

## Pronamide Applied to Sunflower Seeds for *Orobanche cumana* Control<sup>1</sup>

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**Abstract:** Field and laboratory studies were conducted from 1993 to 1997 to determine the feasibility of controlling nodding broomrape in sunflower by treating crop seeds with pronamide. Soaking sunflower seeds for 5 min in 50% pronamide solution or coating at the equivalent of 2 kg/ha with pronamide did not impair seed germination or seedling growth, and controlled nodding broomrape 49 to 68% and 51 to 77%, respectively, up to 105 d after planting. Studies of the effect of treated sunflower seeds on germination and seedling growth, at several time intervals after the herbicide application (0, 30, 60, and 90 d after treatment [DAT]), were conducted. Soaking with 50% solution or coating at 2 kg/ha reduced seedling growth by 20 and 24% 60 DAT, respectively, compared with the control.

**Nomenclature:** Pronamide; nodding broomrape, *Orobanche cumana* Wallr. #<sup>3</sup> ORACU; sunflower, *Helianthus annuus* L.

**Additional index words:** Herbicide seed treatment, parasitic weed.

**Abbreviations:** DAP, days after planting; DAT, days after treatment; HST, herbicide seed treatment.

### INTRODUCTION

*Orobanche* spp. (broomrapes) are obligate root holoparasites of many broadleaf species. Several *Orobanche* species parasitize economically important crops. Nodding broomrape mainly attacks sunflowers in extensive areas of southern and eastern Europe and the Middle East (Parker 1994), causing considerable losses in crop yield. In Spain, this species recently infested about 100,000 and 350,000 ha in the central and southern parts of the country, respectively (García-Torres et al. 1994a).

Herbicides are the most important methods available to control broomrapes in diverse crops (Foy et al. 1989; García-Torres 1994). These include late postemergence application of glyphosate at 60 g/ha on broad bean (*Vicia faba* L.) (García-Torres et al. 1987; Jacobsohn and Levy 1986), sunflower (Castejón-Muñoz et al. 1991), and tobacco (*Nicotiana tabacum* L.) (Lolas 1986), and of imazapyr at 10 to 15 g/ha on sunflower (García-Torres et al. 1995), broad bean, and pea (*Pisum sativum* L.) (García-Torres 1994). Preemergence applications of imazethapyr and imazapyr were also effective for crenata

broomrape (*Orobanche crenata* Forsk) control in broad bean and pea (García-Torres and López-Granados 1991) and for nodding broomrape control in sunflower (García-Torres et al. 1994b). However, the effectiveness of pre-emergence herbicide treatments is considerably influenced by soil texture and precipitation.

Pronamide belongs to the substituted amides family, inhibits mitosis in susceptible species (Vaughn and Lehnen 1991), and is a selective herbicide for control of a wide range of grasses and certain broadleaf weeds in many crops (fruit, vines, vegetables, legumes, and forestry). This herbicide can be applied preemergence and postemergence and is readily absorbed by plants through the root system, translocated upward, and distributed throughout the whole plant. Control of weeds with pre-emergence applications of pronamide in sunflower is influenced by soil characteristics (organic matter, clay content, and pH) (Carlson et al. 1975). According to Kleinfeld et al. (1987), the limiting factor for effective control of nodding broomrape by pronamide seems to be its short-term persistence in wet, warm soil.

Commercial seed treatments with fungicides or insecticides are commonly used in modern agriculture. In contrast, several cases of the use of herbicide-treated seeds have been reported only for control of alfalfa (*Medicago sativa* L.) (Dawson 1981, 1987), grass weeds (Dale 1983), and witchweed (*Striga* spp.) in cowpea (*Vigna unguiculata* L.) (Berner et al. 1994, 1997) or maize (*Zea mays* L.) (Kanampiu et al. 2001). A herbicide

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<sup>3</sup> Letters following this symbol are a WSSA-approved computer code from *Composite List of Weeds*, Revised 1989. Available only on computer disk from WSSA, 810 East 10th Street, Lawrence, KS 66044-8897.

applied to crop seeds may be of particular interest for broomrape control because parasite infection mainly occurs in the root system zone, near where the crop seed is located. Recently, new herbicide seed treatments (HST) for crenata broomrape control have been developed for winter legumes (Jurado-Expósito et al. 1996, 1997) but not for nodding broomrape in sunflower. The objective of this study was to determine the feasibility of pronamide treatments for nodding broomrape control in sunflower seeds.

