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## BIOENGINEERED CRANBERRIES AND THE FUTURE

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The Agracetus Company  
Dan Mahr, Professor, UW  
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The complex of techniques and science now being lumped under the term 'biotechnology' is at the stage of rapidly being applied to agricultural problems. With the cooperative support of the Wisconsin Cranberry Board, the University of Wisconsin has launched a major program to determine the benefits that biotechnology may offer the cranberry industry. This talk will be a progress report of the first 3 years of what is turning-out to be a most exciting effort.

Two major areas of biotechnology are being applied to cranberry production and improvement. The first is 'micropropagation' and involves the rapid cloning of superior cultivars of cranberry in test-tube type environments. The second is 'genetic engineering' and involves inserting new genes into cranberry that will confer pest resistance. Probably the most instructive format for this presentation is to show how the various technological hurdles were (and are being) overcome with cranberry.

In order to successfully genetically engineer any plant, 4 requirements must be met:

1. Have a useable cell/tissue culture system. Since genes are inserted into individual cells, one must have the capability to grow such cells into whole plants. This is done in sterile, test-tube type environments where the environmental and nutrient conditions surrounding the cells/tissues can be precisely controlled.

2. Have the needed genes identified and isolated. There are three types of genes usually needed for genetic engineering. The marker genes allow one to identify those cells and tissues that have incorporated the new genes and in which these genes are functioning or expressing. Such cells are termed 'transformed'. We use the GUS gene which turns transformed cells blue when put under special conditions. The selection genes give the transformed cells a competitive advantage in comparison to cells not containing the new genes; this allows us to preferentially promote the growth of transformed cells even though they may be grossly outnumbered by other cells in the surrounding tissues. Here we use the KAN gene that makes the cells tolerant of an antibiotic, kanamycin, that usually kills plant cells. Finally, one needs the genes of interest, that is the genes that code for the characteristics that we want to put into cranberry. In this case, we are concentrating on pest resistance being conferred by a BT gene.

3. A method to insert the genes into the cells. Here, we are using a new and patented technology called 'particle bombardment' where the genes piggyback on gold pellets that are "shot" into the cells.

4. Control of gene function while in the plant. Although all the cells in a genetically-engineered plant will contain the new genes, we may not want these genes to be functioning (expressing) all the time and in all the tissues. Here we attach control elements, called promoters, onto each gene. Promoters allow the genes to express only under certain conditions.

We are pleased to say that after only a few years of work with the cranberry, all of these requirements are now in place. We have perfected a tissue culture system that we can use to (1) generate the 'targets' that are most effective in the gene transfer system, (2) differentiate shoots from the transformed cells, and (3) multiply the regenerated shoots so that we can rapidly obtain plants for testing and evaluation. The genes, the gene promoters, the gene transfer technology, and many of the analyses are all being provided by Agracetus Company in Middleton, Wisconsin. The close cooperation of Agracetus is in large part why we have been able to make such rapid progress in this research.

A second cooperative effort with Professor Dan Mahr and his laboratory has allowed us to evaluate the pest resistance of our recovered transformed plants. Such analyses are at this time underway. The advice and cooperation of such cranberry experts as Dan Mahr and Elden Stang as well as enthusiastic cranberry growers is yet another reason why the project has progressed so well. Such continued cooperation is absolutely essential for a project like this to realize its goals.

The second part of the project, that of developing the techniques for micropropagating cranberries and evaluating the response of such plants in the field, is also well underway and showing unusual success. In two years of comparative plots, micropropagated plants have outperformed field cuttings in vigor and establishment. This coming season will test the potential fruit productivity of micropropagated plants.

What are some of the remaining hurdles to be overcome? The scope of this project will continue to enlarge as we get closer to actual release of any genetically-engineered plants and so will the problems. A sampling of such concerns includes:

1. Will the genetically-engineered plants be efficacious, that is, will they control pests in the field? We already have indications that we may have a problem here in that two of our inserted genes, the GUS and the BT, are not expressing well in cranberry. Our current hypothesis is that the promoter driving both of these genes is not functioning well. We are already taking steps to overcome this problem by using other promoters, however this means generating a whole new line of transformed plants and thus will delay our evaluations.

A second aspect of field evaluations is obtaining the permits to establish such plots. Again, the cooperation of growers and industry will be critical in expediting such entanglements.

2. Who will own the plants? Much of the technology and genes that we are now using are proprietary. Agreements will have to be worked-out to address the licensing and payments for use of these technologies. We (myself, Rodney Serres, and Agracetus) have already filed a patent on the cranberry transformation system. We expect to file for plant patents on the final products of this research. Our

intention is to turn-over the rights to these patents to the Wisconsin Cranberry Board and Agracetus so that any royalties or licensing fees can be plowed back into research and technology development in Wisconsin.

3. Will the public accept the product? Our intention is to engineer a plant that will produce cranberry fruits that are chemically and aesthetically indistinguishable from non-engineered fruits. One approach is to use promoters that will function only in green tissues. Thus the leaves and green fruits of the plant will be protected by the gene products, but the ripened fruits will not contain such products. Testing this hypothesis will itself be a fascinating biological research endeavor.

4. What are the environmental concerns? Will there be a problem with 'escape' of the genes to native cranberries? Will the insects develop tolerance to the engineered plants, thus circumventing the whole effort? Will non-target (non-pest) insects be impacted? We are already beginning to determine how such questions should be addressed so that reasonable answers can be obtained.

5. What else can we do to improve the cranberry? Actually, here, there is really no limit. The advances that have been made in this biology over the last few years have been truly revolutionary. Continued and even more rapid progress is anticipated. We view this project as only a beginning in a never-ending application of biotechnology to cranberry development and production.

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In summary, our first 3 years of working on cranberry biotechnology has been incredibly stimulating, exciting, and successful. The cranberry is now the leader in the application of biotechnology to fruits. We hope to continue this effort, again with the close cooperation and advice of the Wisconsin cranberry and biotechnological industries.

## COVERS ON CRANBERRIES: A POSITIVE RESPONSE?

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Use of spunbonded fabrics as covers over actively growing or dormant plants is now commonplace in high-value horticultural crops such as strawberries, ornamentals and tobacco seedbeds. Covers also serve as insect barriers and weed barriers when applied over soil. On strawberry, rowcovers are widely used and are reported to provide increased accumulation of heat units, some protection from frost, earlier flowering, enhanced plant growth and increased yield.

Spunbonded fabrics are continuous, porous sheets created from plastics extruded as tiny fibers. Heat, pressure and chemicals are used to bond the fibers together into strong light sheets of varying thicknesses. Water and air pass freely through the openings in the fabric, thus plants can readily be irrigated without removing the cover. Depending on fabric thickness, soil heat loss is delayed and higher humidity around plants is maintained.

The positive effects of spunbonded covers on other crops suggested a test of covers on cranberries was warranted. An initial test in 1989 at R.S. Brazeau, Inc., Wisconsin Rapids is reported in the attached publication. The trend to increased fruit set and yield with covers in 1989 (Table 1) stimulated our interest in further testing in 1990.

A replicated, more detailed test was initiated at Dubay Cranberries, Inc. on 'Searles' in 1990. Typar covers as described in the 1989 report were applied in mid April after ice melted. Covers were again selectively removed at 3 week intervals up to 21 June. Unfortunately, a severe hail storm on 29 June caused up to 30% losses in final yield. Results of the test were thus complicated by crop injury.

A trend to increased fruit set was again noted in 1990 with covers removed early in the season (Table 2). In 1990, however, yield responses did not parallel the trend in 1989. No clear-cut explanation for this response is apparent at this time. High air temperatures under the covers in late May and June might well have altered plant physiology sufficiently in some way to result in failure of fruit to develop past pinhead and pea stages. Berry weight and anthocyanin content were not influenced by covers in either season.

A strong trend to increased chlorophyll content (Table 3) and increased photosynthesis (PN) in plants under covers is limited evidence covers might alter and enhance earlier carbohydrate synthesis despite shading and reduced light levels (PAR) under the covers.

Despite some conflicting results, cranberry plant responses under early season covers suggest further experimentation is warranted. We recognize of course, at this time no reasonable and feasible means for easily applying and maintaining covers on beds in a commercial cranberry production system exists.

In a different experiment in 1990 designed to evaluate influences of various preplant and postplant treatments on newly established cranberries, Typar spunbonded covers were applied over 'Stevens' immediately after planting. Covers

were kept on the planting through late August. Although some interactions with other factors occurred, main responses on treatments with covers vs no covers generally showed 20% increased upright length and greater bed cover by vines. Additional tests in 1991 are proposed to verify if fabric covers on new beds can consistently enhance bed establishment.

