

Light Intensity Is a Main Factor Affecting Fresh Market Spinach Tolerance for Phenmedipham

Author(s): Ran N. Lati, Beiquan Mou, John S. Rachuy, and Steven A. Fennimore

Source: Weed Science, 64(1):146-153.

Published By: Weed Science Society of America

DOI: <http://dx.doi.org/10.1614/WS-D-15-00056.1>

URL: <http://www.bioone.org/doi/full/10.1614/WS-D-15-00056.1>

BioOne (www.bioone.org) is a nonprofit, online aggregation of core research in the biological, ecological, and environmental sciences. BioOne provides a sustainable online platform for over 170 journals and books published by nonprofit societies, associations, museums, institutions, and presses.

Your use of this PDF, the BioOne Web site, and all posted and associated content indicates your acceptance of BioOne's Terms of Use, available at www.bioone.org/page/terms_of_use.

Usage of BioOne content is strictly limited to personal, educational, and non-commercial use. Commercial inquiries or rights and permissions requests should be directed to the individual publisher as copyright holder.

Light Intensity Is a Main Factor Affecting Fresh Market Spinach Tolerance for Phenmedipham

Ran N. Lati, Beiquan Mou, John S. Rachuy, and Steven A. Fennimore*

The few available herbicides for fresh market spinach do not provide adequate weed control, and there is need for additional herbicide tools. Phenmedipham is registered for use in processing spinach but not in fresh spinach, because of potential injury and the short interval between application and spinach harvest. The objectives of this study were to evaluate the tolerance level of fresh spinach varieties to phenmedipham and evaluate the impact of light intensity on tolerance of spinach to phenmedipham. In the greenhouse, nine spinach varieties were treated with phenmedipham ($0.55 \text{ kg ai ha}^{-1}$). Spinach varieties exhibited a wide range of tolerance, and dry weights of treated plants ranged from 40 to 78% compared to the nontreated control. Based on the phenmedipham tolerance screen, two varieties with low (Nordic) and high (Regal) tolerance to phenmedipham were treated, then exposed to half (shaded) and full (nonshaded) sunlight. Nonshaded Nordic treated with phenmedipham had 65% lower dry weight compared to similarly treated plants grown under shade, suggesting that spinach tolerance to phenmedipham was mainly affected by light intensity. Measurements of electron transfer intensity in photosystem II also showed tolerance to phenmedipham that varied among spinach varieties and light intensity. The maximum values of electron transfer in photosystem II of Regal treated with phenmedipham were higher than those of similarly treated Nordic. In the field, phenmedipham was applied under varied light and temperature conditions. The impact of light intensity on yield of treated spinach was greater than the impact of temperature. Phenmedipham applied under high light conditions was more injurious than when applied under low light conditions. Results from this study can contribute to successful integration of phenmedipham into currently used fresh spinach weed management, which in turn can allow more efficient production of this crop.

Nomenclature: Phenmedipham; spinach, *Spinacia oleracea* L. SPQOL; ‘Nordic’; ‘Regal’.

Key words: Fresh spinach yield losses, light, phenmedipham, selectivity, temperature.

Fresh market spinach is an important vegetable crop in the United States and in other parts of the world, and has several categories—baby leaf, teenage leaf, and bunched (Morelock and Correll 2008). The baby leaf and the teenage leaf are seeded in high density (between 24 and 48 seed lines per 2-m bed) and harvested automatically at a very young growth stage; the bunched spinach is seeded at lower density (between 12 and 21 seed lines per 2-m bed) and is manually harvested at a more developed stage (Koike et al. 2011). Recent development of the baby-leaf spinach and its high nutritional values are possible reasons for increased demand for this crop (Correll et al. 2011). Over the last decade the production area of fresh spinach has increased by 155% in the

United States, and California leads its production with 8,400 ha which constitutes 60% of the nation’s total production area (U.S. Department of Agriculture [USDA] 2013). Most spinach grown in California is baby leaf (Koike et al. 2011).

Fresh spinach has low tolerance for weed contamination and is very susceptible to weed competition (Fennimore et al. 2001). Nevertheless, like many other specialty crops, fresh spinach has a complex weed management program. The few herbicides registered for spinach provide only partial weed control (Fennimore and Doohan 2008). In California, cycloate applied PRE is the primary herbicide for fresh spinach and no POST broadleaf herbicide is available for this crop (California Department of Pesticide Regulation [CA-DPR] 2013; USDA 2013). Cycloate application often results in partial control of weeds such as burning nettle (*Urtica urens* L.), common purslane (*Portulaca oleracea* L.), and common chickweed [*Stellaria media* (L.) Vill.] (Fennimore et al. 2001; Lati et al. 2015; Smith et al. 2013). Furthermore, the high seeding densities of baby- and teen-leaf spinach makes cultivation

DOI: 10.1614/WS-D-15-00056.1

* First, third, and fourth authors: Postdoctoral Researcher, Staff Research Associate, and Extension Specialist, respectively, Department of Plant Sciences, University of California at Davis, 1636 East Alisal Street, Salinas, CA 93905; second author: Research Geneticist, U.S. Department of Agriculture, Agricultural Research Service, 1636 East Alisal Street, Salinas, CA 93905. Corresponding author’s E-mail: ranlati@ucdavis.edu

between the crop rows impossible (Fennimore et al. 2001). Therefore, spinach producers hand weed to achieve commercially acceptable weed control. Hand weeding is usually conducted immediately before harvesting when weeds are large, and as a result, it is a slow and costly practice (Takele 2013). Buyers will reject spinach contaminated with weeds, so growers have no choice but to hand weed rather than risk loss of their crop (Smith et al. 2013).

