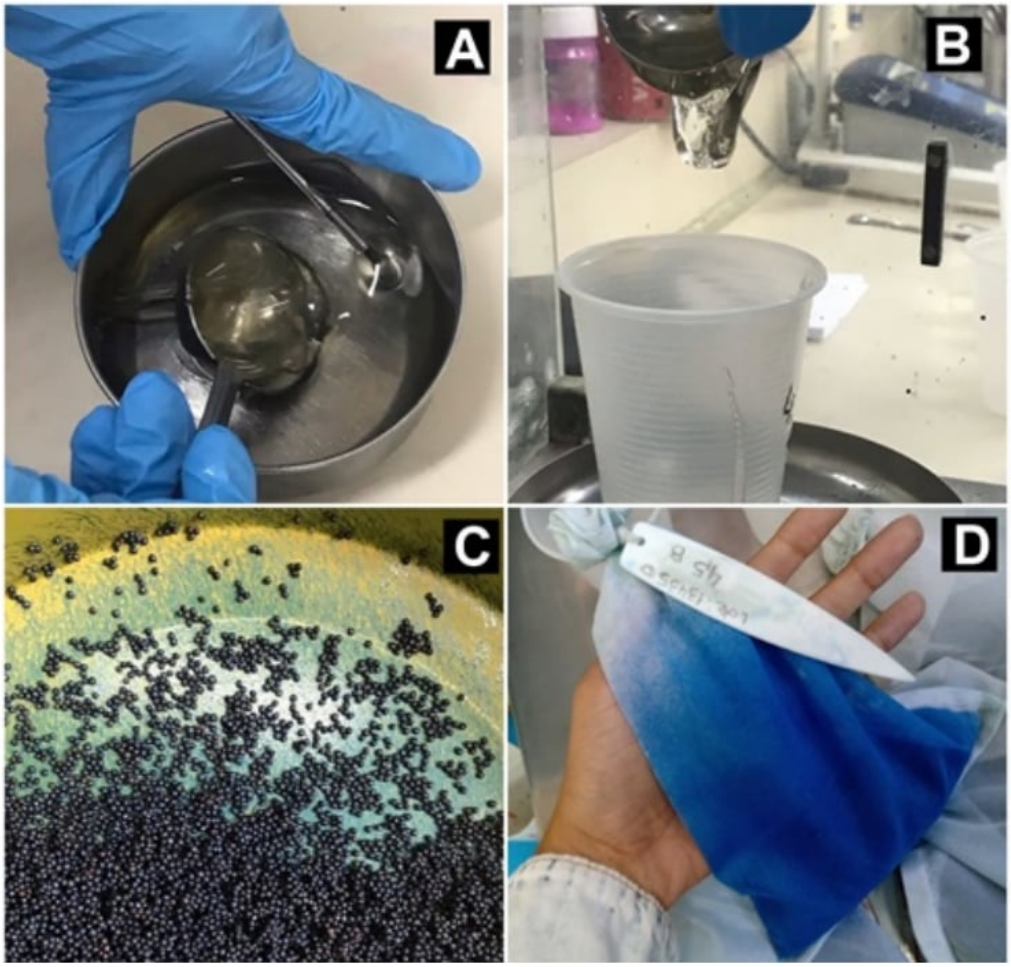


Figure 3. (**A**) Appearance of the boron source; (**B**) resistance to leakage of the boron source; (**C**) the end of the process of film-coating with boron source product; (**D**) boron-treated seeds in the dose of 4.5 g B kg^{-1} seed after drying, with residues of the product in the packaging.

3.1.3. Seedling Emergence and Early Growth

Despite the reduction in the quality of the seeds treated with B in the tests carried out in the laboratory, when the evaluation was carried out in trays with substrate, no difference was observed in relation to the control treatments for film coating, nor for pelleting at most doses (Table 2). The minimum emergence was 94%, which can be considered high and well above the minimum allowed for commercialization when germination is considered (minimum 80%) [38].