Positive responses with plant covers on other plant species are well documented. Enhancement of early season chlorophyll development, increased plant growth and potential for enhancing fruit set in cranberry with fabric covers should be verified by continued experimentation. Fabric covers appear to offer a viable, non-chemical and renewable means for positively altering cranberry plant growth and fruiting responses in our initial tests.

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## Spunbonded Fabric Covers Suggest Possibilities to Alter Early Season Growth and Fruiting in Cranberry

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Additional index words. *Vaccinium macrocarpon*, rowcovers

Use of spunbonded fabrics as covers to alter plant development, modify microclimate, provide protection from insects and animals, and improve productivity, as highlighted by Wells and Loy (1985), is now commonplace for a host of high-value horticultural crops. In strawberry, rowcovers are reported to provide enhanced accumulation of heat units, limited protection from frost, earlier flowering, increased plant growth, and potential for increased yield (Gast and Pollard, 1988; Pollard and Cundari, 1988).

Evaluation of spunbonded fabric covers on cranberry plant development and fruiting has not been reported and was the objective of this preliminary research. Recent research on fruit set in cranberry indicates high levels of soluble carbohydrates in cranberry shoot upright growth at early blossom stages are important in determining subsequent fruit set (Birrenkott, 1989). Onset of earlier seasonal growth as a result of covers, if concurrent with enhanced photosynthesis and higher carbohydrate levels, might offer potential for increased fruit set and productivity.

Typar spunbonded polypropylene fabric (Reemay, Hickory, Tenn.) was placed over a mature, producing 'Searles' cranberry planting in north-central Wisconsin on 5 Dec. 1988. Typar is a heavy-duty fabric with ultraviolet stabilizers added for resistance to degradation by sunlight. Weight of the sheet is  $70.6 \text{ g} \cdot \text{m}^{-2}$ , with substantial resistance to puncturing or tearing and potential for reuse.

A total area of  $2100 \text{ m}^2$  was covered using four large sheets of Typar. Uncovered con-

trol areas were located adjacent to covered plots in the same cranberry bed. Fabric covers were held in place with 9-mm-diameter steel concrete-reinforcing rods laid along the fabric edges, pinned down with heavy-gauge wire hooks inserted into the soil. Remote thermocouples were tied to stakes at plant height ( $\approx 10 \text{ cm}$ ) under the covers and in control areas for temperature monitoring. Soil temperatures were measured using a Taylor dial probe thermometer inserted to 15-cm depth through the fabric covers.

The bed was flooded for winter protection by 10 Dec. 1988 and a 25-cm-thick ice layer was frozen over control and covered areas. Ice remained throughout Winter 1988-89 until melting and drainage in early April. Individual Typar covers,  $\approx 500 \text{ m}^2$  each, were removed at intervals of  $\approx 3$  weeks, beginning 17 Apr.

The plantings were observed at intervals during the growing season to determine effects of covers on growth and fruiting. Six random samples of  $\approx 50$  upright shoots and four samples of fruit ( $\approx 300 \text{ g}$  each) were taken from each plot on 29 Sept. 1989 for determination of percent fruit set, fruit weight, yield, and anthocyanin content of fruit.

During the cool early spring, canopy air temperatures under spunbonded fabric were 5 to 6°C higher than in exposed control plants. Differences ranged as great as 17°C, with a maximum temperature of 45°C measured under covers with full sunshine in late May and early June. Temperatures suitable for initiation of active metabolism, estimated to be

at 6 to 7°C, resulted in earlier greening of leaf tissue under fabric covers. Nevertheless, differences in recognizable stages of current season's growth and flower development were negligible. Despite earlier greening, lack of visible earlier elongation may in part be related to a negligible effect of the covers on soil temperature (Table 1). The cranberry plant canopy appears to provide an insulating effect on soil temperature at least during early spring. However, soil temperature began to increase substantially by late May.

Although no significant differences existed, a trend to increased fruit set was apparent when plant covers remained in place until late May and early June (Table 1). This trend also was noted in samples taken solely for fruit set determination in early September (data not shown). However, no clear trend for increased yield was evident in samples taken at normal harvest. Anthocyanin content of fruit at harvest varied, but did not appear to be related to the time of cover removal.

Current evidence for the positive influence of soluble carbohydrates at early blossom on fruit set in cranberry (Birrenkott, 1989) suggests that earlier leaf greening, if concomitant with enhanced soluble carbohydrate production, could enhance subsequent fruit set. Evidence in this study is not adequate to warrant a conclusion that spunbonded fabric covers either provided earlier enhanced carbohydrate levels or enhanced fruit set.

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Table 1. Soil temperature and fruit characteristics in 'Searles' cranberry as influenced by date of removal of Typar polypropylene fabric cover.

Date of cover removal	Soil temp (°C)			Fruit set (%)	Yield <sup>a</sup> (g/81 cm <sup>2</sup> )	Anthocyanin <sup>b</sup> (mg/100 g)
	2 May	23 May	6 June			
17 Apr.	2.3	12.4	15.3	35.8	37.8	20.5 c
2 May	1.3	12.5	14.2	34.4	31.7	17.5 a
23 May	1.7	12.5	15.1	42.0	43.2	19.3 b
6 June	3.3	13.7	17.6	42.6	37.2	21.5 c
No cover	2.5	11.4	14.0	39.6	41.4	21.0 c
				NS	NS	

<sup>a</sup>Adjusted yield using number of flowering uprights as a covariate.

<sup>b</sup>Mean separation at  $P = 0.05$ , Fisher's protected LSD; NS, nonsignificant.

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Table 2. Flowering and yield, 1990.

Cover removal date	No. flowering shoots <sup>a</sup>	% Fruit set	Yield(g) <sup>a</sup>	Mean berry weight(g)	Anthocyanin (mg 100g <sup>-1</sup> )
No cover (control)	16.4a	22.5b	11.7a	1.12	23.1
9 May	12.0b	29.2a	9.5a	1.11	21.2
30 May	10.5b	23.7ab	6.4b	1.11	23.6
21 June	10.4b	17.7b	5.1b	1.10	22.0
	*	*	*	NS	NS

<sup>a</sup>Sample size reduced in 1990 vs. 1989.

Table 3. Photosynthesis and chlorophyll, 1990.

Cover removal date	Photosynthesis PN ( $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ )		Light PAR ( $\mu\text{mol}\cdot\text{s}^{-1}\cdot\text{m}^{-2}$ )		Chlorophyll ( $\mu\text{g}\cdot\text{g}^{-1}$ FW)
No cover (control)	<u>30 May</u>	<u>21 June</u>	<u>30 May</u>	<u>21 June</u>	
	2.38	2.07	1931a	1773a	.250
9 May	1.44	2.73	1975a	1798a	.236
30 May	2.44	2.79	1973a	1535a	.304
21 June	1.48	3.00	1257b	988b	.417
	NS	NS	*	*	NS

## RESEARCH REPORT: MANAGING CRANBERRY COTTONBALL WITH FUNGICIDES

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### INTRODUCTION

Cottonball, caused by *Monilinia oxycocci*, is the most economically important disease affecting cranberries grown in Wisconsin. This disease has two distinct stages, which differ both in the type of inoculum (i.e., spore) causing infection and the portion of the plant infected. Primary infection by ascospores causes tip blight, which affects young upright shoots (primarily), flower pedicels, and flowers. A layer of fungus tissue (gray to white in color) bearing conidia eventually forms on blighted organs, and these conidia cause secondary infection of flowers that results in cottonball fruit rot later in the season. In North America, cottonball is economically important only in Wisconsin and British Columbia although losses from the disease have occurred recently in Ontario, Canada as well. The economic importance of cottonball in Wisconsin has increased considerably during the past 10-15 years.

Currently, triforine (FUNGINEX) is the only fungicide registered for cottonball management in Wisconsin, but control in the field has not been consistent. Additional fungicides that provide alternatives to triforine and that may improve disease management are needed. Only limited research on chemical control of cottonball has been conducted previously, in the 1920's by H. F. Bain in Washington and in the 1970's by H. S. Pepin in British Columbia. This previous research and that on other diseases caused by different species of *Monilinia* suggested candidate fungicides for evaluation. The accurate timing of fungicide applications is essential to optimize the effectiveness of the chemicals applied. Ideally, fungicides should be applied only when infection is likely to occur, that is, during "infection periods". Infection periods are times when inoculum of the pathogen is present, environmental conditions are favorable for infection, and host plants are susceptible to infection.

