# Effect of Preemergence Herbicides on the Cold Hardiness of Container-grown Kurume Hybrid Azalea (*Rhododendron* x *hybrida* 'Tradition')[1]

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## Abstract

Since there is considerable use of preemergence herbicides in the containerized nursery industry, a study was conducted to determine if they affect the ability of plants to cold acclimate. Six preemergence herbicides with different modes-of-action were chosen for the study, and were applied to a Kurume type azalea (*Rhododendron* x *hybrida* 'Tradition') on October 8, 2004. Laboratory cold hardiness evaluations were conducted at 10, 17, and 24 weeks after treatment (WAT) to both stems and leaves. None of the preemergent herbicides tested caused a significant trend in cold acclimation with either stems or leaves when compared over all sampling dates and temperatures. However, some preemergence herbicides caused a significant increase or decrease in cold acclimation of stems and leaves at selected sampling dates and temperatures.

**Index words:** 'Tradition azalea', Kurume hybrid azalea, herbicides, cold acclimation, plant hardiness, dithiopyr, flumioxazin, isoxaben, metolachlor, norflurazon, oryzalin.

**Species used in this study:** *Rhododendron* x *hybrida* 'Tradition'.

**Chemicals used in this study:** Dimension, (dithiopyr), *S,S*-dimethyl 2-(difluoromethyl)-4-(2-methylpropyl)-6-(trifluoromethyl)-3,5-pyridinedicarbothioate; Broadstar, (flumioxazin), 2-[7-fluoro-3,4-dihydro-3-oxo-4-(2-propynyl)-2H-1,4-benzoxazin-6-yl]-4,5,6,7-tetrahydro-1H-isoindole-1,3(2H)-dione; Gallery, (isoxaben), *N*-[3-(1-ethyl-1-methylpropyl)-5-isoxazolyl]-2,6-dimethoxybenzamide; Pennant, (metolachlor), 2-chloro-*N*-(2-ethyl-6-methylphenyl)-*N*-(2-methyloxy-1-methylethyl)acetamide; Predict, (norflurazon), 4-chloro-5-(methylamino)-2-(3-(trifluoromethyl)phenyl)-3(2*H*)-pyridazinone; Surflan, (oryzalin), 4-(dipropylamino)-3,5-dinitrobenzenesulfonamide.

## Significance to the Nursery Industry

Cold damage is a major concern to the nursery industry throughout the United States. Cold injury to woody plants grown in nursery containers is costly (1), and any information that could help prevent cold damage would be welcomed by the industry. Many containerizer nurseries grow plants that are marginally hardy for their area (i.e. Azaleas), and often losses due to cold damage can be substantial. Acclimation of a plant to the cold is a complicated process influenced by many factors. Some of these factors are controllable and include fertility, watering regimes, and pesticide applications. Preemergence herbicides are often applied before plants are moved to over-winter houses, and may contribute to loss of cold hardiness in specific plants. There was no clear trend that the preemergence herbicides tested caused a loss or gain in cold hardiness in stems and leaves of 'Tradition' azalea when compared to the untreated control (UTC). However, some preemergence herbicides caused a significant increase or decrease in cold acclimation of stems and leaves at selected sampling dates and temperatures. More research is needed on the potential of preemergence herbicides, as well as other pesticides, to reduce the cold hardiness of ornamentals.

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## Introduction

Acclimation of a plant to the cold is a complicated process that can be influenced by many factors (2–5). One factor that has not been explored in great detail in the scientific community is the effect of various pesticides on plant cold acclimation. Preemergence herbicides are commonly used in the nursery industry, but the effects on plant cold acclimation are unknown. In the containerized nursery industry, preemergence herbicides are often applied in the fall before plants are placed in overwintering so as to reduce the amount of winter weeds within the containers. During this time period, most plants are beginning the process of cold acclimation. Since cold acclimation involves many metabolic and physical changes within the plant, preemergence herbicide applications may affect these processes. All herbicides have a mode-of-action by which they work to control or disrupt plants processes, and conversely, all plants have a means or mode-of-action by which they attempt to detoxify or eliminate the effect of herbicides. Some of these processes are well understood while others are not (6–7). Many of the preemergence herbicides used in the nursery industry are from families that can inhibit photosystem II, mitosis, cell wall or chlorophyll biosynthesis (Table 1). These systems are also important in cold acclimation and therefore the application of certain preemergence herbicides may affect cold acclimation. This study was designed to determine the effect of common preemergence herbicides on the cold acclimation process in 'Tradition' azalea leaves and stems.