Only the two highest doses of B with pelleting had lower ESI values than the control treatment (untreated seeds) (Table 2). One possible reason for this result is the effect of irrigation, which may have removed the nutrient from close to the seeds, washing them and preventing the nutrient from having a phytotoxic effect on the seeds. Giménez-Sampaio and Sampaio [39] point out that the effects of film coating may be more restricted to the physiological potential of seeds and possibly to the initial growth of seedlings during a relatively short period after emergence.

In the nursery, by contrast to laboratory tests, seedlings were irrigated daily in a free-draining substrate, which probably diluted and leached the boron concentrated around the seed coat before it reached phytotoxic levels at the root surface, explaining why emergence and subsequent seedling growth were largely unaffected even at doses that suppressed germination in the laboratory assay. A comparable dose-dependent reduction in germina-

tion without a proportional effect on later seedling establishment has also been reported for boron-stressed canola and safflower seeds [40,41], reinforcing that the critical window of boron sensitivity in Brassicaceae seeds is concentrated in the earliest, most localized phase of germination, rather than in the seedling establishment phase that follows it.

However, even if seeds treated with B have shown the potential to produce normal seedlings, their low germination value would make it impossible to sell them, since the Ministry of Agriculture, Livestock and Food Supply (MAPA) establishes minimum germination values of 80% for the commercialization of cauliflower seeds [38]. Therefore, the only treatment that could be commercialized would be at a dose of 0.5 g B kg^{-1} of seed, for both film coating and pelleting. For film coating at this dose, mean germination reached 86.0%, a margin of six percentage points above the 80% commercial threshold; this margin should nonetheless be interpreted with some caution, as it reflects the mean of a limited number of replicates and does not by itself guarantee that all commercial seed lots would consistently clear the threshold.

Looking at the characteristics of the seedlings assessed in the greenhouse, there were differences between the seed treatment technologies for the shoot and root dry mass and seedling height, where film coating was the technology that showed the best results and there was no difference for the fresh mass of either the shoot or the root (Table 3).

Table 3. Emergence averages (EMERG), emergence speed index (ESI), shoot fresh mass (SFM), root fresh mass (RFM), shoot dry mass (SDM), root dry mass (RDM) and height (HEIG) of cauliflower seedlings originating from seeds film coated and pelletized with boron.

	EMERG	ESI	SFM	RFM	SDM	RDM	HEIG
	%			mg			cm
Film coating	97.0 a	3.5 a	448.3 a	55.8 a	51.3 a	9.7 a	7.6 a
Pelleting	97.8 a	3.4 a	389.3 a	57.8 a	40.9 b	7.7 b	6.7 a

Averages followed by the same letter in the columns do not differ according to the Tukey test ($p < 0.05$).

Probably, the boron that was in the pellets and went into the substrate via the irrigations had a toxic effect on the roots of the seedlings, reducing the accumulation of dry mass in them, without, however, damaging the fresh mass. This response is consistent with the boron toxicity effects described in Brassicaceae seedlings, where excess boron triggers oxidative stress and increased antioxidant enzyme activity, leading to reduced dry mass accumulation [30].

The only characteristic that showed a difference for the boron dose factor was the emergence speed index (ESI), in which the speed decreased linearly with the increase in doses, both for film coating and pelleting, with the reduction being more pronounced with pelleting (Figure 2F). Therefore, it is likely that the higher the dose, the greater the number of irrigations that would be needed to remove the boron-based product near the seeds, delaying emergence, but without affecting the final emergence assessed at ten DAS.