## MATERIALS AND METHODS

**Herbicide Seed Treatments.** Coating treatments consisted of stirring the sunflower (cv. 'Agrosur') seeds with a dressing substance<sup>4</sup> and distilled water in the proportion of 10 g seeds per milliliter of dressing substance per 0.5 milliliter of water. After 20 to 24 h at room temperature, the seeds were added to the herbicide solution (1 g seeds per milliliter of herbicide solution) and shaken for 3 min. Coating treatments consisted of 0, 1, and 2 kg ai/ha of pronamide<sup>5</sup>.

Soaking treatments were performed by immersing the crop seeds for 5 min in 0, 25, and 50% ai pronamide solutions. Afterward, the seeds were dried at room temperature for 24 h and kept in the dark until sowing.

**Germination and Seedling Growth.** The germination test was carried out on 28- by 24-cm Whatman GF/A paper sheets moistened with sterile water, on which 20 treated sunflower seeds were placed 6 cm apart. The paper with the treated seeds was then rolled in a germination filter paper and covered with aluminum foil to prevent evaporation. After 7 d at  $22 \pm 1$  C in darkness, the number of germinated seeds (coleoptile > 5 mm) per treatment was determined. The experimental design was a randomized complete block with three replications, and the experiment was repeated. Means and standard error were calculated.

The seedling growth test was set up in the growth chamber at  $20 \pm 1$  C with a 12-h light-dark photoperiod ( $220 \mu\text{E}/\text{m}^2/\text{s}$ ). Pots (55 ml) were filled with 4 g of wet perlite (10 ml water), one treated crop seed, and 45 g of wet soil (4.5 ml water). After 30 d, the number of healthy seedlings (> 5 to 7 cm with no phytotoxic symptoms) of each treatment was determined and expressed as a percentage of the untreated control. The experimental design was a randomized complete block with three

replications of 10 pots each, with each experiment being repeated. The means and standard error were calculated.

To determine the effect of prolonged exposure of sunflower seeds to the herbicide, similar tests of germination and seedling growth were conducted at several time intervals after herbicide application (0, 30, 60, and 90 d after treatment [DAT]).

**Field Studies with HST.** Field studies were carried out in 1993, 1994, 1995, 1996, and 1997 in different fields located in Seville (southern Spain) and in clayey soils naturally infested with nodding broomrape with pH 7.8 and 1.2% organic matter. Mean air temperature and rainfall were recorded daily throughout the experimental period, and the rainfall during the crop-growing season (February to July) in the different years was considered.

The sowing date was around mid-February, and the sowing rate was 6 kg/ha. Experimental plots consisted of four rows 0.7 m apart and 5 m long. Trifluralin<sup>6</sup> [2,6-dinitro-*N,N*-dipropyl-4-(trifluoromethyl)-benzenamide] at 0.8 kg/ha was preplant incorporated for general weed control. Hoeing and hand weeding were conducted as needed but did not include nodding broomrape. Coating and soaking treatments were 0, 1, and 2 kg ai/ha of pronamide and 0, 25, and 50% ai pronamide solutions, respectively.

In selected treatments, the two central rows from each plot were hand harvested by late July. Sunflower plants were then dried, cleaned, and the seeds weighed to determine yield. Nodding broomrape control was evaluated at several time intervals (75, 90, and 105 d after planting [DAP]) by determining the number of parasitic plants that had emerged from the soil.

**Field Studies on Crop Tolerance.** To study the tolerance of sunflower to pronamide seed treatments by coating or soaking, two similar field experiments were conducted in 1995 in two locations in a soil that was not infested with nodding broomrape. (province of Seville, southern Spain). The experimental plots consisted of four rows 0.7 m apart and 10 m long. Herbicide treatments were performed by soaking in 0, 25, and 50% ai pronamide solutions and by coating with 0, 1, and 2 kg ai/ha of pronamide.

All the experimental plots were subjected to a visual evaluation of crop phytotoxicity 4 to 5 wk after crop emergence, using a scale of 0 to 100, with 0 indicating no injury, 10 to 30 indicating slight injury, 40 to 60 indicating moderate injury, 70 to 90 indicating severe injury, and 100 indicating complete kill. Tolerance of

<sup>4</sup> Peridiam Blue®, dressing substance, Rhône-Pulenc, Follège-Mereville, France.