## Materials and Methods

In Griffin, Georgia on July of 2004, six-month-old rooted cuttings of 'Tradition' azalea (*Rhododendron* x *hybrida* 'Tra-

**Table 1. Herbicide and rates used in cold hardiness evaluations of Kurume Azaleas.**

| Trade name | Active ingredient | Chemical family | Mode-of-action | Formulation | Rate kg ai/ha (lbs ai/A) |
|---|---|---|---|---|---|
| Dimension | dithiopyr | Pyridine | Inhibits mitosis | 40 WSP | 0.56 (0.5) |
| Gallery | isoxaben | Benzamide | Inhibits cell wall biosynthesis | 75 DF | 1.12 (1) |
| Surflan | oryzalin | Dinitroaniline | Inhibits microtubule production | 4 AS | 4.48 (4) |
| Pennant | metolachlor | Chloroacetamide | Unclear | 7.62 L | 2.24 (2) |
| BroadStar | flumioxazin | N-phenylphthalimide | Inhibits protoporphyrinogen oxidase | 0.25 GR | 0.42 (0.375) |
| Predict | norflurazon | Pyzidazinone | Inhibits carotenoid biosynthesis | 78.6 WG | 3.36 (3) |
| UTC | | | | | |

dition') were potted into 3.8 liter (1 gal) pots using Fafard 52 potting mix. Azaleas were placed under shade (30% light reduction) and irrigated with approximately 1.75 cm (0.7 in) of water twice daily. On August 18, 2004, plants were top dressed with 18 gm (0.6 oz) of Osmocote Pro 13–10–13. Herbicide treatments were applied on October 1, 2004 (Table 1). One hundred and five containers of 'Tradition' azalea were used. For each treatment, fifteen containers were placed in a 1.8 × 1.8 m (6 × 6 ft) area. Herbicide treatments were applied, and containers were moved to assigned test areas where they were arranged in a randomized complete block (RCB) design. There were 5 replications per treatment, and each treatment contained 3 subsamples. Sprayable herbicides were applied with a $CO_2$ backpack sprayer calibrated to deliver 186.9 liters/ha (20 gal/A). Granular herbicides were applied with a cheese shaker jar[3]. Plants were maintained under shade for the duration of the study.

No visual injury symptoms were noted to any treated azalea leaves or stems at 2, 4, and 8 weeks after treatment (WAT) and the visual herbicide injury ratings were discontinued. Plant samples used for the freeze study were taken at 10, 17, and 24 WAT. Minimum outdoor temperatures for the duration of the experiment are presented in Fig. 1. Laboratory cold hardiness evaluations were performed as described by Lindstrom and Dirr (5) to determine the maximum midwinter hardiness of leaves and stems. Twelve leaves and stem segments of 'Tradtion' azalea from each herbicide treatment were randomly collected and placed into a refrigerator at 2 to 6C (36 to 43F). Three replications of four leaves and stem segments for each herbicide treatment were randomly selected and wrapped in cheesecloth and placed in a test tube (25 × 200 mm). Tubes were submerged in ethylene glycol-water solution (1:1 by vol) in a temperature bath precooled to –2 ± 0.5C (28 ± 0.5F) and held for 14 hours.