Phenmedipham is a photosystem II (PS II) inhibitor that can be applied POST for the control of annual broadleaf weeds and some grasses (Davies et al. 1990). Phenmedipham binds to the D1 protein of PS II and blocks the electron transfer between the primary and secondary quinones (Q_A and Q_B). As a result, the photosynthetic electron transport chain is disturbed, and ATP production and carbon fixation are inhibited (Roberts et al. 2003). Plant death is mainly due to the formation of reactive oxygen species (ROS), which cause lipid peroxidation, and proteolysis of thylakoid membranes (Fufezan et al. 2002).

Phenmedipham is registered for use on spinach grown for processing or seed and on sugar beet (*Beta vulgaris* L.). Its application may cause temporary injury such as growth retardation, chlorosis, leaf margin burn, and tip burn (Anonymous 2013; Wallace and Petty 2007). Additionally, crop tolerance to phenmedipham (or phenmedipham selectivity) is not absolute; under some environmental conditions application might result in injury from which the crop does not recover and result in yield loss (Norris 1991). Starke and Renner (1996) mentioned that phenmedipham applied at temperatures above 22 C in sugar beet can lead to yield losses. They suggested that phenmedipham should be applied in the evening or at night rather than morning to improve crop safety. Norsworthy and Smith (2005) evaluated phenmedipham in kale (*Brassica oleracea* var. *acephala*), leafy turnip greens (*Brassica rapa* L. Alamo), mustard greens (*Brassica juncea* (L.)), and collard (*Brassica oleracea* L.). They reported variations in crop tolerance and yield between years, which was also related to differences in temperature conditions at time of application.

Phenmedipham is not registered for fresh spinach because spinach has a short growth cycle (~33 d in midsummer in coastal California), which leaves little time for recovery from herbicide injury (Wallace and Petty 2007). Lati et al. (2015) showed that sequential applications of phenmedipham at the four-leaf stage after cycloate PRE provided safe and effective

weed control. However, the 21-d preharvest interval (PHI) specified on the phenmedipham label means that this treatment is not feasible for spinach—the four-leaf-stage treatment (~17 d) plus the 21-d PHI means that the harvest must be delayed, which can affect net returns and produce quality (Anonymous 2015; Koike et al. 2011; Lati et al. 2015). To make phenmedipham more useful for fresh spinach production, this herbicide must be applied at the earliest possible growth stage that is safe. One way to reduce phenmedipham injury to spinach and allow earlier application timing may be by spraying this herbicide under low light conditions. Light intensity is known to impact the selectivity and phytotoxicity of other PS II herbicides (Brain et al. 2012). However, there are no data about the impact of light on phenmedipham selectivity in fresh spinach. In addition, there were no reports of differences in the tolerance of fresh spinach variety to phenmedipham; such data can extend the herbicide options and improve weed control programs for this crop (Leon and Tillman 2015). Therefore, the main objectives of this study were (1) to evaluate the tolerance of different fresh spinach varieties to phenmedipham, (2) to evaluate the impact of light on the tolerance of fresh spinach to phenmedipham, and (3) to evaluate the impact of application time of day on phenmedipham weed control.

Material and Methods

Greenhouse Studies. *Variety Screen.* The tolerance of several fresh spinach varieties to phenmedipham was evaluated. Pots (8-cm diameter) were filled with sandy loam soil (2.1% organic matter and pH 7.0) and seeded with nine spinach varieties separately: Santorini, Crocodile, Polar Bear (Snow Seed Co., Salinas, CA 93908), Bolero, Cypress (Seminis Vegetable Seeds, Inc., St. Louis, MO 63167), Unipack 12, Tye, Regal (Siegers Seed Co., Holland, MI 49424) and Nordic IV (Gowan Seed Co., Chualar, CA 93925). Plants were grown outside the greenhouse under direct sun conditions. Pots were sprayed with 0.55 kg ai ha⁻¹ phenmedipham (Spin-Aid, Engage Agro, Phoenix, AZ 86303) at the four-leaf stage or left not sprayed. Phenmedipham was applied with a CO₂-pressurized backpack sprayer equipped with 8002VS flat-fan nozzles (Tee Jet Technologies, Wheaton, IL 60189) calibrated to deliver 337 L ha⁻¹ at 290 kPa. After being sprayed, plants were left outside the greenhouse. Ten days after treatment the aboveground parts of the plants were harvested, dried at 80 C for 6 d,

and their dry weights recorded. The experiment was conducted in July 2013 and repeated in September–October 2013. Average temperature and photoperiod were 12.5 and 17.6 C, and 15/9 and 12/12 day/night hours, for the July and September–October trials, respectively.