The objectives of our research projects were: 1) to evaluate fungicides for efficacy in managing cranberry cottonball under Wisconsin field conditions and 2) to identify infection periods so that fungicide applications could be scheduled more accurately.

### EXPERIMENTAL METHODS

Experiments were conducted from 1987 to 1989 in a commercial cranberry bed (cultivar Bain McFarlin) that was over 20-yr-old and heavily infested with *M. oxycocci*. The bed was located in the southwestern portion of Wood County. All research was conducted in the southern section of the bed where disease severity typically was greatest. In all years, regular fungicide applications were withheld from the portion of the bed in which research was conducted.

## Evaluation of Candidate Fungicides

Fungicides were evaluated in 1987, 1988, and 1989. Each year, 10 treatments were evaluated--nine fungicides and an untreated control. Table 1 lists the fungicides evaluated each year with the formulations and rates of application used. In all, 11 fungicides were evaluated over the 3-yr period. Each fungicide was evaluated in at least two years except for myclobutanil and thiophanate-methyl, which were tested only the first year. Fungicides were used at the high end of the range of rates suggested by the manufacturers or registered on other crops for related diseases to optimize the opportunity for efficacy. In addition, in 1989, several of the most promising fungicides also were evaluated at reduced rates. Fungicides were applied to plots at a rate equivalent to 160 gallons of water per acre by a CO<sub>2</sub>-powered sprayer equipped with a single boom and a hollow-cone nozzle.

Three applications beginning at budbreak and then at 7- to 9-day intervals were made to protect plants from primary infection by ascospores (Table 2), which is one more application than is applied commercially. Two applications to protect flowers from secondary infection by conidia were applied 7 to 10 days apart with applications beginning progressively earlier in relation to bloom each year (Table 2). In each year, a total of five applications were made to manage both stages of disease development. Primary infection was assessed during bloom, and secondary infection was assessed at the end of the growing season prior to harvest.

## Identification of Infection Periods

The presence of airborne inocula (spores) was monitored with a Burkard 7-day recording volumetric spore trap. In 1987 and 1988, the trap was placed in the field immediately after the last spring flood waters were removed (17 and 29 April, respectively) and operated continuously until 29 and 28 July, respectively (10-12 days after bloom had ended). To determine if any ascospores were coming from outside the study area prior to removal of the spring flood, the spore trap was placed in the field on 12 April in 1989 and mounted on a platform just above the water surface. After the flood was removed from the field (16 May), the spore trap was placed back in the bed and run continuously until 2 August, approximately 12 days after bloom had ended. Environmental parameters were recorded with a Campbell 21X micrologger. Parameters measured were temperatures of the soil, duff, upper plant canopy, and ambient air; percent relative humidity; wind speed; rain and irrigation; and leaf wetness. A hygrothermograph was operated to verify and backup the micrologger.

Observations on cranberry growth and development and disease progress were recorded at regular intervals each year. Budbreak was defined to be when greater than 50% of shoots had begun to elongate. Stages of early season shoot development were divided into three classes based on shoot length and corresponding morphological changes. Percent bloom was the percentage of flowering upright shoots with open blossoms. Samples of fruit were collected and examined weekly from the end of bloom until harvest to determine if incidence of fruit rot increased after bloom.

## RESULTS

### Evaluation of Candidate Fungicides

Triforine, RH-7592, and terbutrazole consistently provided the best protection from primary infection; vinclozolin and RH-3486 also controlled primary infection but not as effectively. In 1989, the reduced rate of terbutrazole was as effective as the full rate at controlling tip blight. Chlorothalonil and benomyl provided moderate control whereas control by iprodione, copper hydroxide, thiophanate-methyl, myclobutanil, and the reduced rate of benomyl was inadequate. In managing cottonball fruit rot caused by secondary infection of flowers by conidia, benomyl, triforine, terbutrazole, and chlorothalonil consistently provided the best control over the 3-yr period. In 1989, the reduced rates of terbutrazole and chlorothalonil gave protection similar to their corresponding full rates; however, the reduced rate of benomyl was significantly less effective than its full rate. In addition to these four fungicides, RH-7592, RH-3486, and vinclozolin also provided effective control of secondary infections; but iprodione, thiophanate-methyl, myclobutanil, and copper hydroxide offered little or no protection.

In both 1987 and 1988 yield was greatest in plots treated with benomyl and was reduced in those treated with copper hydroxide or left untreated. These yield reductions likely were due to the abundance of cottonball fruit rot that occurred. However, in 1988, plots treated with chlorothalonil also had significantly reduced yield (i.e., a reduction of 49% compared to benomyl treated plots) but one of the lowest amounts of fruit rot (2.4%). Although there was no significant treatment effect in 1989, plots treated with either rate of benomyl again were among the highest yielding and those treated with chlorothalonil at either the full or the reduced rate were the lowest yielding despite again having some of the lowest amounts of fruit rot.

Individual berry weight (i.e., berry size) was affected by fungicide treatments in two of the three years. Compared to the treatment that produced berries with the greatest weight, only chlorothalonil reduced berry weight in both 1988 and 1989 although terbutrazole, triforine, vinclozolin, or the untreated control also reduced berry weight in one of the years. In addition, chlorothalonil and copper hydroxide reduced fruit retention in 1988.

### Identification of Infection Periods

Similar patterns of spore dispersal were observed each year. Ascospores were collected in a single spore shower that lasted 31 days in 1987, 35 days in 1988, and 25 days in 1989. In each year, a single, distinct peak in the shower occurred that lasted 11, 12, and 10 days in 1987, 1988, and 1989, respectively. Conidia also were collected in a single spore shower each year, which lasted 30 days in 1987, 26 days in 1988, and 33 days in 1989. Similar to the ascospore shower, there was a single conidium peak that lasted 13, 15, and 14 days in 1987, 1988, and 1989, respectively. In 1987 and 1988, the time from the beginning of the ascospore peak to the beginning of the conidium peak was 33 days; in 1989, it was 32 days.

The pattern of ascospores caught over a 24-hr period showed a distinct diurnal periodicity with most spores collected between 11:00 AM and 9:00 PM; the maximum spore catch occurred between 5:00-6:00 PM. Catches of conidia also exhibited diurnal periodicity although less pronounced than that for ascospores. Most conidia were caught during

daylight hours, and the greatest numbers of spores were caught between 11:00 AM and 6:00 PM.

Of the environmental variables measured, duff and canopy temperatures and percent relative humidity correlated best with hourly catches of both ascospores and conidia during peak periods of dispersal; leaf wetness and wind speed correlated less, and rain/irrigation correlated poorly. More ascospores were caught during dry periods than during wet periods (i.e., from leaf wetness or rain/irrigation) in each year. A similar relationship was found between numbers of conidia and periods of leaf wetness; in contrast, however, more conidia usually were trapped during periods of rain/irrigation than during dry periods.

Dispersal of both ascospores and conidia was closely associated with cranberry growth and development. The peak of the ascospore shower occurred around budbreak, and the peak of the conidium shower occurred during bloom. The median number of conidia caught (one-half of the accumulated total) coincided with peak bloom in both 1988 and 1989. In 1988 and 1989, 37% and 33%, respectively, of the fruit collected on all sampling dates combined was diseased, and there was no significant difference among the percentages of diseased fruit at each sample date in either year.

## CONCLUSIONS

Many of the fungicides managed cottonball effectively. Triforine, terbutrazole, and RH-7592 were most effective at controlling both tip blight and fruit rot. Vinclozolin and RH-3486 consistently managed both disease stages but less effectively. Of these, only triforine and vinclozolin currently are registered for use on agricultural crops; the other three are still experimental compounds. Benomyl and chlorothalonil also were very effective at inhibiting secondary infection of flowers by conidia but were not effective at limiting primary infection of shoots by ascospores.

Over the 3-yr period, plots treated with benomyl consistently had the highest yields whereas those treated with chlorothalonil had the lowest yields. In 1988, chlorothalonil treated plots had reduced yields despite having only 2.4% fruit rot; in fact, yield was comparable to that in the untreated plots, which had 30.5% rot. Consequently, reduced yields from chlorothalonil could not be attributed to cottonball fruit rot but were likely due to reductions in berry weight and fruit retention. Berry weight also was reduced by both rates of chlorothalonil in 1989. These data confirm that this fungicide should not be applied to cranberry during bloom in Wisconsin.

An appropriate time to begin fungicide applications to control fruit rot is estimated to be around 5-20% bloom and may depend on the fungicide being used. The benefit of making a third application during bloom should be investigated as fruit rot was not eliminated by any treatment in any year. A third application would be justified economically since a yield reduction of only 1% can represent a significant economic loss.