Considering that there was almost no effect on the characteristics of the seedlings, except for root dry mass and ESI, it can be concluded that, under nursery conditions, the doses tested had little effect on the development of cauliflower seedlings. Pessoa, Luchese and Luchese [26], in their research with maize, concluded that toxic concentrations of boron can accumulate and cause phytotoxicity, even killing the embryo, since the germinating seed does not have efficient physiological mechanisms to control the entry of boron via seed soaking, which occurs passively. However, treating maize seeds with boron increased the initial availability of this element in future plants grown from these enriched seeds. On the other hand, Rosal et al. [28], when pelleting maize seeds with boron (0, 0.05, 0.10, 0.15 and 0.20 g B kg^{-1} of seed), observed that seeds pelleted with the nutrient obtained greater

seedling length. The authors also found that the doses used had no significant effect on fresh and dry mass, so they could be used as seed treatments without causing phytotoxicity to the seedling.

In short, it is most likely that the high availability and rapid absorption of boron–polyol led to toxic accumulation or osmotic stress that critically affected the most vulnerable phase of seed germination. However, once the seedling overcame this phase and established itself in the soil/substrate, with its vascular system fully functioning, it was able to manage boron more efficiently, allowing for normal further development.

3.2. Agronomic Performance

3.2.1. Seedling Characteristics

The use of seeds pelleted with boron at the highest dose favored an increase in the B content in the seedlings (Table 4) and was the only treatment that differed from seedlings grown from untreated seeds (Table 5), resulting in seedlings with an average content of 21.5 mg kg^{-1}. The probable reason for this is that pelleting protects the nutrient that was added during the treatment, making it less susceptible to leaching with the irrigations carried out over the 25 days that the trays remained in the nursery. Studying the effects of boron fertilization on cauliflower production, Pizetta et al. [42] found B contents ranging from 18 to 40 mg kg^{-1} in the plants. Alves [43], on the other hand, determined an accumulation of boron in the leaves of 0.18 mg per plant at 30 days after transplanting 'Verona' cauliflower using a nutrient solution without boron. In *Brassica napus* and *B. rapa*, synchrotron-based speciation analyses have demonstrated that boron-tolerant lines accumulate less boron in shoot tissues relative to sensitive lines, suggesting that tissue-level boron distribution is an important determinant of deficiency versus toxicity responses [44].

Table 4. Boron content in cauliflower seedlings as a result of seed treatment through film coating and pelleting with different doses of boron.

	Film Coating	**Pelleting**
g B kg^{-1} Seed	**mg B kg^{-1}**	
0.5	10.5 Aa	8.0 Ba
4.5	13.8 Ab	21.5 Aa

Averages followed by the same uppercase letter in the column and lowercase letter in the line do not differ by the Tukey test ($p < 0.05$).

Table 5. Comparison of boron film coating and pelleting of cauliflower seeds with control (untreated seeds) for the boron content characteristic in seedlings.

Treatment	**Dose**	**Content**
	g B kg^{-1} Seed	**mg B kg^{-1}**
Film coating	0.5	10.5 *
Film coating	4.5	13.8 *
Pelleting	0.5	8.0 *
Pelleting	4.5	21.5
Untreated seed	-	13.3 *

* Averages statistically equal to untreated seed by the Dunnett test at 5% probability.

3.2.2. Characteristics Evaluated in Greenhouse-Grown Plants

The cycle was only influenced by the management factor, being 3 days earlier with standard management (Table 6), in which organic compost and B were used, as recommended by Trani et al. [18].

Table 6. Cycle, number of leaves (NL), leaf and stem fresh matter mass (LSFM) and dry matter mass (LSDM), dry inflorescence matter mass (IDM) and inflorescence diameter (DIAM) of cauliflower plants originating from seeds treated with boron in different management.

	CYCLE Days	NL	LSFM	LSDM g	IDM	DIAM cm
Management 1	57.79 a	26.3 b	1054.23 a	100.33 a	44.21 a	29.39 a
Management 2	60.83 b	27.8 a	1141.26 a	89.00 b	37.46 b	27.15 b

Management 1: Organic compost + B via soil and foliar; Management 2: No organic compost and no B. Averages followed by the same letter in the column do not differ by the Tukey test ($p < 0.05$).