Shade Sensitivity. To evaluate the impact of shade on the tolerance of fresh spinach to phenmedipham, pots (8-cm diameter) were filled with soil and seeded with two varieties, ‘Nordic’ and ‘Regal’. These varieties were used because they exhibited different levels of tolerance to phenmedipham: Regal—high, Nordic—low. Plants were grown outside the greenhouse under direct sun conditions. At the four-leaf stage, pots were sprayed with the use of CO₂-pressurized backpack sprayer with phenmedipham at 0.55 kg ha⁻¹, or left not sprayed. After treatment, half of the plants were moved inside a screen house covered with shade cloth (half sunlight), and the other half remained nonshaded outside the screen house (full sunlight). To evaluate the effect of shade cloth on light intensity, measurements were taken three times inside and outside the screen house with the use of a Li-170 light meter (LI-COR Inc., Lincoln, NE 68504). Measurements were taken under clear sky conditions between 1100 and 1300. To evaluate the impact of the shade cloth on temperatures, air temperature was measured every minute inside and outside of the screen house with the use of Hobo H8 Pro Series temperature data loggers (Onset Computers Corporation, Pocasset, MA 02532).

The shade sensitivity experiment included two evaluations: PS II electron transfer and dry weight, which were taken with the use of two different sets of plants. Chlorophyll fluorescence measurements were taken on dark-adapted leaves (30 min dark adapted by covering the leaves with a clip) with the use of a portable chlorophyll fluorometer (Photosynthesis Yield Analyzer Mini-PAM, Walz, Effeltrich, Germany 91090). Measurements were taken from the second leaf (from the bottom) at 1, 2, 3, and 8 d after treatment (DAT). The basic fluorescence parameters measured were F_m —maximum fluorescence in a dark-adapted state, and F_0 —minimal fluorescence in a dark-adapted state. Then, the maximum photochemical potential of PSII (F_v/F_m) was calculated using the formula: $F_v/F_m = (F_m - F_0)/F_m$ (Genty et al. 1989). For the dry-weight evaluation, pots were seeded, treated with phenmedipham at 0.55 kg ha⁻¹ or not sprayed, and grown with and without shade, as previously

described. Ten days after treatment the aboveground parts of the plants were harvested, dried at 80 C for 6 d, and their dry weights recorded. The experiment was conducted twice in September 2014. Average temperature and photoperiod were 17.5 C and approximately 12.5/11.5 day/night hours.

Application Time of Day. To evaluate the effect of application time of day on phenmedipham weed control, 30 pots (8-cm diameter) were filled with soil and seeded with burning nettle seeds that were collected from fields at Salinas, CA, dried in the greenhouse for 3 mo and kept at room temperature until use. At the four-leaf stage, plants were sprayed with the use of CO₂-pressurized backpack sprayer on different timings, morning (0900) and evening (1800), with phenmedipham at 0.27 and 0.55 kg ha⁻¹, or left not sprayed. All plants were grown outside the greenhouse under direct sunlight conditions. Ten days after treatment the aboveground parts of the plants were harvested and fresh biomass was recorded. Control efficacy (CE) was evaluated by

$$CE = 100 - [(I/I_0) \times 100] \quad [1]$$

where I and I_0 are the fresh biomass values of the treated and untreated plants, respectively.

Statistical Analysis for Greenhouse Studies. Analysis was conducted with the use of SAS (Statistical Analysis Systems, version 9.3, SAS Institute Inc., Cary, NC 27513). Experiments were conducted twice and arranged in a completely randomized design with five replicates, with the exception of the shade sensitivity study, which was replicated 10 times. There were no experiment by treatment interactions; therefore, data were pooled. For the screening experiment, dry weights of each variety were subjected to ANOVA (Tukey-Kramer honestly significant difference [HSD] test, $P = 0.05$), and one-way analysis of variance was conducted with the use of PROC GLM to determine the impact of variety on the dry weights of treated plants. For the shade sensitivity experiment two analyses were held. To evaluate the impact of shade on spinach tolerance to phenmedipham, the 95% confidence intervals (CI) of Regal and Nordic dry weights were calculated separately with the use of the CLM option in the MEANS procedure. Then, factorial analysis was held (on each evaluation timing) with the use of PROC GLM, for the shade and no-shade conditions separately, to determine the interaction between phenmedipham rates and spinach variety on F_v/F_m values. For the

Table 1. Year, location, planting date, phenmedipham (0.55 kg ai ha⁻¹) treatment dates, and average light and temperature values taken 3 and 21 d after treatments, in the field experiments (Exp.).