Currently only triforine is registered for cottonball management in Wisconsin and this is by a Special Local Needs (24[c]) registration that expires at the end of 1991. If cottonball in Wisconsin is to be managed effectively in future years, it is imperative that a permanent federal label for triforine be obtained, that registrations for benomyl and vinclozolin be sought, and that manufacturers of the experimental fungicides terbutrazole and RH-7592 be encouraged to pursue cranberry labels for these compounds at the earliest possible dates.

Ascospores and conidia of *M. oxycocci* were dispersed in single showers, and each shower had a discrete period of peak abundance. No clear pattern of environmental events was observed to account for the initiation of these peak periods in any year; however, once spore showers had begun, they were essentially continuous until the shower ceased. Conidium peaks began 32-33 days after the initiation of ascospore peaks.

The seasonal occurrence of inoculum of *M. oxycocci* was closely tied to cranberry growth and development. Peak ascospore dispersal occurred when shoots were 1-3 cm in length, and shoots appeared to be most susceptible to infection around this time. Peak conidium dispersal occurred around peak bloom. Fruit rot incidence did not increase after bloom, which indicated that the fungus did not colonize fruit that escaped infection during bloom.

In Wisconsin, cottonball infection periods were single continuous events that occurred during the 10- to 14-day periods of peak spore dispersal. Cottonball should be managed effectively with the four applications of triforine that currently are recommended by accurately timing applications to plant growth stages. The intervals of time when inocula and susceptible organs were present were relatively narrow, and successful disease management with fungicides would require that applications be scheduled to adequately cover these intervals. Consequently, triforine (or other fungicides when they become available) should be applied at:

- budbreak (i.e., when greater than 50% of the shoots have begun to elongate) and then 7-10 days later to control tip blight, and
- early bloom (5-20%) and then 7-10 days later to control fruit rot.

*Note: This report is a summary of two research projects that have been submitted for publication by the authors. Copies of these publications, which contain additional information and details, are available upon request from the senior author.*

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**Table 1.** Fungicides and application rates evaluated over a 3-yr period for management of cranberry cottonball caused by *Monilinia oxycocci*.

Common or code name	Trade name	Formulation	Years evaluated	Amount per acre <sup>a</sup>	
				A.I.	Product
Benomyl	Benlate	50DF	1987, 1988, 1989	454 g	2 lb
			1989	227 g	1 lb
Chlorothalonil	Bravo 720	6F	1988, 1989	2041 g	6 pt
			1989	1361 g	4 pt
Copper hydroxide	Kocide 101	77WP	1987, 1988	2794 g	8 lb
Iprodione	Rovral	50WP	1987, 1988	454 g	2 lb
Myclobutanil	Nova	60DF	1987	57 g	3.3 oz
RH-7592 <sup>b</sup>		2F	1987, 1988, 1989	28 g	4 fl oz
		50WP	1987, 1988	454 g	2 lb
Terbutrazole	Elite	45DF	1988, 1989	102 g	8 oz
			1989	51 g	4 oz
Thiophanate-methyl	Topsin-M	70WP	1987	635 g	2 lb
Triforine	Funginex	1.6EC	1987, 1988, 1989	136 g	24 fl oz
Vinclozolin	Ronilan	50WP, 50DF <sup>c</sup>	1987, 1988, 1989	454 g	2 lb

a. Amounts are for active ingredient (A.I.) and formulated product (Product).  
b. Numbered experimental fungicides with no common or trade names assigned.  
c. 50WP was used in 1987 and 1988; 50DF was used in 1989.

**Table 2.** Dates fungicides were applied to replicated plots to manage primary or secondary infection by naturally occurring *Monilinia oxycocci*, intervals between applications, and associated stages of plant development in three consecutive years.

1987			1988			1989		
Date	Interval (days)	Stage <sup>a</sup>	Date	Interval (days)	Stage <sup>a</sup>	Date	Interval (days)	Stage <sup>a</sup>
<b>Primary Infection</b>								
12 May 12	budbreak	19 May	budbreak	25 May		budbreak		
20 May 8	shoot growth	26 May 7	shoot growth	1 June 7		shoot growth		
29 May 9	hook	3 June 8	hook	8 June 7		hook		
<b>Secondary Infection</b>								
16 June 18	50% bloom	15 June 12	29% bloom	23 June 15		6% bloom		
24 June 8	late bloom	22 June 7	61% bloom	3 July 10		49% bloom		

a. Stages of plant development were: budbreak -- >50% shoots beginning to elongate; shoot growth -- >50% shoots actively growing; hook -- unopened flowers present on hooked pedicels; % bloom -- mean percent of flowers open; late bloom -- after full bloom, berries beginning to set.

## BIENNIAL BEARING IN CRANBERRY

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 Arthur P. Poole<sup>2</sup>

Biennial bearing has been observed in cranberry for many years. Various studies have shown a percent return bloom ranging from 12% to 65% depending on year, bed vigor, and cultivar. Significant region to region differences have been noted. This research was undertaken to determine the extent of biennial bearing by cranberry cultivar and growing region. The study was conducted in four major cranberry growing areas with four cultivars per area. All sites had Stevens, Ben Lear, and Crowley. In addition, Massachusetts and New Jersey examined Early Black and Howes, Wisconsin examined Searles, and McFarlin was included in Oregon.

In the fall or winter of 1989/1990 six transects were established within a single bed for each cultivar in each region. In MA, WI and NJ uprights were tagged in the fall after harvest, but before the winter flood. The fruiting characteristic was based on the presence of persistent pedicels. In Oregon, uprights were tagged in midwinter in the absence of a winter flood. For each transect 60 to 100 uprights that had produced fruit in 1989 were tagged with a small piece of vinyl tape. After fruit set in 1990, fifty tagged uprights per transect were examined for the presence of flowers for a second year (return bloom) or fruit a second year (return fruit).

Each cultivar had a different amount of return bloom and return fruit depending on where it was grown (Figure 1). In 1990 Wisconsin typically had the highest percent return bloom and return fruit. Massachusetts and Oregon were typically lower, while New Jersey was variable. These data are also supported by 1990 production statistics.

Percent return bloom was highest for Ben Lear in Wisconsin (74%) and lowest for Howes in New Jersey (14%) (Figure 1A). Crowley was the most consistent performer across all production areas. Ben Lear differed most in percent return bloom among regions; from 16% in Massachusetts to 74% in Wisconsin.

Despite high percent return bloom for some cultivars and regions, many flowers did not set viable fruit (Figure 1B). Ben Lear had only 49% return fruit following 74% return bloom in Wisconsin, a 30% reduction. Searles in Wisconsin was highest of the single state cultivars in both return bloom and return fruit. Percent return bloom followed percent return fruit and, of course, in all cases was somewhat lower.

The differences in percent return bloom and return fruit among regions for a given cultivar may have been caused by differences in cultural practices as well as

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environmental factors. The age of the bed, yield in 1989 and 1990 and cultural practices such as sanding, pruning, and fertilization may well have had an influence.