Leaf and stem temperatures were measured by thermocouples placed next to the samples and recorded by a data logger. Crushed ice crystals were applied to the wet cheesecloth to insure that the samples did not undercool. Samples were cooled at a rate of not greater than 4C (39F) per hour. Three replications of four leaves and stems of each treatment were removed from the bath at progressively lower 3C (37F) temperature intervals from –3 to –27C (27 to –17F). Controls were prepared and kept at about 4C (39F) for the duration of the freezing test. After being removed from the low temperature treatment, the samples were allowed to thaw overnight at 4 ± 2C (39 ± 2F). Samples were then removed from the tubes and placed in disposable petri dishes containing filter paper saturated with water to maintain 100% relative humidity. Petri dishes were placed on their sides in the dark at 22 ± 2C (72 ± 2F) for 10 to 14 days. Samples were visually evaluated for injury on a 0–3 scale (0 = 0 to 25% brown or damaged tissue, 1 = 26 to 50% brown or damaged tissue, 2 = 51 to 75% brown or damaged tissue, 3 = 76 to 100% brown or damaged tissue). Data were analyzed using analysis of variance and means were exposed to Fisher's least significant difference (LSD) test with a significance level of $\alpha \leq 0.05$. Test for interaction between treatment/temperature and treatment/date were highly significant with both the leaf and stem data ($\alpha < 0.0001$), thus treatment data were presented separately for both temperature and date.

## Results and Discussion

In general, the cold hardiness of leaves and stems increased regardless of preemergence herbicide treatment applied from early fall to midwinter (Tables 2–7). There was no clear trend that any herbicides reduced or increased cold hardiness of either leaves and stems of Kurume Hybrid Azalea (Tables 2–7).

With leaves and stems at all rating dates, little or no injury was found in the treatments as compared to the untreated control (UTC) through exposure to –9C (16F). Only data from –12C to –27C (10 to –17F) is presented and discussed. At 10 WAT, leaves treated with isoxaben and metolachlor showed more injury than 'Tradition' azalea leaves treated with other preemergence herbicides or the UTC. At –15C (5F) no significant difference in treatments was observed. All leaves regardless of treatment were killed at exposure temperatures less than or equal to –18C (–0.4F).

At 17 WAT, no injury occurred in the leaves of any of the treatments when exposed to –12 or –15C (10 to 5F). At –18C (–0.4F) leaves treated with dithiopyr, metolachlor, and

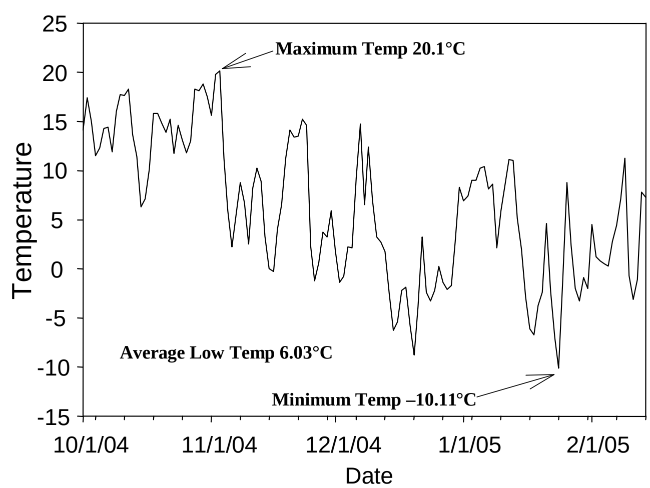


**Fig. 1. Minimum temperatures in Celsius during 2004 experiment.**

[3]Cheese shaker jar. Central Restaurant Products Wholesale Equipment & Supplies, 7750 Georgetown Road, Indianapolis, IN 46268-4135.