Exp.	Year	Location	Planting	Treatment	Light W m ⁻²	Temperature C
1	2013	Hartnell	21 May	11 June	322	16.0
2	2013	Hartnell	30 July	15 August	296	16.1
3	2013	Hartnell	9 September	21 October	195	12.8
4	2014	Spence	20 April	12 May	326	16.0
5	2014	Spence	1 May	19 May	244	14.7

application time of day experiment, CE was subjected to ANOVA (Tukey-Kramer HSD test $P = 0.05$), and full factorial analysis was conducted with the use of PROC GLM to determine the interaction between phenmedipham rates and application time on burning nettle control.

Field Studies. The field studies were conducted to evaluate the impact of light on the tolerance of fresh spinach to phenmedipham. Trials were conducted during 2013 and 2014 on Hartnell and Spence field stations near Salinas, CA. Soil type at Hartnell station is an Antioch sandy loam, fine, smectitic, thermic Typic Natrixeralf (53% sand, 32% silt, and 15% clay) with a pH of 7.0 and organic matter content of 2.1%. Soil type at Spence station is a Chualar loam, fine-loamy, mixed, thermic Typic Argixeroll (79% sand, 14% silt, and 7% clay) with a pH of 7.2 and organic matter content of 1%. Temperatures and light values were recorded hourly from the California Irrigation Management Information System (CIMIS) weather station network (with the use of data from the weather station nearest the test site) throughout the study. Table 1 lists the relevant data for the field experiments, which include year, location, planting date, treatment date, and light and temperature conditions.

All plantings were grown on raised 1-m-wide beds in plots 6 m long with two seed lines per bed, 30 cm apart. One seed line was planted with Nordic and the other with Regal. A tractor-mounted planter (Stanhay Webb Ltd., Grantham, U.K.) was used for planting, and overhead sprinkler irrigation and other common spinach cultural practices were used (LeStrange et al. 2013). Phenmedipham, 0.55 kg ha⁻¹, was applied to four-leaf spinach with a CO₂-pressurized backpack sprayer as described in the previous section. Nontreated (hand-weeded) controls were maintained to estimate the level of yield loss due to herbicide treatment. Spinach yield was evaluated 21 DAT (17 in experiment 1) by harvesting a 3-m section sample area from the center of each plot. The trial was repeated five times (Table 1).

Each experiment was arranged in a randomized complete block design with four replications.

Statistical Analysis. Analysis for the field studies was conducted with SAS. Yield data (percent of control) were plotted against average light and temperature measurements taken 3 and 21 DAT, respectively. Correlations between light, temperature, and yield were analyzed with Pearson product-moment correlations (Kluth et al. 2005) with the use of PROC CORR, followed by a linear regression with PROC REG.

Results and Discussion

Greenhouse Studies. Fresh spinach tolerance to phenmedipham differed by variety, and dry weights of treated plants ranged from 40 to 78% compared to the control (Figure 1). The dry weights of the less tolerant varieties, Tye and Nordic, were approximately half that of Santorini and Regal, the most tolerant varieties (Figure 1). These results indicate that there is a range in tolerance of fresh spinach varieties to phenmedipham. These findings can be

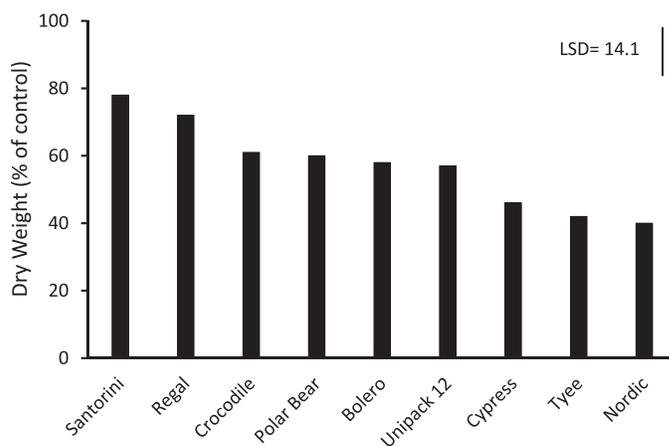


Figure 1. The impact of phenmedipham treatment (0.55 kg ha⁻¹) on the dry weights of nine fresh spinach varieties evaluated 10 d after treatment, in the greenhouse study. Vertical bar represents the least significant difference (LSD) value between varieties dry weight ($n = 10$; $P = 0.05$).

utilized to improve weed control programs in this crop (Leon et al. 2014). Fresh spinach does not have POST herbicides for broadleaf weeds, and the agrichemical industry has no incentive in developing or registering new ones because of spinach's small hectare and high value (Fennimore and Doohan 2008). Identifying varieties with a high tolerance level for nonlabeled herbicides may offer new herbicide options in the tool set available for weed management in fresh spinach (Leon and Tillman 2015). If necessary, registration of this new herbicide can be variety-specific (O'Sullivan et al. 2002). Because phenmedipham and cycloate have different modes of action (MOAs), a sequential application of these herbicides can increase the spectrum and efficacy of controlled weeds and allow MOA rotation (Lati et al. 2015). Furthermore, data about genetic diversity in the tolerance level for phenmedipham (or any other herbicide) across varieties can also be useful for plant breeders (Leon and Tillman 2015). Considering herbicide tolerance early in the selection process of new varieties can lead to high tolerance levels in the end of the process, which in turn may enable herbicide applications at higher rates and better weed control.