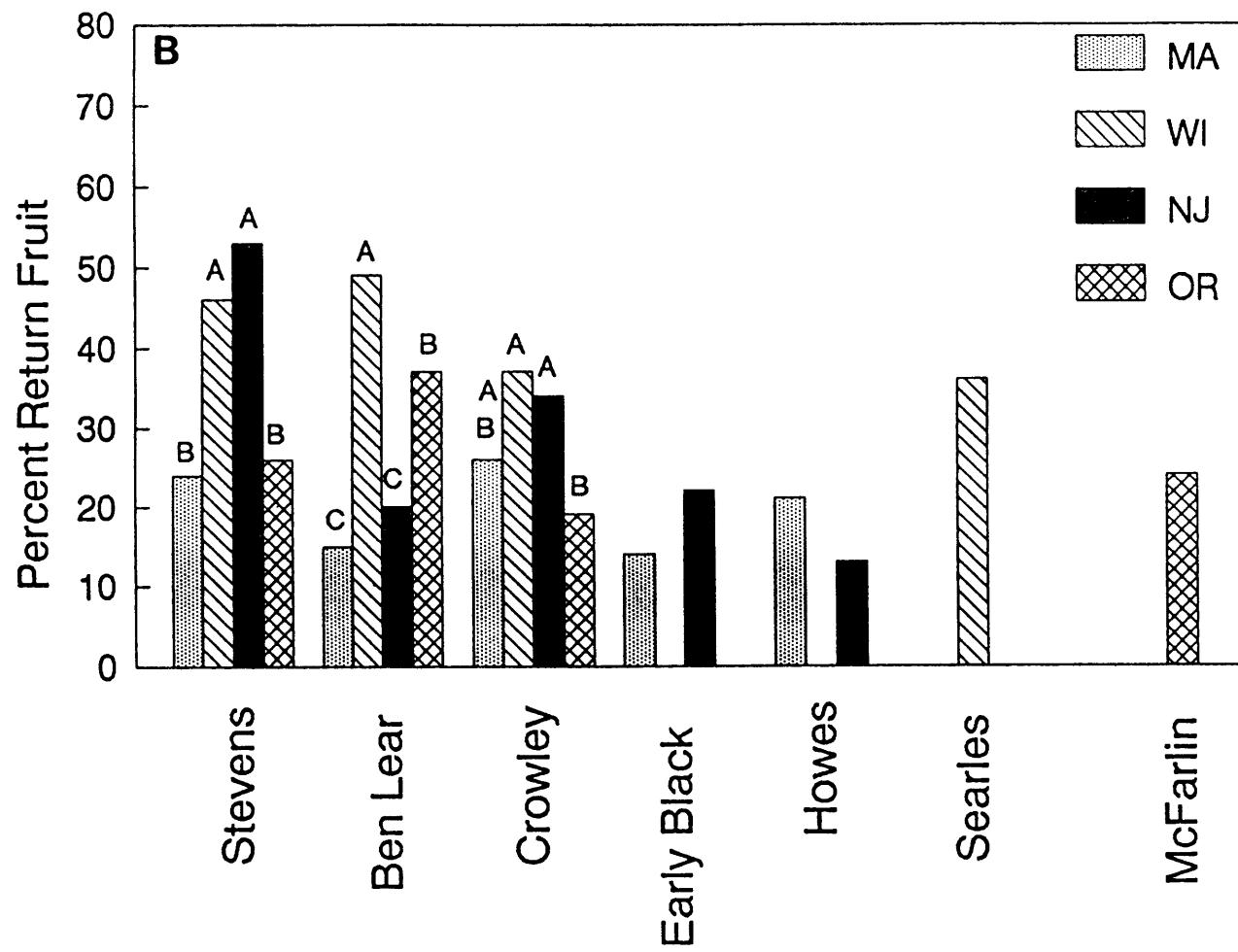
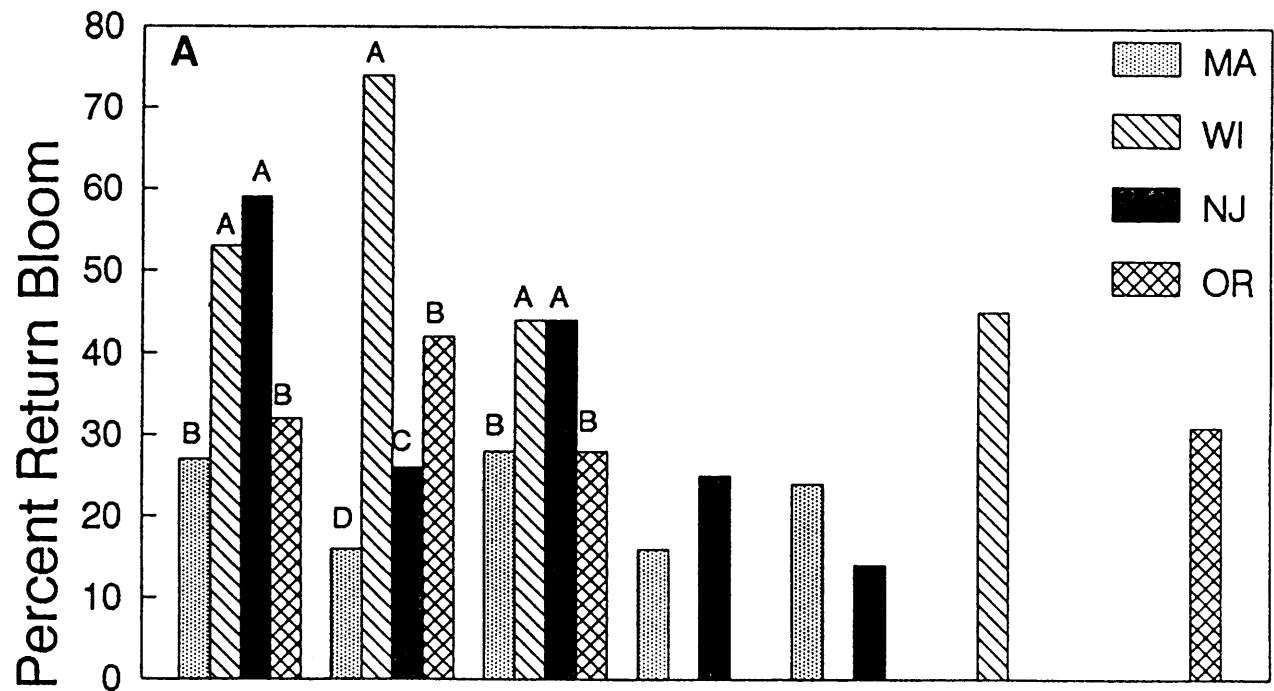
Two internal factors also may be affecting return bloom and return fruiting in cranberry. First, flower initiation, therefore, return bloom may be inhibited by internal plant hormones which are produced by developing flowers and fruit. This has been found to be the case in apples as well as some other fruit. Second, even if flowers open and are pollinated there may be insufficient resources so that the flowers could be weak and would be unlikely to set fruit.

In conclusion, cultivars differed in percent return bloom and set within and among regions. Regional differences for a particular cultivar could have been due to cultural and environmental effects. The low percent return bloom for most cultivars could be due to resource limitation and/or hormonal factors during fruiting, which may adversely affect concurrent flower bud initiation and thus, percent return bloom the following year. Cultivar and regional and cultural differences could be related to photosynthetic efficiency and partitioning of photosynthates. The physiology of either has yet to be determined in Cranberry.

This research will be continued in 1991 with more sites and fewer cultivars as we try to unravel the potential causes for biennial bearing for individual uprights.

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*The cooperation of Gaynor Cranberries, Wisconsin Rapids, in conducting this research is gratefully acknowledged.*



## PLANT NUTRITION OF THE CRANBERRY CROP

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The cranberry plant requires certain chemical elements which we refer to as plant nutrients for normal growth and development. Three of these elements (carbon, hydrogen, oxygen) come from air and water, and another 13 elements (nitrogen, phosphorus, potassium, calcium, magnesium, sulfur, zinc, boron, manganese, iron, copper, chloride, and molybdenum) are supplied by the soil and are absorbed into the plant by the root system. If any one of these 13 elements is not adequately supplied by the soil, it is necessary to supply the element of concern by fertilization. However for a majority of these elements, the soil supplies an adequate amount for normal growth, and as growers you need not be concerned with but a few of the elements. If a reasonable fertilizer program has been followed, fertility will very seldom be a problem.

As growers it is important that a diagnostic procedure be available to evaluate the nutritional status of your crop. One procedure is leaf or tissue analysis. A tissue analysis can provide an almost complete listing of the soil supplied elements which allows for a good evaluation. For a perennial crop like cranberry, tissue analysis is a good diagnostic tool.

For tissue analysis to be effective, it is essential that a set of standards for the nutrient elements be available for comparison to the elemental composition of field tissue samples. This comparison will assist in the determination of the absence or presence of a plant nutritional problem. A set of standards for a number of the nutrient elements was developed by Dr. Dana and is shown in Table 1. We are in the process of evaluating the standards primarily for confirmation and to provide additional information which will permit us to do a more effective evaluation of field samples. Besides composition, definable symptoms can be helpful in determination of possible problems.

For an evaluation of the standards, cranberry plants have been grown in hydroponic systems under greenhouse conditions. This allows for accurate control of nutrient availability and the development of plants with a wide range of elemental composition. Composition is then compared to the type of growth which may be normal or abnormal.