<sup>5</sup> Kerb®, 50W herbicide, Rohm & Haas, Philadelphia, PA.

<sup>6</sup> Treflan®, E.E. 480 g ai/L herbicide, DowElanco, Indianapolis, IN.

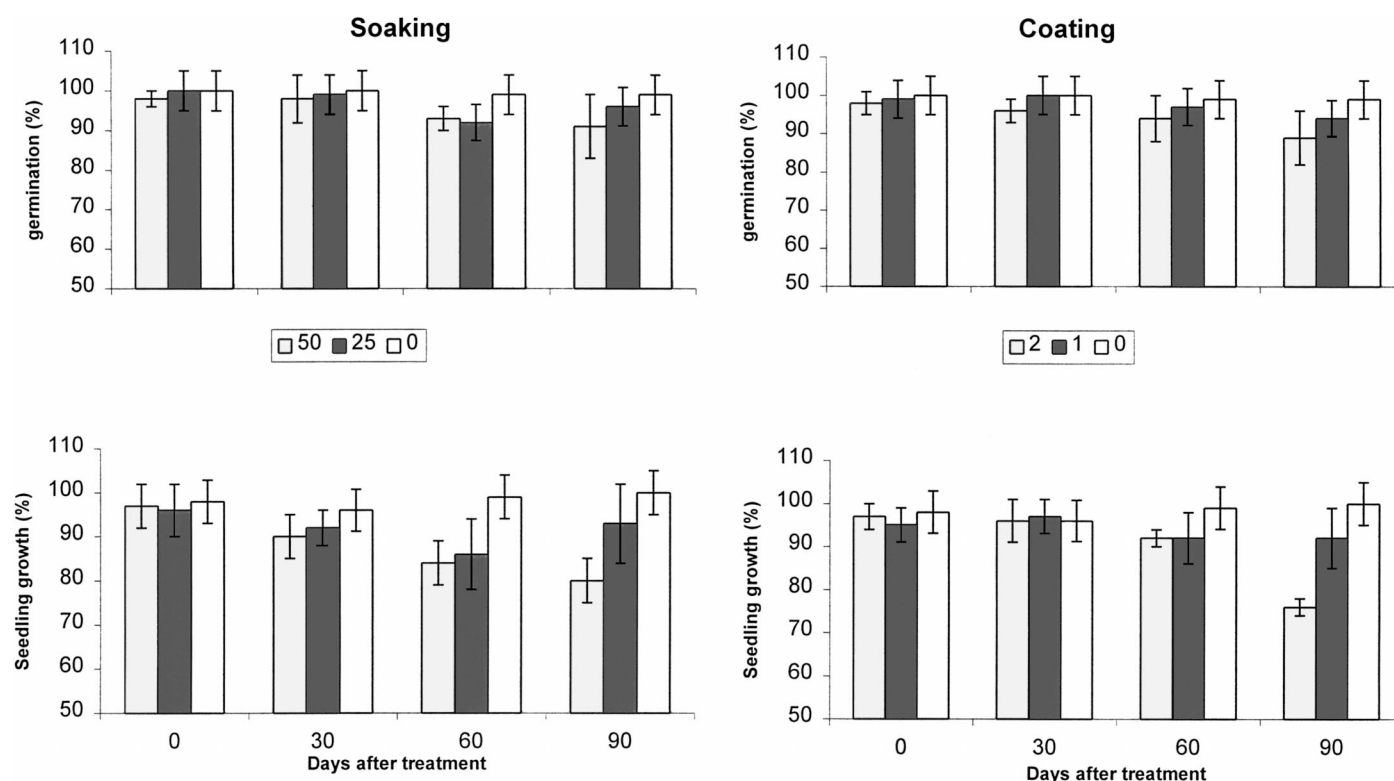


Figure 1. Effect of pronamide seed treatments by coating at 2, 1, and 0 kg/ha and by soaking at 50, 25, and 0% on germination and seedling growth. Bars indicate standard error of the mean.

sunflower to the seed treatments was evaluated by determining biomass (dry weight), head diameter and weight, and seed yield.

All field experiments carried out in the different locations and years were designed as completely randomized blocks with three replications. An ANOVA was done, and a separation of means test (Duncan's) was conducted along with the ANOVA.