**Table 2. Injury rating of *Rhododendron* x *hybrida* 'Tradition' leaves sampled on December 10, 2004 (10 WAT)[z].**

| Trade name | Active ingredient | –12 | –15 | –18 | –21 | –24 | –27 |
|---|---|---|---|---|---|---|---|
| | | Temperature (C) | | | | | |
| | | Injury ratings[y] | | | | | |
| Dimension | dithiopyr | 0.0c | 2.3a | 3.0a | 3.0a | 3.0a | 3.0a |
| Gallery | isoxaben | 2.5a | 3.0a | 3.0a | 3.0a | 3.0a | 3.0a |
| Surflan | oryzalin | 0.0c | 1.8a | 3.0a | 3.0a | 3.0a | 3.0a |
| Pennant | metolachlor | 1.8ab | 2.5a | 3.0a | 3.0a | 3.0a | 3.0a |
| BroadStar | flumioxazin | 1.0bc | 2.0a | 3.0a | 3.0a | 3.0a | 3.0a |
| Predict | norflurazon | 0.5c | 2.5a | 3.0a | 3.0a | 3.0a | 3.0a |
| UTC | | 0.3c | 2.8a | 3.0a | 3.0a | 3.0a | 3.0a |
| LSD | | 1.03 | 1.15 | 0.00 | 0.00 | 0.00 | 0.00 |
| Standard deviation | | 0.71 | 0.79 | 0.00 | 0.00 | 0.00 | 0.00 |

[z]Means within a column followed by the same letter are not significantly different at α = 0.05 as determined by Fisher's Least Significant test.

[y]Cold injury ratings on 0–3 scale, where 0 = no damaged and 3 = dead.

**Table 3. Injury rating of *Rhododendron* x *hybrida* 'Tradition' leaves sampled on January 10, 2005 (17 WAT)[z].**

| Trade name | Active ingredient | –12 | –15 | –18 | –21 | –24 | –27 |
|---|---|---|---|---|---|---|---|
| | | Temperature (C) | | | | | |
| | | Injury ratings[y] | | | | | |
| Dimension | dithiopyr | 0.0a | 0.0a | 1.7a | 2.0a | 2.5b | 3.0a |
| Gallery | isoxaben | 0.0a | 0.0a | 0.0c | 1.5b | 2.3b | 3.0a |
| Surflan | oryzalin | 0.0a | 0.0a | 0.0c | 1.0c | 2.5b | 3.0a |
| Pennant | metolachlor | 0.0a | 0.0a | 1.5a | 2.0a | 3.0a | 3.0a |
| BroadStar | flumioxazin | 0.0a | 0.0a | 1.5a | 1.5b | 3.0a | 3.0a |
| Predict | norflurazon | 0.0a | 0.0a | 0.0c | 2.0a | 3.0a | 3.0a |
| UTC | | 0.0a | 0.0a | 0.5b | 1.5b | 3.0a | 3.0a |
| LSD | | 0.00 | 0.00 | 0.48 | 0.42 | 0.41 | 0.00 |
| Standard deviation | | 0.00 | 0.00 | 0.41 | 0.36 | 0.35 | 0.00 |

[z]Means within a column followed by the same letter are not significantly different at α = 0.05 as determined by Fisher's Least Significant test.

[y]Cold injury ratings on 0–3 scale, where 0 = no damaged and 3 = dead.

**Table 4. Injury rating of *Rhododendron* x *hybrida* 'Tradition' leaves sampled on February 14, 2005 (24 WAT)[z].**

| Trade name | Active ingredient | –12 | –15 | –18 | –21 | –24 | –27 |
|---|---|---|---|---|---|---|---|
| | | Temperature (C) | | | | | |
| | | Injury ratings[y] | | | | | |
| Dimension | dithiopyr | 0.0a | 0.0a | 0.0c | 0.3cd | 0.8d | 3.0a |
| Gallery | isoxaben | 0.0a | 0.0a | 0.0c | 0.0d | 0.3d | 2.0b |
| Surflan | oryzalin | 0.0a | 0.0a | 0.0c | 1.4a | 2.0ab | 1.8bc |
| Pennant | metolachlor | 0.0a | 0.0a | 0.4ab | 0.8bc | 2.2a | 2.8a |
| BroadStar | flumioxazin | 0.0a | 0.0a | 0.2bc | 0.9ab | 1.5bc | 1.6bc |
| Predict | norflurazon | 0.0a | 0.1a | 0.5a | 0.8bc | 2.3a | 2.8a |
| UTC | | 0.0a | 0.0a | 0.1c | 0.3cd | 1.3c | 1.4c |
| LSD | | 0.00 | 0.09 | 0.30 | 0.54 | 0.56 | 0.56 |
| Standard deviation | | 0.00 | 0.08 | 0.25 | 0.46 | 0.47 | 0.48 |

[z]Means within a column followed by the same letter are not significantly different at α = 0.05 as determined by Fisher's Least Significant test

[y]Cold injury ratings on 0–3 scale, where 0 = no damaged and 3 = dead.