The 22% dry weight reduction of Santorini and Regal treated with phenmedipham should not discourage future development of phenmedipham-based programs for fresh spinach. Fennimore et al. (2001) reported that significant yield reductions in fresh spinach resulted from cycloate applied under high temperature conditions. In addition, this study included full phenmedipham rate (0.55 kg ha^{-1}), whereas Lati et al. (2015) used a lower rate (0.27 kg ha^{-1}) following cycloate PRE, which was effective and did not cause spinach injury or yield reduction.

The shade sensitivity experiment revealed differences between varieties and shading conditions. Shade had no impact on the nontreated plants, and for both varieties there was no difference between the dry weights of plants grown with and without shade; dry weight of nontreated Nordic grown with and without the shade was 0.32 ± 0.03 and 0.33 ± 0.03 , respectively (Table 2). Shade had an impact on the phenmedipham-treated plants, but the response differed by variety. For Regal, means and 95% CI of treated plants grown with and without shade were 0.26 ± 0.03 and 0.27 ± 0.04 g, respectively (Table 2). The complete overlap between means and 95% CI of these treatments suggest that Regal is tolerant to phenmedipham under a wider spectrum of shade conditions. For Nordic, means and 95% CI of treated plants grown with and without shade did not overlap and reached values of

Table 2. Dry weight mean and 95% confidence interval (CI) of 'Regal' and 'Nordic' treated or not treated with phenmedipham and grown with or without shade.

Variety	Phenmedipham		Mean	95% CI	
	rate	Shade			
	kg ai ha^{-1}		g plant^{-1}		
Regal	0	With	0.37	0.33	0.41
	0	Without	0.36	0.32	0.40
	0.55	With	0.26	0.23	0.29
	0.55	Without	0.27	0.24	0.30
Nordic	0	With	0.32	0.29	0.35
	0	Without	0.33	0.29	0.36
	0.55	With	0.20	0.17	0.23
	0.55	Without	0.07	0.17	0.23

0.20 ± 0.03 and 0.07 ± 0.03 g, respectively (Table 2). According to Cumming (2009), lack of overlap between the 95% CI arms of different treatments indicates that differences were highly significant ($P < 0.01$). These results indicate that Nordic tolerance to phenmedipham was affected by shade, and that there was greater injury under no-shade conditions.

The light and temperature measurements in the shade sensitivity experiments showed that the shade cloth reduced light to 50% of full sunlight, but reduced the average daily temperature just by 0.5 C ($\sim 3\%$). Based on these environmental measurements, the high proximity (2 m) between plants growing with and without shade and the small size of the screen house (1 by 1.5 m), it can be assumed that light intensity was the major environmental factor affecting the tolerance of Nordic to phenmedipham. These findings are in agreement with Brain et al. (2012) and Fufezan et al. (2002), who evaluated the impact of light intensity on the selectivity of PS II inhibitors, bromoxynil and atrazine, and found them to be more injurious under high light intensities. Phytotoxic effects of PS II inhibitors in plants have been attributed to the formation of ROS, which are promoted under high light intensities (Hess 2000).

Phenmedipham effect on Regal and Nordic was also monitored by measure of chlorophyll fluorescence. The F_v/F_m parameter indicates PS II function, a component of the photosynthetic system that is most sensitive to PS II inhibitors (Abbaspoor and Streibig 2005). The F_v/F_m has a constant value of 830 in healthy leaves of most species, and any reduction implies sensitivity to the herbicide, although tolerance can be noticed via the recovery of this value (Follak and Hurlle 2004). Treated Regal had higher

Table 3. Effect of variety and phenmedipham rate on the F_v/F_m values of the spinach varieties 'Regal' and 'Nordic' taken 1 and 8 d after treatment (DAT) in the greenhouse study.^a

Variety	Phenmedipham rate kg ai ha ⁻¹	Fluorescence ^b	
		1 DAT	8 DAT
		F_v/F_m	
No shade			
Regal	0	779 a	857 a
Regal	0.55	656 b	858 a
Nordic	0	805 a	854 a
Nordic	0.55	417 c	138 b
ANOVA			
V		<0.0001	<0.0001
PR		<0.0001	<0.0001
V by PR		<0.0001	<0.0001
Shade			
Regal	0	832 a	861 a
Regal	0.55	769 b	861 a
Nordic	0	828 ab	855 a
Nordic	0.55	580 c	812 b
ANOVA			
V		<0.0001	0.0185
PR		<0.0001	0.0612
V by PR		<0.0001	0.0576

^a Abbreviations: V, variety; PR, phenmedipham rate.