Most likely, this earlier cycle is due to the use of organic compost, which, in addition to providing nutrients, can improve the physical, chemical and biological conditions of the soil [45] and favors faster development of cauliflower plants [46]. The cycle, with an average of 59 days, was earlier than that obtained in Jaboticabal with the summer hybrid Verona (69 days) [47]. The cycle is generally influenced by environmental and management conditions, and, in the case of cauliflower, the genotype has a major influence, with summer hybrids being earlier than winter hybrids, whose cycle can exceed 120 days after transplanting [48].

Although management without organic fertilizer and B sources resulted in a greater number of leaves, it showed a lower accumulation of dry matter in the leaves and inflorescences (Table 6), since organic fertilizer favors a greater accumulation of fresh and dry matter in cauliflower and other brassicas, as observed in this research and also highlighted by other authors [46]. This result confirms the importance of organic matter for plant development, and its use is recommended as a basic fertilizer for cauliflower in the state of São Paulo [18].

The average number of leaves per plant in this study was 27, higher than the average found by Kano et al. [48] of 23.4 leaves per plant for the winter cultivar Teresópolis Gigante and similar to the value found by Castoldi et al. [47] of 26 leaves per plant for the summer hybrid Verona. Monteiro et al. [49] found an average of 81.2 g of leaf dry mass, a value equivalent to this study's 94.7 g, since, in this case, the stem was also included. For inflorescence diameter, the average obtained was 28.3 cm, higher than the average of 18.5 cm for the Flamenco hybrid [49] and 20.5 cm for the Teresópolis Gigante cultivar [48]. Overall, the recommended management resulted in a greater inflorescence diameter, regardless of the dose of B in the seeds (Table 6), and greater fresh mass and total yield of inflorescences.

The average fresh mass of the inflorescence was 478 g, higher than that reported by Godoy and Cardoso [50] of 377 g for the Shiromaru II hybrid and lower than that obtained by Castoldi et al. [47] of 774 g for the Verona hybrid. The average yield obtained of 11.95 t ha^{-1} was similar to that found by Ferreira et al. [51] of 11.3 t ha^{-1} for the Desert hybrid and higher than that reported by Zanuzo et al. [52] of 6.67 t ha^{-1} for the Piracicaba Precoce cultivar. Of course, these values can vary depending on the cultivar, climatic conditions and management, but they show the good development of the plants in the pots, with the formation of inflorescences with a good size pattern.

When comparing the performance of plants originating from boron-treated seeds with plants originating from untreated seeds, it can be seen that when the correct fertilization management is carried out (Management 1), all the plants show satisfactory growth and development, since even the untreated seeds resulted in plants with averages similar to those from seeds treated with boron for all the characteristics evaluated (Table 7). Therefore, under these conditions, there is no advantage in treating the seeds with boron.

Table 7. Comparison of cauliflower plants originating from untreated seeds grown under management with organic compost before planting and boron via soil and foliar application with treatments in which the seeds were treated with boron for the characteristics: cycle, number of leaves (NL), leaf and stem fresh matter mass (LSFM) and dry matter mass (LSDM), inflorescence fresh matter mass (IFM), inflorescence dry matter mass (IDM), inflorescence diameter (DIAM), rate of plants with symptoms of B deficiency (DEF), total yield (TY) and commercial yield (CY).

Treatment	Dose g B kg^{-1} Seed	CYCLE	NL	LSFM	LSDM	IFM	IDM	DIAM	DEF	TY	CY
		Days			g			cm	%	t ha^{-1}	
Film coating	0.5	58.2 *	26.0 *	1007.5 *	101.2 *	504.9 *	42.3 *	29.1 *	0.0 *	12.6 *	12.6 *
Pelleting	0.5	57.8 *	26.2 *	1086.3 *	102.7 *	547.7 *	43.2 *	29.8 *	0.0 *	13.7 *	13.7 *
Film coating	4.5	57.0 *	27.0 *	1105.2 *	99.0 *	580.7 *	49.7 *	30.4 *	0.0 *	14.5 *	14.5 *
Pelleting	4.5	58.2 *	26.0 *	1017.9 *	98.5 *	454.5 *	41.7 *	28.3 *	0.0 *	11.4 *	11.4 *
Untreated seed	0	58.3 *	26.7 *	1039.5 *	88.8 *	521.0 *	43.5 *	29.0 *	0.0 *	13.0 *	13.0 *

* Averages statistically equal to the control (untreated seed) by Dunnett's test at 5% probability.