**Table 5. Injury rating of *Rhododendron* x *hybrida* 'Tradition' stems sampled on December 10, 2004 (10 WAT)[z].**

| Trade name | Active ingredient | –12 | –15 | –18 | –21 | –24 | –27 |
|---|---|---|---|---|---|---|---|
| | | Temperature (C) | | | | | |
| | | Injury ratings[y] | | | | | |
| Dimension | dithiopyr | 0.0b | 0.5bc | 1.0b | 2.0a | 2.0b | 3.0a |
| Gallery | isoxaben | 0.0b | 2.0a | 1.8a | 2.3a | 3.0a | 3.0a |
| Surflan | oryzalin | 0.8a | 1.0b | 1.0b | 1.3bc | 2.3b | 3.0a |
| Pennant | metolachlor | 0.0b | 1.0b | 1.3ab | 2.0a | 2.0b | 3.0a |
| BroadStar | flumioxazin | 0.0b | 0.0c | 0.0c | 0.0d | 1.5c | 3.0a |
| Predict | norflurazon | 0.0b | 0.0c | 1.0b | 1.8ab | 2.0b | 3.0a |
| UTC | | 0.0b | 1.0b | 1.0b | 1.0c | 2.0b | 2.0b |
| LSD | | 0.26 | 0.53 | 0.57 | 0.63 | 0.40 | 0.00 |
| Standard deviation | | 0.18 | 0.36 | 0.39 | 0.43 | 0.28 | 0.00 |

[z]Means within a column followed by the same letter are not significantly different at α = 0.05 as determined by Fisher's Least Significant test.

[y]Cold injury ratings on 0–3 scale, where 0 = no damaged and 3 = dead.

**Table 6. Injury rating of *Rhododendron* x *hybrida* 'Tradition' stems sampled on January 10, 2005 (17 WAT)[z].**

| Trade name | Active ingredient | –12 | –15 | –18 | –21 | –24 | –27 |
|---|---|---|---|---|---|---|---|
| | | Temperature (C) | | | | | |
| | | Injury ratings[y] | | | | | |
| Dimension | dithiopyr | 0.0a | 0.9a | 0.8a | 1.8a | 3.0a | 3.0a |
| Gallery | isoxaben | 0.0a | 0.0a | 0.0a | 0.0b | 0.0b | 0.0c |
| Surflan | oryzalin | 0.0a | 0.0a | 0.0a | 0.3b | 0.7b | 1.3b |
| Pennant | metolachlor | 0.0a | 0.0a | 0.0a | 0.3b | 0.3b | 0.4c |
| BroadStar | flumioxazin | 0.0a | 0.0a | 0.0a | 0.0b | 0.4b | 0.3c |
| Predict | norflurazon | 0.0a | 0.0a | 0.0a | 0.1b | 0.1b | 0.6c |
| UTC | | 0.0a | 0.0a | 0.0a | 0.1b | 0.2b | 0.3c |
| LSD | | 0.00 | 0.09 | 0.19 | 0.35 | 0.67 | 0.70 |
| Standard deviation | | 0.00 | 0.08 | 0.16 | 0.29 | 0.57 | 0.60 |

[z]Means within a column followed by the same letter are not significantly different at α = 0.05 as determined by Fisher's Least Significant test.

[y]Cold injury ratings on 0–3 scale, where 0 = no damaged and 3 = dead.