^b Means within a column followed by the same letter within shade and no-shade conditions are not significantly different according to Tukey-Kramer HSD test ($P = 0.05$).

F_v/F_m values than Nordic under both shade and no-shade conditions and at all evaluations, which suggest that Regal is less sensitive to phenmedipham than Nordic (Table 3). The greatest differences were observed 8 DAT under full sunlight conditions (without shade), where F_v/F_m values of Regal and Nordic were 858 and 138, respectively (Table 3). F_v/F_m values of treated Regal increased during the interval between 1 and 8 DAT, regardless of shade conditions (Table 3). For treated Nordic, the trend of F_v/F_m values was the opposite; F_v/F_m of treated plants developed without shade declined from 417 (1 DAT) to 138 (8 DAT), and F_v/F_m of treated plants developed under shade increased from 580 (1 DAT) to 812 (8 DAT) (Table 3).

The fluorescence measurements provided an insight into the influence of shade on the tolerance and ability to recover from phenmedipham by Regal and Nordic. Higher F_v/F_m values suggest that Regal had higher recovery rate than Nordic, which in turn provides greater herbicide tolerance; Regal was able to recover even when plants developed without shading. Nordic recovered only when plants were grown under shade. Under full sunlight (without shade) Nordic PS II function was irreversibly

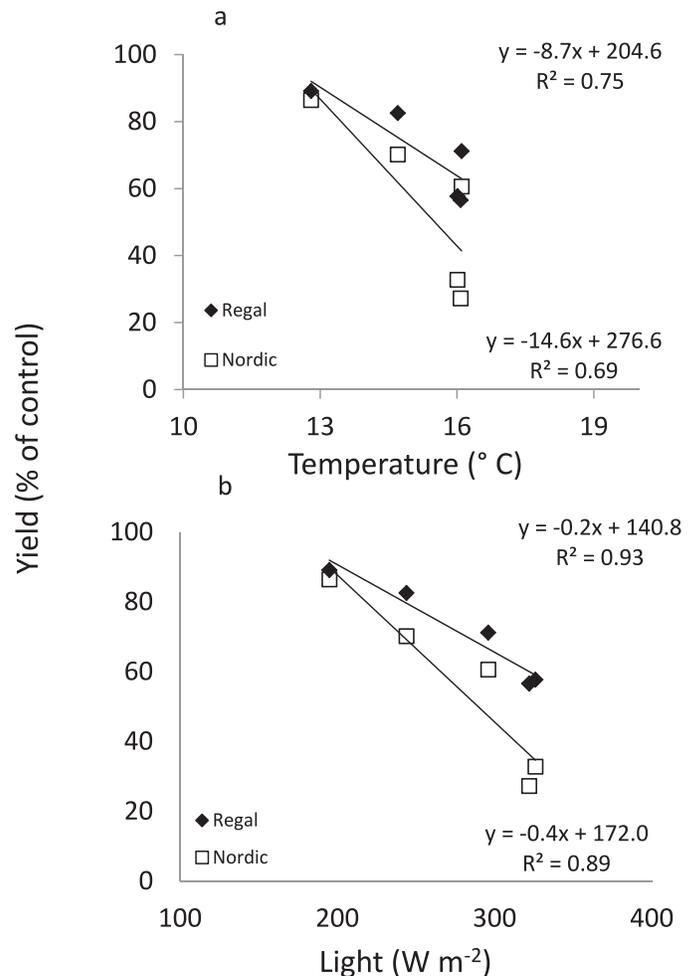


Figure 2. The relationship between 'Regal' and 'Nordic' yield and average temperatures measured over 21 d after treatment (a) or light intensity measured 3 d after treatment (b) in the field study.

damaged and plants did not recover. Abbaspoor and Streibig (2007) used fluorescence measurements to compare recovery from phenmedipham between sugar beet and black nightshade (*Solanum nigrum* L.). These authors used differences in F_v/F_m values between these species as indicators for better recovery rates and tolerance in sugar beet.

Field Studies. In the field, experimental plots could not be covered with shade cloth because of the large area. Therefore, experiments were held under a range of light and temperature conditions, which impacted on Regal and Nordic plants treated with phenmedipham (Table 1). Yield data were regressed against average temperature and light values taken 3 and 21 DAT, respectively. For light, values measured 3 DAT resulted in better fit than values taken 21 DAT. For temperature, 21 DAT measurements resulted in better fit than 3 DAT measurements (data not shown). Both temperature and light had a

Table 4. Effect of phenmedipham rate and application time on burning nettle (*Urtica urens*) control efficacy in the greenhouse study.^a

Application time	Phenmedipham rate	Control efficiency ^{a,b}
	kg ai ha ⁻¹	% of control
9 A.M.	0.27	97
9 A.M.	0.55	96
6 P.M.	0.27	95
6 P.M.	0.55	97
ANOVA		
AT		0.94
PR		<0.0001 ^d
AT by PR		0.98

^a Abbreviations: AT, application time; PR, phenmedipham rate.