Specific evaluation procedures have been to grow cranberries from rooted cutting for 10 weeks on a complete nutrient solution in a hydroponic system. Excellent growth is obtained during this time period with 1.5 to 2 foot runners as a common response. At 10 weeks, the plants are transferred to a complete nutrient solution minus the element to be evaluated. The common response is a gradual reduction in the concentration of the nutrient element in the tissue to a point where it will become low enough to reduce plant growth and also to develop definable symptoms. The elemental tissue concentration at this point will assist in defining the low level where the cranberry plant will function normally and will set the low level for the plant analysis standards which is a most important standard. The data

developed for the nutrient elements phosphorus (P), potassium (K), and sulfur (S) are shown in Figures 1, 2 and 3, respectively. For phosphorus in Fig. 1, yield continues to increase even after 6 weeks without available phosphorus in the nutrient solution. During this period the phosphorus concentration in the tissue dropped below the proposed low level of P without a growth reduction. The response to potassium is different (Fig. 2). When the potassium concentration in the tissue dropped below the proposed low level, growth stopped indicating that the present proposed level is accurate. No standard has been established for sulfur. The data for sulfur (Fig. 3) indicate that growth continues even at a very low tissue concentration of sulfur. This type of data has been collected for all of the essential nutrient elements except iron, chlorine, and molybdenum and will be analyzed and provided in a form which can be used by you the grower.

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**Table 1.** Nutrient concentration of cranberry tissue where deficiency symptoms were observed and the proposed concentrations of cranberry tissue samples for determining the nutrient status of a crop.

Nutrient	Conc. at observed deficiency	<u>Proposed conc.</u>		
		Low	Sufficiency range	High
<u>%</u>				
Nitrogen	0.70	<0.90	0.90 to 1.00	>1.00
Phosphorus	0.09	<0.13	0.14 to 0.18	>0.18
Potassium	0.17	<0.50	0.50 to 0.90	>0.90
Calcium	0.05	<0.30	0.30 to 0.60	>0.60
Magnesium	0.02	<0.15	0.16 to 0.20	>0.20
Sulfur	no estimates			
<u>ppm</u>				
Zinc	3.8	<15	15 to 30	>30
Boron	1.0	<10	10 to 20	>20
Manganese	2.0	<10	10 to 200	>200
Iron	2.6	<40	40 to 80	>80
Copper	3.1	<5	6 to 10	>10

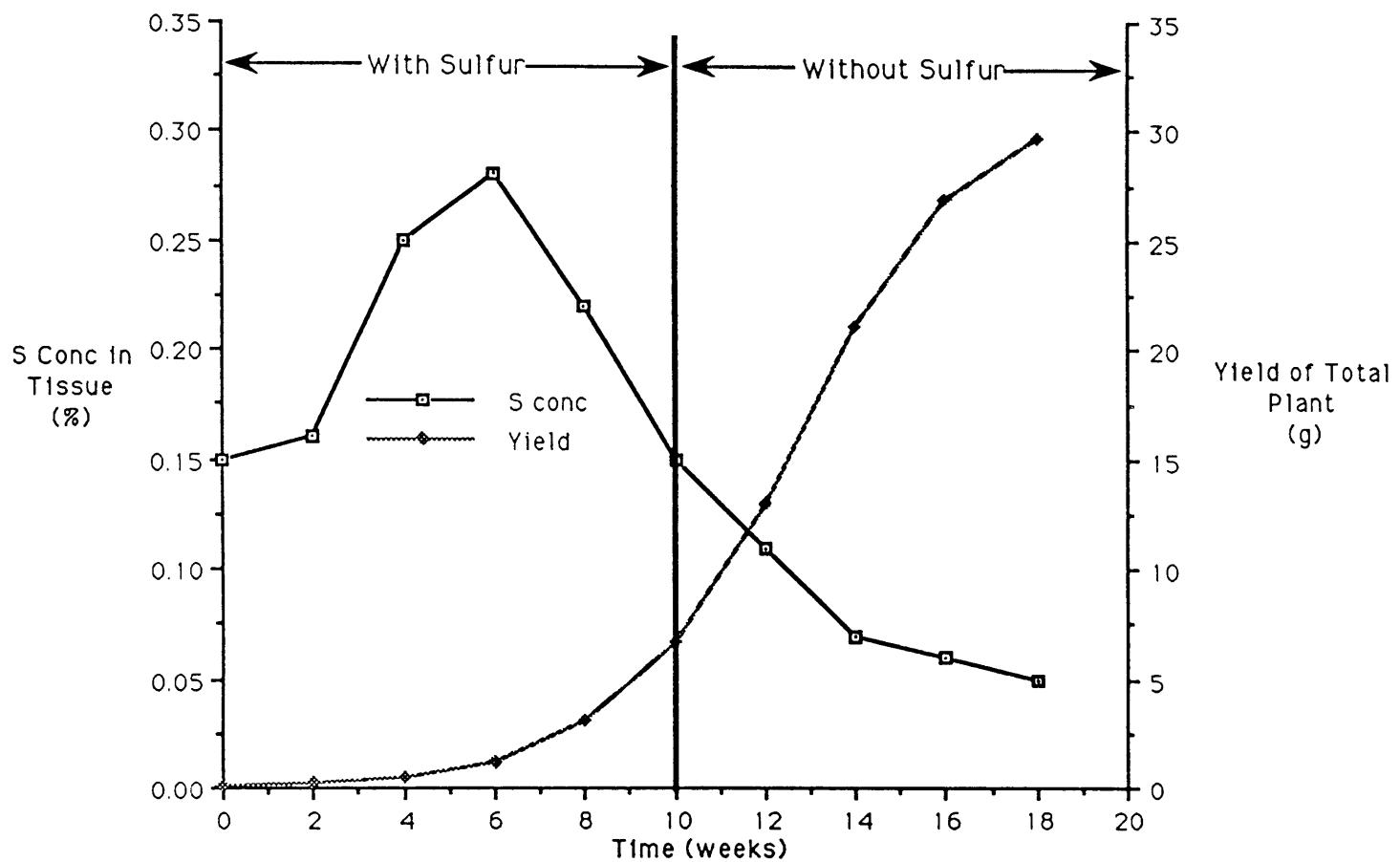


Fig. 3. Relation between S concentration and yield of cranberry plants.

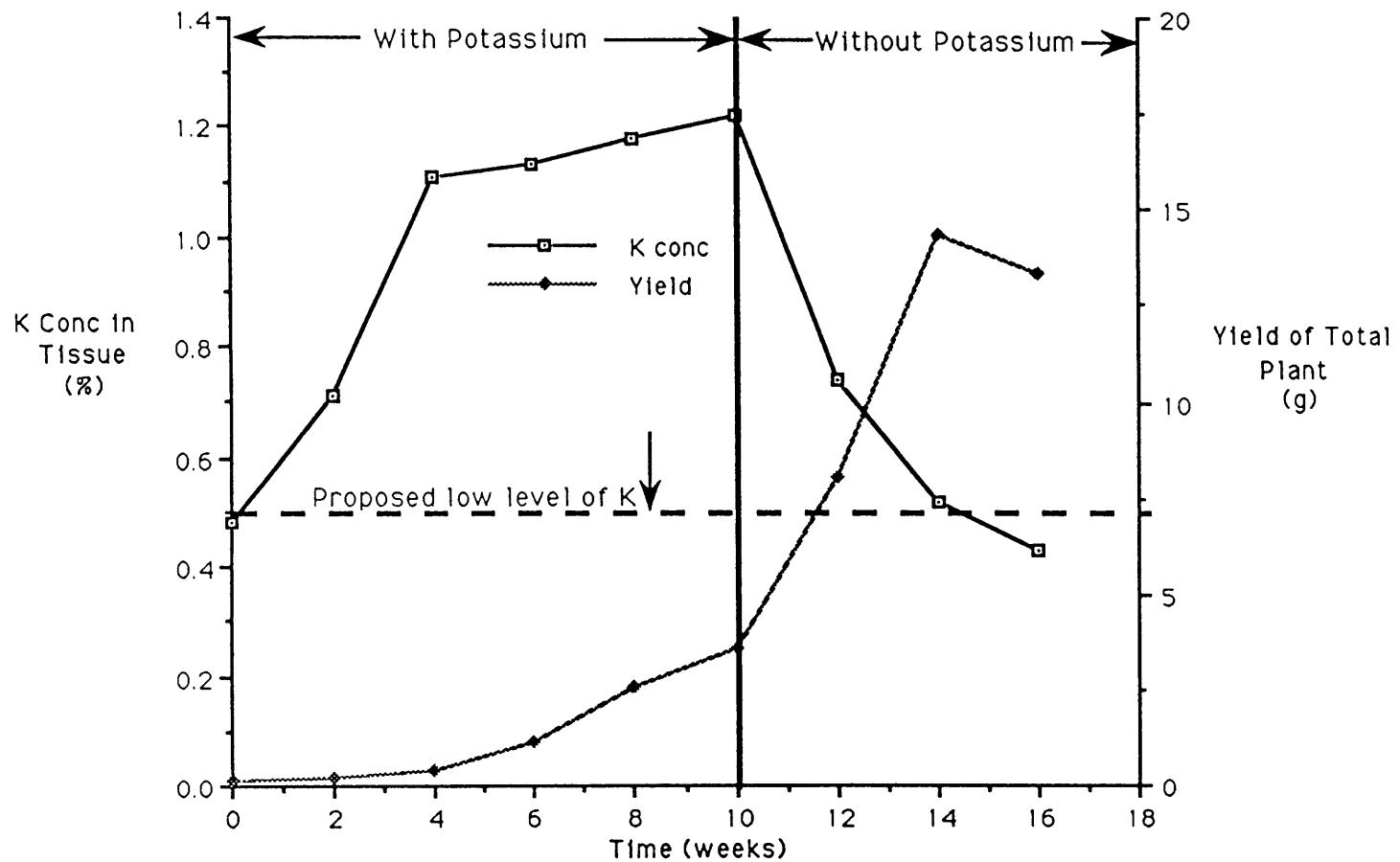


Fig. 2. Relation between K concentration and yield of cranberry plants.