**Table 7. Injury rating of *Rhododendron* x *hybrida* 'Tradition' stems sampled on February 14, 2005 (24 WAT)[z].**

| Trade name | Active ingredient | –12 | –15 | –18 | –21 | –24 | –27 |
|---|---|---|---|---|---|---|---|
| | | Temperature (C) | | | | | |
| | | Injury ratings[y] | | | | | |
| Dimension | dithiopyr | 0.0a | 0.9a | 0.0a | 0.0c | 2.0a | 3.0a |
| Gallery | isoxaben | 0.0a | 0.0a | 0.0a | 0.0c | 0.0d | 2.5ab |
| Surflan | oryzalin | 0.0a | 0.0a | 0.0a | 0.4b | 0.8bc | 1.3c |
| Pennant | metolachlor | 0.0a | 0.0a | 0.0a | 0.0c | 0.2d | 0.5d |
| BroadStar | flumioxazin | 0.0a | 0.0a | 0.4a | 0.3bc | 1.3b | 1.3c |
| Predict | norflurazon | 0.0a | 0.0a | 0.1a | 0.3bc | 0.7c | 1.4c |
| UTC | | 0.0a | 0.0a | 0.3a | 0.8a | 1.8a | 2.3b |
| LSD | | 0.00 | 0.09 | 0.36 | 0.41 | 0.43 | 0.57 |
| Standard deviation | | 0.00 | 0.08 | 0.30 | 0.35 | 0.37 | 0.49 |

[z]Means within a column followed by the same letter are not significantly different at α = 0.05 as determined by Fisher's Least Significant test.

[y]Cold injury ratings on 0-3 scale, where 0 = no damaged and 3 = dead.

flumioxazin showed significantly more cold injury compared to the other treatments and the UTC; where as, leaves treated with isoxaben, oryzalin, and norflurazon showed significantly less injury than the UTC (Table 3). At –21C (–5.8F), dithiopyr, metolachlor, and norflurazon expressed more cold injury than the UTC, while oryzalin showed less cold injury than the UTC. At –24C (–11.2F) and below all leaves, regardless of treatment showed severe cold damage.

At 24 WAT, no injury occurred to the leaves on any of the treatments at –12 or –15C (10 to 5F). At –18C (–0.4F) metolachlor and norflurazon showed more injury than leaves of the other treatments and the UTC. At –21C (–5.8F) the oryzalin and flumioxazin treated leaves showed more injury than the UTC and other treated samples. At –24C (–11F) the leaves treated with oryzalin, metolachlor, and norflurazon showed more cold injury than the other treatments and the UTC; whereas, dithiopyr and isoxaben showed less injury compared to the UTC. At –27C (–17F) the dithiopyr, isoxaben, metolachlor, and norflurazon treated leaves showed more cold injury than the other treatments and the UTC (Table 4).

With stems at 10 WAT, very little damage was apparent for any of the treatments at –12C (10F). From exposure temperatures of –15 to –24C (5 to –11F), isoxaben showed significantly more cold injury to stems than the UTC and other treatments except dithiopyr and metolachlor at –21C (–6F). Flumioxazin treated stems from –15 to –24C (5 to –11F) showed less cold injury than the UTC and stems of the other treatments (Table 6). At –27C (–17C) stems treated with all herbicides expressed more injury than the UTC.

At 17 WAT no cold damage was shown when stem segments were exposed from –12 to –18C (10 to –0.4F). Stems treated with dithiopyr showed more cold damage than stems of other treatments and the UTC when exposed to temperatures ranging from –21 to –27C (–6 to –17F). Also at –27C (–17F) stems treated with oryzalin expressed more injury than the UTC.

At 24 WAT, there were no differences among injury ratings from exposure temperatures ranging from –12 to –18C (10 to –0.4F). Stems of all treatments were slightly less injured than the UTC when exposed to a –21C (–6F). At –24C (–11F) all treatments, except dithiopyr, were slightly less injured than the UTC. This same trend was seen at –27C (–17F), as dithiopyr caused more damage to the stems. All other treatments at –27C (–17F) (but isoxaben) were slightly less injured than the UTC (Table 7).

According to these data, there was no clear trend for a single mode-of-action preemergence herbicide to cause a reduction or increase in cold acclimation of leaves or stems. Further research is needed to determine if an interaction with cold acclimation and herbicides exists, and if applications of preemergence herbicides affect the acclimation of select ornamentals.

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