## RESULTS AND DISCUSSION

**Germination and Seedling Growth.** Germination of sunflower seeds after pronamide treatments was 89% or higher for all treatments (Figure 1). The elapsed time between the herbicide applications and the germination tests did not substantially alter the germination percentage. Seedling growth was generally similar to that in the untreated control for the higher rates of soaking and coating treatments up to 30 DAT. HST adversely affected seedling growth 60 d or more after treatment. For example, 60 DAT, seedling growth was reduced by 20 and 24% for soaking at 50% and coating at 2 kg/ha, respectively, compared with the control (Figure 1).

**Field Studies with HST.** Neither soaking nor coating with pronamide affected germination or seedling emergence from the soil. Furthermore, no crop phytotoxicity symptoms were observed except at the higher rates of soaking (50% of pronamide) or coating (2 kg/ha), which caused slight injury (10 to 30) when compared with the control.

Pronamide seed treatment reduced nodding broomrape emergence per host plant. Nodding broomrape control with sunflower seeds treated by coating at 1 and 2 kg/ha was medium to high in 1993 (Figure 2a, 70 to 77%), 1994 (Figure 2b, 60 to 75%), and 1995 (Figure 2c, 60 to 69%) and slightly lower in 1996 (Figure 2d, 59 to 68%) and 1997 (Figure 2e, 41 to 51%) when compared with the untreated control (0 kg/ha).

Generally, sunflower yield increased with pronamide HST as compared with the untreated control during the 5 yr when the experiments were conducted (Table 1). Seed coating with pronamide at 1 and 2 kg/ha increased yield significantly from 17 to 45% in 1993, from 25 to 40% in 1994, and from 26 to 33% in 1995 as compared with nodding broomrape-infested untreated seeds, and slightly lower benefits were obtained in 1996 and 1997 (Table 1).