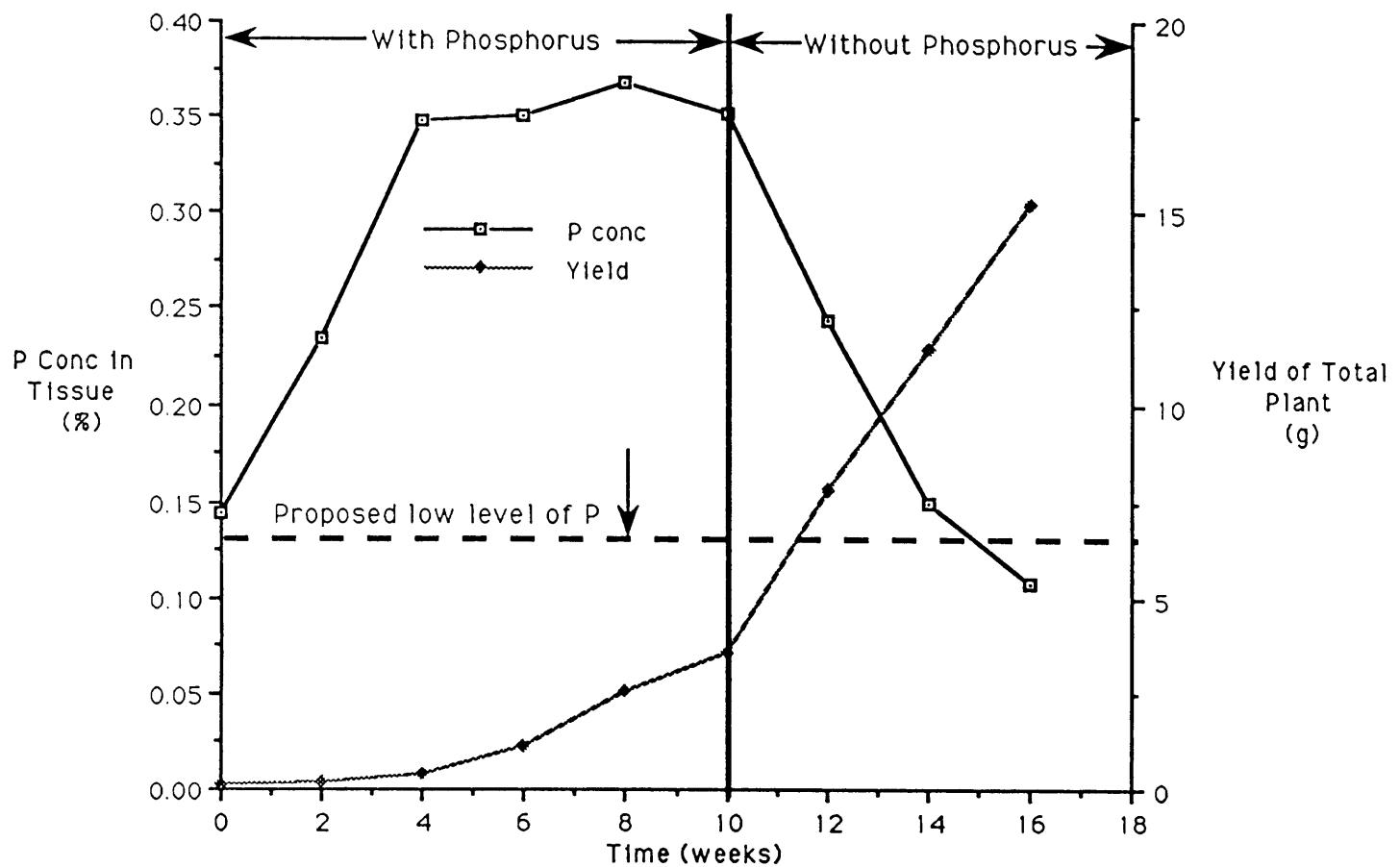


Fig. 1. Relation between P concentration and yield of cranberry plants.

## CONTROLLING CRANBERRY GIRDLER WITH BIOSAFE-N FOR CRANBERRIES

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Cranberry girdler is one of several serious insect pests that infest cranberry beds. It is the larval stage that is destructive to the cranberry vine. The larvae feed on the woody parts such as the runners and larger roots and only occasionally feed on the finer roots.

Injury occurs in the larval stage, beginning in early June and continuing throughout the summer months and into fall, usually into mid-October. The newly emerged larvae do not cause the extensive damage that the larger maturing larvae cause. For this reason, evidence of the presence of cranberry girdler sometimes goes unnoticed until vines begin to express feeding damage through the early fall expression of red or brown foliage in September and October. Large quantities of leaves begin to drop from the cranberry vine, leaving areas of dead vines. In severe cases, the vines become severed in portions of the field and one is able to roll the vines back, like a carpet, exposing the soil beneath.

In the cranberry field, larval feeding carries on concealed in the trash layer that consists of fallen leaves, and other organic matter. This concealed feeding area also provides the larvae with a haven for protection from natural parasitic enemies.

As the larvae mature into larger forms, they become more ravenous and consume a great deal of cranberry vine bark, often feeding right through the vine and completely severing the vine.

### **The Use of BioSafe-N for Cranberries to Control Cranberry Girdler**

BioSafe-N for Cranberries can be applied from mid-July through mid-September. The application is timed with the larval stage of the cranberry girdler. Biosys now recommends a new reduced rate for the control of cranberry girdler. That rate is now 2 billion beneficial nematodes per acre.

BioSafe-N for Cranberries can be applied with most spray equipment. Overhead sprinkler systems, backpack sprayers as well as other application equipment may be used to apply the product. Always apply the product to moist soil. The natural environment for beneficial nematodes is moist soil. Pre-irrigate the bed for 30 minutes. This will prepare the bed for the application and also flush and cool irrigation lines. After applying BioSafe-N for Cranberries, follow the label and continue irrigating another  $\frac{1}{4}$  inch of water. Be careful not to flood the bed. The key is to get the beneficial nematodes down to the soil through the trash layer.

Once the product is applied, begin monitoring your beds after a week and look for signs of girdler infection. You will notice a change in activity as the larvae become less active and begin to turn light brown in color. If you have access to laboratory equipment, collect several larvae from the treated bed and carefully

dissect them under a microscope in several drops of water in a petri dish. You should see many new beneficial nematodes that reproduced inside the girdler larvae.

BioSafe-N for Cranberries will last up to 4 to 6 weeks in the soil after following label directions. When applied properly, BioSafe-N for Cranberries can result in the control of girdler larvae up to 95%.

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# PESTICIDE STORAGE, MIXING, AND LOADING SITES

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March 1991

## INTRODUCTION

Commercial, aerial, and private storage and mixing/loading sites are at risk of spills of pesticides and fertilizers which may contaminate groundwater. The rules written by the WDATCP fall under Wisconsin's groundwater legislation and were implemented to reduce the risk of accidental spills and/or poor management practices from contaminating soil and water at these sites. Ag 29, which took effect on May 1, 1990 requires compliance by end users to reduce this risk by several ways. Figure 1 describes the compliance of all end users for installing a mixing/loading pad for handling pesticides. In Wisconsin these rules are impacting small commercial applicators, lawn companies, vegetable and fruit growers, golf course operators, and private applicators depending on the amount of pesticide used. These rules deal with the mixing and handling of pesticides, but the safe storage of the pesticides is also implied in the rules.