^b Means within a column followed by the same letter are not significantly different according to Tukey-Kramer honestly significant difference test ($P = 0.05$).

^c The control efficacy data were based on fresh biomass evaluated 10 d after treatment.

^d Control (phenmedipham at 0 kg ai ha⁻¹) was included in the analysis.

negative relationship with spinach yield (Figure 2). However, in both varieties the relationship between light (3 DAT) and yield was stronger than the relationship between temperature (21 DAT) and yield (Figure 2). For Nordic, the r^2 value was 0.69 and 0.89 for temperature and light, respectively (Figure 2). Additionally, the coefficient of determination resulted from the Pearson product-moment analysis was higher for light and yield ($r^2 = 0.83$, $P = 0.0024$) than for temperature and yield ($r^2 = 0.73$, $P = 0.0148$). These results suggest that light intensity has a greater impact on spinach tolerance to phenmedipham than temperature.

Our findings contrast with prior studies that evaluated phenmedipham control and phytotoxicity, and linked temperature as the main factor affecting its selectivity (Norris 1991; Starke and Renner 1996; Norsworthy and Smith 2005). A possible explanation for that disagreement is that temperature and light are confounded factors, where in this study they were managed to some extent. The spring and summer in Salinas Valley are normally very mild because of the proximity to the ocean, and variations in light intensity are more significant than temperature (Shem-Tov and Fennimore 2003). Experiments 2 and 4 had similar temperatures, whereas light intensity varied by 10%, 31 W m⁻² (Table 1). Correspondingly, Regal and Nordic yields varied by 14 and 28% (significant decrease, data not shown), respectively, between these experiments (Figure 2). Hence, evaluation of the effect of light and

temperature on yield suggested that light is the main factor influencing spinach tolerance to phenmedipham.

The positive impact of low temperatures and light condition on fresh spinach tolerance to phenmedipham (Figure 2) suggests that evening and night applications might be safer for this crop (as Starke and Renner 1996 recommended in sugar beet) and might allow earlier application timing. Still, the weed control efficacy of these applications must be as good as the morning application. Table 4 shows that the evening phenmedipham application was as effective on burning nettle as the morning one, despite the lower light condition in the immediate hours after application (evening and night time). There were no differences in burning nettle control between application timing for both phenmedipham rates. The higher phenmedipham rate (0.55 kg ha⁻¹) is the recommended label rate, and the 0.27–kg ha⁻¹ rate was successfully used by Lati et al. (2015) for control of burning nettle and common purslane. However, data show that both rates were effective and had no interaction with application timing on burning nettle control. Another practical aspect of light intensity and spinach sensitivity for phenmedipham relates to the differences between baby and bunched spinach cropping methods. Light intensity might be less of a factor for phenmedipham injury in baby leaf production than in bunching spinach because of the denser seeding and higher self shading between plants. However, the tolerance level of baby and bunched spinach varieties for phenmedipham was not compared here.

This study demonstrated for the first time significant differences in the tolerances for phenmedipham between fresh spinach varieties. In addition, it demonstrated that light intensity is a major factor that affects the tolerance level of fresh spinach to phenmedipham. By identifying spinach lines highly tolerant to phenmedipham, results here can be useful for future breeding programs of fresh spinach, and for the development of novel phenmedipham-based weed control programs for this crop. New commercial cultivars with high tolerance levels for phenmedipham and successful and safe integration of new POST herbicides into the currently used fresh spinach weed-management programs can reduce hand-weeding costs and allow more efficient production.

Literature Cited

Abbaspoor M, Streibig JC (2005) Clodinafop changes the chlorophyll fluorescence induction curve. *Weed Sci* 53:1–9