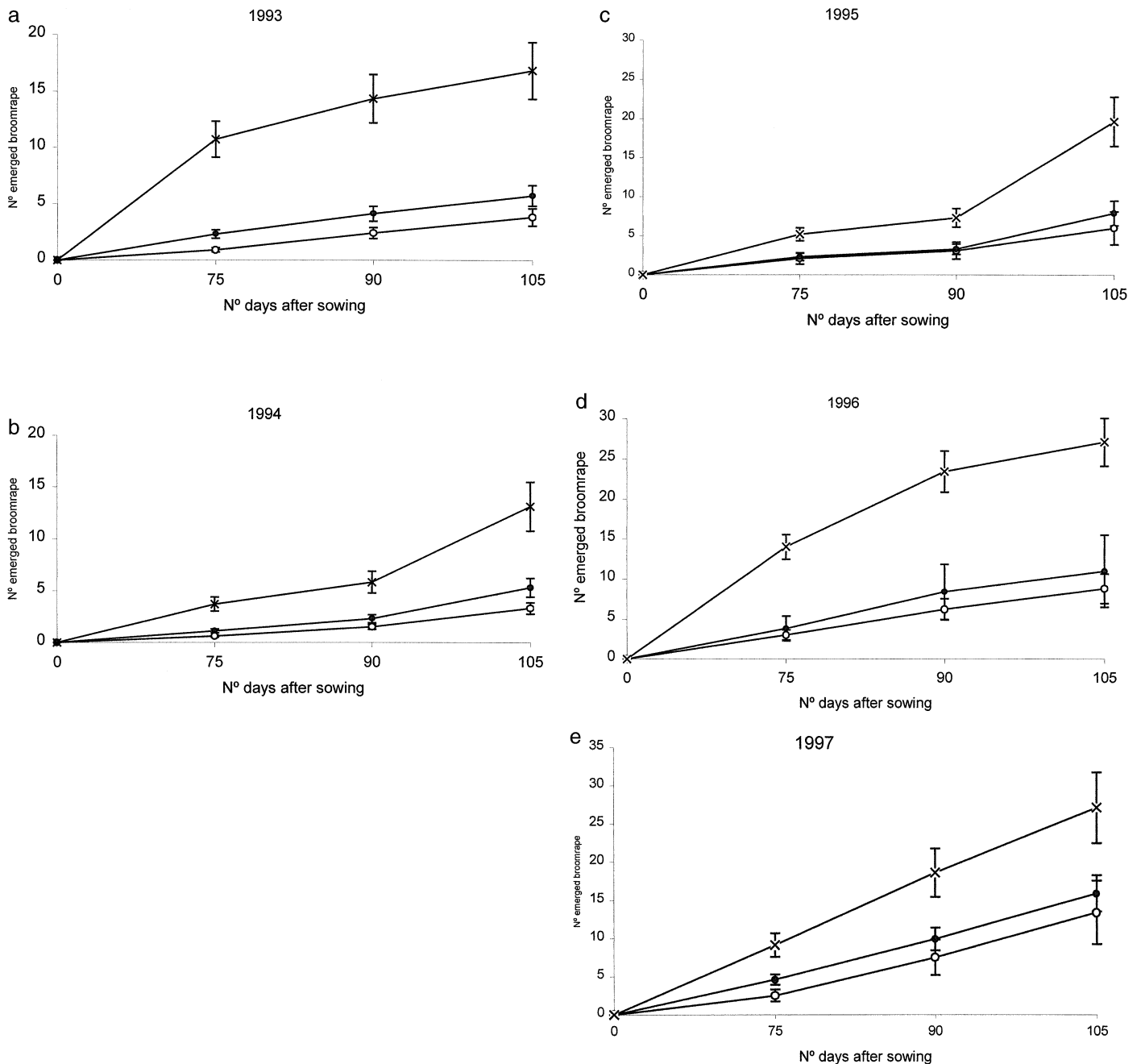


Figure 2. Nodding broomrape emergence per sunflower plant as affected by treatment of sunflower seeds with pronamide by coating at 2 kg/ha (○), 1 kg/ha (●), and 0 kg/ha (×). (a) 1993, (b) 1994, (c) 1995, (d) 1996, (e) 1997. Bars indicate standard error of the mean.

Pronamide applied by soaking controlled nodding broomrape less than when it was applied by coating. Thus, at 25 to 50%, pronamide herbicide solutions resulted in medium to high control of nodding broomrape (57 to 68%) in 1995 (Figure 3a) and 1996 (Figure 3b, 22 to 49%). Increases in sunflower seed yield after 5 min seed soaking in 25 to 50% pronamide solutions ranged from 17 to 45% in 1995 and from 20 to 17% in 1996 compared with the control, although there were no

significant differences with the untreated control (Table 1).

Differences between years in the efficiency of nodding broomrape control after coating or soaking treatments can be attributed mainly to environmental conditions, as in the case of conventional preemergence applications. Generally, nodding broomrape infections decreased and preemergence herbicide efficiency increased in low-precipitation years as compared with medium- or high-precipitation years.

Table 1. Sunflower yield as affected by pronamide-treated seed by coating (1993–1997) and soaking (1995–1996).<sup>a</sup>

Treatment	Rates	Yield				
		1993	1994	1995	1996	1997
		kg/ha				
Coating (kg ai/ha)	2.0	1,978 c	1,827 c	2,849 b	1,548 b	1,550 b
	1.0	1,605 b	1,628 b	2,689 b	1,369 b	1,628 b
	0.0	1,368 a	1,302 a	2,141 a	930 a	1,274 a
Soaking (% ai)	50.0			1,795 b	1,570 b	
	25.0			1,447 b	1,610 b	
	0.0			1,240 a	1,338 a	

<sup>a</sup> Values for treatment within columns followed by the same letter are not significantly different according to Duncan's test ( $P \leq 0.05$ ).

precipitation years, which was probably the result of enhanced herbicide degradation (García-Torres and López-Granados 1991; García-Torres et al. 1994b). Total rainfall during the crop-growing season (from February to July) in the different years when the field experiments were carried out was 131, 153, 180, 263, and 271 mm in 1993, 1994, 1995, 1996, and 1997, respectively. This could explain the better nodding broomrape control during the first 3 yr than during the last 2 yr. According to

Kleifeld et al. (1987), the limiting factor for effective nodding broomrape control by pronamide seems to be its short-term persistence in wet, warm soil.

**Crop Tolerance.** Pronamide applied by coating or by soaking did not produce crop phytotoxicity symptoms except at the higher rates assayed by soaking (50% of pronamide) or coating (2 kg/ha), which caused slight injury (10 to 30) as compared with the control. Generally, seeds growing in nodding broomrape-free soils and treated by coating with pronamide at 1 and 2 kg/ha or by soaking at 25 and 50% did not show decreased sunflower plant biomass, head diameter, or yield (Table 2). Thus, pronamide applied at 2 kg/ha by coating and at 50% by soaking resulted in a yield of 106 and 108%, respectively, as compared with the untreated control, and the differences between treatments were not statistically significant. It should be pointed out that these herbicide treatments also were effective for nodding broomrape control as previously indicated. The differences between the coating and soaking treatments were not evident in terms of crop tolerance.