There were no plants with boron deficiency symptoms in all treatments under Management 1 (Table 7), confirming that correct boron fertilization (before planting and foliar application) is the best preventive strategy against the appearance of deficient plants, as highlighted by Pizetta et al. [42], Camargo et al. [53], Filgueira [54], Shelp and Shattuck [55] and Batabyal et al. [56]. However, farmers do not always have enough organic fertilizer and do not always fertilize correctly with boron, leading to serious losses, as observed in this research, because in the management without organic compost and in the absence of soil and foliar B fertilization (Management 2), when untreated seeds were used, the highest rate of boron-deficient plants (62.5%) and the lowest commercial yield were obtained (Table 8), consistent with the soil's initial available boron content (B = 0.21 mg dm^{-3}), well below the critical deficiency threshold of 0.48 mg kg^{-1} established for cauliflower [56], and lower than those of all the boron treatments, with the exception of pelleting at 0.5 g B kg^{-1} seed. The treatment with the highest numerical value for commercial yield (pelleted seeds with a dose of 4.5 g B kg^{-1} seed) achieved a 145.7% increase in commercial yield compared to the control with untreated seeds, and was the only treatment in which the rate of deficient plants was statistically equal to the recommended management.

Table 8. Comparison of cauliflower plants originating from untreated seeds grown under management without the use of organic compost before planting and without soil and foliar boron application with the treatments in which seeds received boron application for the characteristics: cycle, number of leaves (NL), leaf and stem fresh matter mass (LSFM) and dry matter mass (LSDM), inflorescence fresh matter mass (IFM), inflorescence dry matter mass (IDM), inflorescence diameter (DIAM), rate of plants with symptoms of B deficiency (DEF), total yield (TY) and commercial yield (CY).

Treatment	Dose g B kg^{-1} Seed	CYCLE	NL	LSFM	LSDM	IFM	IDM	DIAM	DEF	TY	CY
		Days			g			cm	%	t ha^{-1}	
Film coating	0.5	60.8 *	27.8 *	1195.1 *	96.7 *	468.9 *	34.0 *	27.4 *	33.3	11.7 *	7.9
Pelleting	0.5	61.3 *	27.7 *	1160.1 *	82.8 *	423.4 *	37.0 *	26.8 *	50.0 *	10.6 *	5.2 *
Film coating	4.5	60.5 *	27.3 *	1147.0 *	98.8 *	429.6 *	40.2 *	27.3 *	25.0	10.7 *	7.7
Pelleting	4.5	60.7 *	28.3 *	1062.8 *	77.7 *	414.0 *	38.7 *	27.1 *	12.5	10.4 *	8.6
Untreated seed	0	59.5 *	28.0 *	1174.5 *	95.5 *	424.3 *	32.7 *	26.7 *	62.5 *	10.6 *	3.5 *

* Averages statistically equal to the control (untreated seed) by Dunnett's test at 5% probability.

Film coating at 0.5 g B kg^{-1} seed, the only boron seed treatment that also met the commercial germination standard, increased commercial yield from 3.5 to 7.9 t ha^{-1}, a 125.7% gain relative to untreated seeds (Table 8). It should be emphasized that these gains refer to commercial yield: total yield did not differ between boron-treated and untreated seeds under Management 2 (Table 8), indicating that the benefit of seed-applied boron

resulted from the lower proportion of inflorescences with deficiency symptoms (brown rot), which were, therefore, unmarketable, rather than from greater inflorescence mass.