- Abbaspoor M, Streibig JC (2007) Monitoring the efficacy and metabolism of phenylcarbamates in sugar beet and black nightshade by chlorophyll fluorescence parameters. *Pest Manag Sci* 63:576–585
- Anonymous (2013) Spin-Aid H specimen Label. <http://www.agrian.com/labelcenter/results.cfm>. Accessed May 9, 2014
- Brain RA, Hoberg J, Hosmer AJ, Wall SB (2012) Influence of light intensity on the toxicity of atrazine to the submerged freshwater aquatic macrophyte *Elodea canadensis*. *Ecotox Environ Safe* 79:55–61.
- [CA-DPR] California Department of Pesticide Regulation. (2013) Summary of Pesticide Use Report Data 2011. <http://www.cdpr.ca.gov/docs/pur11rep/comrpt11.pdf> Accessed June 11, 2014
- Correll JC, Bluhm BH, Feng C, Lamour K, du Toit LJ, Koike ST (2011) Spinach: better management of downy mildew and white rust through genomics. *Eur J Plant Pathol* 129:193–205
- Cumming G (2009) Inference by eye: reading the overlap of independent confidence intervals. *Stat Med* 28:205–220
- Davies HM, Merydith A, Mende-Müller L (1990) Metabolic detoxification of phenmedipham in leaf tissue of tolerant and susceptible species. *Weed Sci* 38:206–214
- Fennimore SA, Doohan DJ (2008) The Challenges of Specialty Crop Weed Control, Future Directions. *Weed Technol* 22:364–372
- Fennimore SA, Smith RF, McGiffen E Jr (2001) Weed management in fresh market spinach (*Spinacia oleracea*) with S-metolachlor. *Weed Technol* 15:511–516
- Follak S, Hurler K (2004) Recovery of non-target plants affected by airborne bromoxynil-octanoate and metribuzin. *Weed Res* 44:142–147
- Fufezan C, Rutherford AW, Liskya AK (2002) Singlet oxygen production in herbicide-treated photosystem II. *FEBS Lett* 532:407–410
- Genty B, Briantais JM, Baker NR (1989) The relationship between the quantum yield of photosynthetic electron-transport and quenching of chlorophyll fluorescence. *Biochim Biophys Acta* 990:87–92
- Hess FD (2000) Light-dependent herbicides: an overview. *Weed Sci* 48:160–170
- Kluth S, Kruess A, Tschardt T (2005) Effects of two pathogens on the performance of *Cirsium arvense* in a successional fallow. *Weed Res* 45:261–269
- Koike ST, Cahn M, Cantwell M, Fennimore SF, LeStrange M, Natwick E, Smith RF, Takele E (2011) Spinach production in California. University of California, Vegetable Research and Information Center. <http://anrcatalog.ucdavis.edu/pdf/7212.pdf>. Accessed May 26, 2015
- LeStrange M, Koike S, Valencia J, Chaney WE (2013) Spinach Production in California. <http://ucanr.edu/repository/fileaccess.cfm?article=54021&p=%20PUIVXP&CFID=6609474&CFTOKEN=39025186>. Accessed April 20, 2014
- Lati RN, Rachuy JS, Fennimore SA (2015) Weed management in fresh market spinach (*Spinacia oleracea*) with phenmedipham and cycloate. *Weed Technol* 29:101–107
- Leon RG, Tillman BL (2015) Postemergence herbicide tolerance variation in peanut germplasm. *Weed Sci* 63:546–554
- Leon RG, Unruh JB, Brecke BJ, Kenworthy KE (2014) Characterization of fluazifop-P-butyl tolerance in zoysiagrass cultivars. *Weed Technol* 28:385–394
- Morelock TE, Correll JC (2008) Spinach breeding. Pages 183–212 in Prohens J, Nuez F, eds. *Vegetables I*. New York: Springer
- Norris RF (1991) Sugarbeet tolerance and weed control efficacy with split applications of phenmedipham and desmedipham. *Weed Res* 31:317–331
- Norsworthy JK, Smith JP (2005) Tolerance of leafy greens to pre-emergence and postemergence herbicides. *Weed Technol* 19:724–730
- O’Sullivan J, Zandstra J, Sikkema P (2002) Sweet corn (*Zea mays*) cultivar sensitivity to mesotrione. *Weed Technol* 16:421–425
- Roberts AG, Gregor W, Britt RD, Kramer DM (2003) Acceptor and donor-side interactions of phenolic inhibitors in photosystem II. *Biochim Biophys Acta* 1604:23–30
- Shem-Tov S, Fennimore SA (2003). Seasonal changes in annual bluegrass (*Poa annua* L.) germinability and emergence. *Weed Sci* 51:690–695
- Smith RF, LeStrange M, Fennimore SA (2013) Integrated weed control in spinach. University of California, Pest Management Guidelines. Division of Agriculture and Natural Resources publication. <http://www.ipm.ucdavis.edu/PMG/r732700111.html>. Accessed May 11, 2014
- Starke RJ, Renner KA (1996) Velvetleaf (*Abutilon theophrasti*) and sugarbeet (*Beta vulgaris*) response to triflurosulfuron and desmedipham plus phenmedipham. *Weed Technol* 10:121–126
- Takele E (2013) Spinach production: sample costs and profitability analysis. University of California. Division of Agriculture and Natural Resources publication. <http://anrcatalog.ucdavis.edu/pdf/8032.pdf>. Accessed June 18, 2014
- [USDA] United States Department of Agriculture (2013) Vegetables: 2012 Summary. <http://usda.mannlib.cornell.edu.80/usda/>. Accessed July 11, 2014
- Wallace RW, Petty AK (2007) Differential response of processing spinach varieties to clopyralid tank-mixes. *Weed Technol* 21:678–682

Received April 8, 2015, and approved July 25, 2015.

Associate Editor for this paper: Muthukumar V. Bagavathiannan, The University of Queensland



Spin-Aid
1.0 Pint/A
morning