In general, seed herbicide treatments for parasitic weed (witchweed or nodding broomrape) control are considered an advantageous technology because farmers will not need to purchase and calibrate sprayers (Kan-

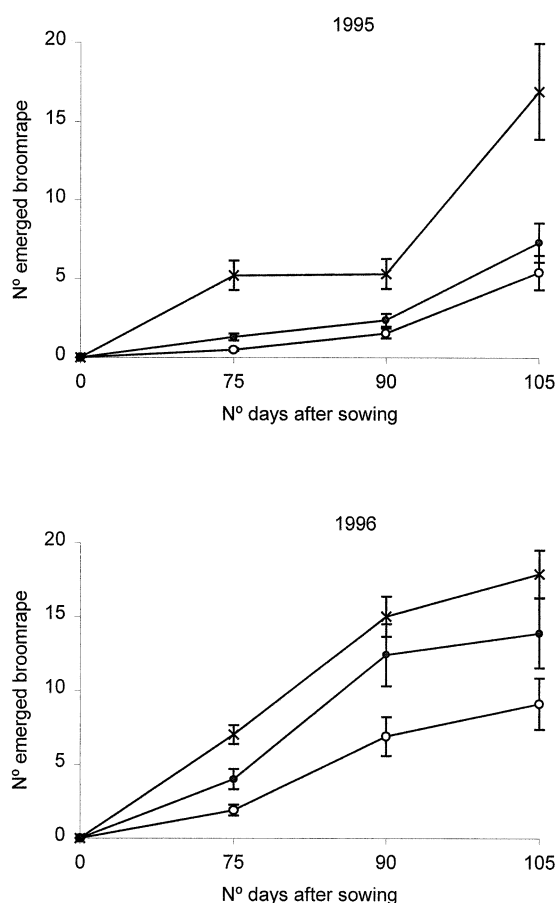


Figure 3. Nodding broomrape emergence per sunflower plant as affected by treatment of sunflower seeds with pronamide by soaking at 50% (○), 25% (●), and 0% (×). (a) 1995, (b) 1996. Bars indicate standard error of the mean.

Table 2. Effect of pronamide seed treatment on nodding broomrape-free sunflower (1995). Numbers in parentheses are standard errors.

Rates	Aerial biomass <sup>a</sup>	Head weight <sup>a</sup>	Head diameter	Yield
	g/plant	cm/plant	cm/plant	kg/ha
Coating (kg ai/ha)				
0	216 (63)	91 (31)	10.0 (1.5)	1,170 (405)
1	242 (83)	88 (38)	9.9 (2.2)	1,130 (373)
2	264 (126)	96 (25)	10.4 (1.7)	1,243 (334)
Soaking (% ai)				
0	255 (73)	107 (24)	10.7 (2.9)	1,380 (300)
25	275 (98)	121 (31)	10.2 (1.0)	1,548 (255)
50	269 (5)	116 (8)	10.4 (1.1)	1,501 (98)

<sup>a</sup> Sunflower dry weight.

ampiu et al. 2001) and because mechanical application costs for preemergence herbicides are unnecessary (US\$9/ha, Spanish current price). Also, HST could be advantageous for the nodding broomrape-resistant sunflower seed companies because they could offer seeds with an additional value. However, under high nodding broomrape infestations or in very rainy years (or both), which enhance and prolong the parasite attachment, neither the conventional preemergence treatments nor the seed herbicide treatments are expected to control nodding broomrape satisfactorily by themselves. In these unfavorable situations, subsequent and specific postemergence treatments with single applications of imazapyr<sup>7</sup> at 10 to 15 g/ha or double treatments of glyphosate<sup>8</sup> at 40 g/ha on sunflower plants with 12–19 and 22 leaves, respectively (Castejón-Muñoz et al. 1991; García-Torres et al. 1995), also should be applied to obtain acceptable nodding broomrape control and crop yield.

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<sup>7</sup> Arsenal®, 240 g ae/L herbicide, American Cyanamid Co, Bridgewater, NJ.

<sup>8</sup> Roudup®, 360 g ae/L herbicide, Monsanto, St. Louis, MO.