The agronomic benefit observed under Management 2 is plausible considering what is known about boron mobility in cauliflower itself. Working with the same species under greenhouse conditions, Shelp and Shattuck [55] showed that, although boron is largely phloem-immobile in cauliflower under adequate supply, the element is actively remobilized from older leaves toward the developing inflorescence once boron becomes limiting, indicating that even a small seed-applied dose can be redirected toward the curd under deficiency. Because the polyol-complexed boron used here is known to display greater phloem mobility than diffusion-limited boric acid in species that synthesize sugar alcohols [33,34], a small seed-localized boron reservoir may have continued to supply the root zone over the crop cycle, complementing cauliflower's natural remobilization response and explaining why even the lowest commercially viable dose (0.5 g B kg^{-1}, film coating) was sufficient to reduce deficiency incidence and raise commercial yield relative to the untreated control. This is consistent with reports from other cauliflower-growing regions, where modest, well-positioned boron doses were sufficient to maximize curd yield, while substantially higher doses became either unnecessary or phytotoxic [42,56–58]. It should be noted that boron dynamics in the substrate, soil, and plant tissue were not directly measured over the crop cycle in the present study, so this remobilization-based explanation remains inferential rather than directly demonstrated.

However, knowing that brassica species do not require pelleting for precision planting, as the seeds are not particulary small and have a rounded shape, film coating is the technology often used by companies [59], considering the economic point of view.

In Management 1, there was no advantage in applying boron to seeds. On the other hand, in Management 2 (without applying boron to the soil and via foliar application), treating the seeds with this nutrient was efficient in reducing the rate of boron-deficient plants and, consequently, increasing commercial yield. Therefore, in a situation where the farmer does not usually manage B sources correctly in the field, applying B to seeds is an alternative that reduces the occurrence of deficient plants, resulting in higher commercial yield. In addition, it is a technology that, if adopted, is potentially more cost-effective than soil and foliar B application and, above all, than incorporating large quantities of organic fertilizer into the soil before planting. However, a formal economic analysis was beyond the scope of this study and would be needed to confirm this advantage.

When evaluating agronomic performance, it is important to emphasize that Management 1 and Management 2 differ in both organic amendment and boron supply. Because castor bean cake contains background boron (36 mg B kg^{-1}), its omission in Management 2 was necessary to achieve a B-limiting state, but this also omitted the physical and nutritional benefits of organic matter. Therefore, the overall differences between the two managements reflect the combined effect of organic fertilization and total B availability rather than boron alone. Consequently, performance evaluations and treatment comparisons are focused strictly within each fertilization management regime.

It is important to acknowledge that the field results presented here were obtained under a protected, pot-based environment (8 L pots, controlled irrigation, greenhouse), rather than under open-field commercial conditions. Although this approach allowed precise control of fertilization treatments and substrate uniformity, factors such as natural spatial variability in soil boron availability, rainfall-driven leaching, and the root-volume restriction inherent to pot cultivation may affect the magnitude of the responses reported here. Therefore, the use of seed-applied boron as a complementary strategy to mitigate B deficiency should be validated under open-field, multi-environment trials before broad extrapolation to commercial cauliflower production.

4. Conclusions

Cauliflower seeds treated with boron can meet commercial germination standards (≥80%) at the lowest dose tested (0.5 g B kg^{-1} seed) with both film coating and pelleting technologies, maintaining acceptable final germination despite a reduction in initial seed vigor (first germination count and germination speed index).

Under the greenhouse pot trial without soil or foliar boron fertilization (B-deficient baseline management), boron seed treatment substantially reduced deficiency incidence and enhanced commercial inflorescence yield. Film coating at 0.5 g B kg^{-1} seed was the most suitable strategy, as it effectively maintained seed marketability while providing a 125.7% increase in commercial yield (from 3.5 to 7.9 t ha^{-1}) relative to untreated seeds, representing the standard technology routinely adopted by seed companies for *Brassica* species [59]. While these pot-scale findings demonstrate the agronomic potential of boron-coated seeds, further trials under open-field commercial conditions are warranted to validate practical yield responses.

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