## LIABILITY

There are other reasons for installing proper chemical storage and handling facilities. The liability of the owner for accidental spills on the farm causing water contamination may prompt construction of a facility. Insurance carriers are limiting policies on environmental damage caused by a fire or a spill involving pesticides, and may require certain practices be put in place before a policy can be written. The owner/operator can also protect the well from contamination for his/her own family's safety.

## FUNCTIONAL DESIGN

Functional planning of an pesticide facility should be the first item addressed in developing new or remodeling existing facilities. A well planned and designed facility is needed for environmental and human protection. Agricultural facilities are especially visible and potentially risky due to the nature of the activities at these sites. Typically, highly concentrated chemicals are stored and handled at these sites. The pesticide facility provides several distinct and separate functional areas shown in Figure 2:

- Pesticide Storage
- Secondary Containment
- Mixing/Handling Equipment
- Loading/Washpad
- Worker Safety
- Waste Disposal

## Pesticide Storage

The storage area is used to store pesticides. It should be designed to protect pesticides from theft, temperature extremes if needed, and unauthorized use by untrained personnel. The storage facility should be isolated from other buildings used to store feeds or fuel and should only store pesticides. There should be visible signs indicating the materials are toxic and dangerous. The area should be ventilated to prevent a concentration of fumes from building up in the area. The storage area should also be secured by a fenced area or a locked building to prevent vandalism and unauthorized entry.

## Secondary Containment

Secondary containment provides protection of the environment from accidental leaks and spills of bulk liquid storage tanks and small handling spills during the normal daily routine of mixing and loading chemicals and fertilizer into spraying equipment. Bulk liquid storage tanks are placed within a containment area to prevent release of the fertilizer or chemical in the event that the primary storage tank fails in some way. The spilled material can then be easily recovered instead of leaching into the soil or draining into surface water. Pesticides in minibulk containers and rinsate storage tanks are typically contained on a curbed pad constructed from concrete or other impermeable material.

The secondary containment is sized to hold 125% of the largest storage tank or sprayer tank in the secondary containment. The displaced volume of any other storage tanks or equipment must be considered in determining the size of secondary containment. The inside dimensions of the walls or curbed area must be used in calculating the containment volume.

## Mixing/Handling Equipment

The mixing/handling area is most often located adjacent to or between the storage/containment area and loading/wash pad area for convenience. The mixing/handling area provides secondary containment during the transfer of chemicals from storage to the loading area. Pesticide containers, and minibulks are temporarily stored and handled in this area. The pumps, valves, hoses, and meters used to transfer chemicals from containers are also located in this area. Leaks and small spills occurring during the transfer and handling of chemicals is contained and collected in this area.

## Loading/Washpad

The loading/wash pad is used to park application or transport equipment during the loading of pesticides. Unloading of chemicals into the storage building also takes place over the pad. Repair of sprayer equipment should also be done over this area to collect any material that leaks or is drained from the tanks or booms of the spray equipment. Typically, this area is a sloped and curbed concrete pad or other impermeable surface. The pad is sloped to a shallow sump where contaminated water (rinsate) is collected, pumped to above ground rinsate storage tanks and used as makeup water for

subsequent sprayer loads. The rinsate is segregated and reused on the target crop reducing waste. If this area does not have a roof, rainwater falling on the pad is collected and stored for future use if it is contaminated.

### Worker Safety

The worker safety area should be equipped with all the necessary emergency equipment needed to prevent harm or provide emergency aid to the workers. An eyewash and/or deluge shower should be provided to rinse spilled chemical from the eyes, face and body. A first aid kit and spill response kit should also be provided to deal with accidents in a timely manner. A fire extinguisher should also be provided. Personal protection equipment should also be available at the chemigation site.

### Waste Disposal

The waste disposal area is used to hold empty containers temporarily until they can be disposed of properly. The pressure rinsed empty containers should be stored on a covered, curbed area to prevent rain entry into the container or leaks from containers contaminating the soil. Old burn piles and uncovered empty container piles have been identified as a major contamination source at many existing storage facilities. Empty minibulk or returnable containers should also be stored in this area until they can be returned or refilled. Returnable and/or reusable containers should be used whenever possible. The private applicators training manual should be consulted to determine best method for cleaning up a spill and to determine when a spill is to be reported.

### Chemigation System Equipment

The pesticide container, pesticide injection unit, and all connections between them must be located in secondary containment. This may be concrete curbed area, or a large tank which is sized to hold the chemigation system equipment and the pesticide container. A transportable secondary containment system may be of benefit for use at multiple application sites. The secondary containment must be located at least 8 feet from the water supply.

Figure 3 shows a general schematic of the equipment requirements for chemigation. The construction materials must be resistant to corrosion, puncture and cracking and be chemically compatible with the pesticides used in the system. Written confirmation of compatibility must be kept on file by the operator and be available for inspection by WDATCP upon request. Backflow prevention must be incorporated into the system to protect the water supply from contamination. Most chemigation systems will find the reduced pressure principle backflow preventor to be the most practical for protecting a well or surface water supply. Alternatives for protecting a well water supply are a fixed air gap/repump system or barometric loop. Alternatives for protecting a surface water supply are a double check valve system, gooseneck loop and check valve, or a barometric loop. A low pressure switch must be installed to shut off the irrigation system power supply if water pressure decreases to the point where pesticide is no longer being applied according to label rates. A pump interlock between the water supply and the pesticide injection pump shuts the injection pump off if the water supply is interrupted.

A flow interrupter is placed in the pesticide supply line which shuts off the pesticide supply if the injection pump fails. A flow sensor installed in the pesticide injection line near the injection pump shuts off the injection pump if the injection line fails. A check valve with a minimum opening or cracking pressure of 10 psi must be installed in the pesticide injection line between the injection pump and the point of injection. This prevents backflow of irrigation water into the pesticide tank, and prevents low pressure flow of pesticide into the irrigation system if the injection pump fails.

### **Farm Sized Facility**

No matter what the size of the operation each of the functional areas should be incorporated into the total system facility plan. The size or scale of the functional areas of an operation is dependent on the amount of pesticide and/or fertilizer stored at the facility and the number of employees. In many cases a single space can be used to provide for several functional areas such as minibulk storage, rinsate storage and mixing and loading chemicals. This provides for flexibility in the layout and design of the facility. Noyes and Kammel 1989 discuss a detailed set of plans for a mixing/loading pad that can also incorporate agrichemical storage. As the size of the facility increases the space needed for each functional area becomes larger and more well defined. Functional areas should be placed adjacent to each other to provide for efficient traffic flow and easy access from one area to another. Remote chemigation sites should also incorporate these same design concepts as much as possible.

### **Facility Management Plan**

Pesticide storage and handling on the farm is carried out according to a facility management plan. The plan specifies the storage, handling, cleaning, and application of pesticide materials and equipment used in the farm operation. The facility management plan is to be used in conjunction with the plans and specifications of the chemical storage building and loading pad to reduce the potential for groundwater contamination from the storage and handling of pesticides on the farm, and to provide a safe environment for the operator/owner using the pesticides.

### **Chemigation Operation Plan**

The chemigation operation plan is part of the facility management plan. The owner/operator of the chemigation system must prepare a written observation plan prior to operating the system. The plan must be followed during system operation. The plan must be kept at the site or with the operator while the system is functioning, and a copy must be kept at the residence or business office of the owner/operator. The operation plan must include a) A list or drawing indicating sensitive areas which may be subject to drift. This includes nontarget areas such as roads within 100 feet of system, public or populated areas within 300 feet of the system, surface water and wetlands. b) a description of methods and procedures used to prevent drift and overspray. c) A description of the backflow prevention system. d) A description of methods and procedures to assure calibration of the system. e) A description of the monitoring procedure to follow to assure the system is functioning properly according to the plan and the law. f) A statement indicating the flush time for the system. g) A description of

the safety protection for persons observing the system or entering the treated area for repairs.

### Emergency Response Plan

An emergency response plan is part of the facility management plan. This response plan contains:

- A set of plans and specifications for the facility.
- A facility site plan.
- An inventory list of chemicals stored.
- The facility management plan.

Copies of the emergency response plan should be available:

- At the facility.
- At the owner's house.
- Local emergency government office.
- Local fire official office.

### Summary

With proper planning and consideration of design fundamentals, a pesticide application facility can be incorporated into commercial and private applicator sites. These facilities will reduce potential harm to workers and also protect the environment, especially the groundwater. These sites are especially visible and potentially dangerous because of the quantity and concentration of the chemicals used. Farm sites may not require a complex design, but still need to address certain design fundamentals to reduce groundwater contamination.

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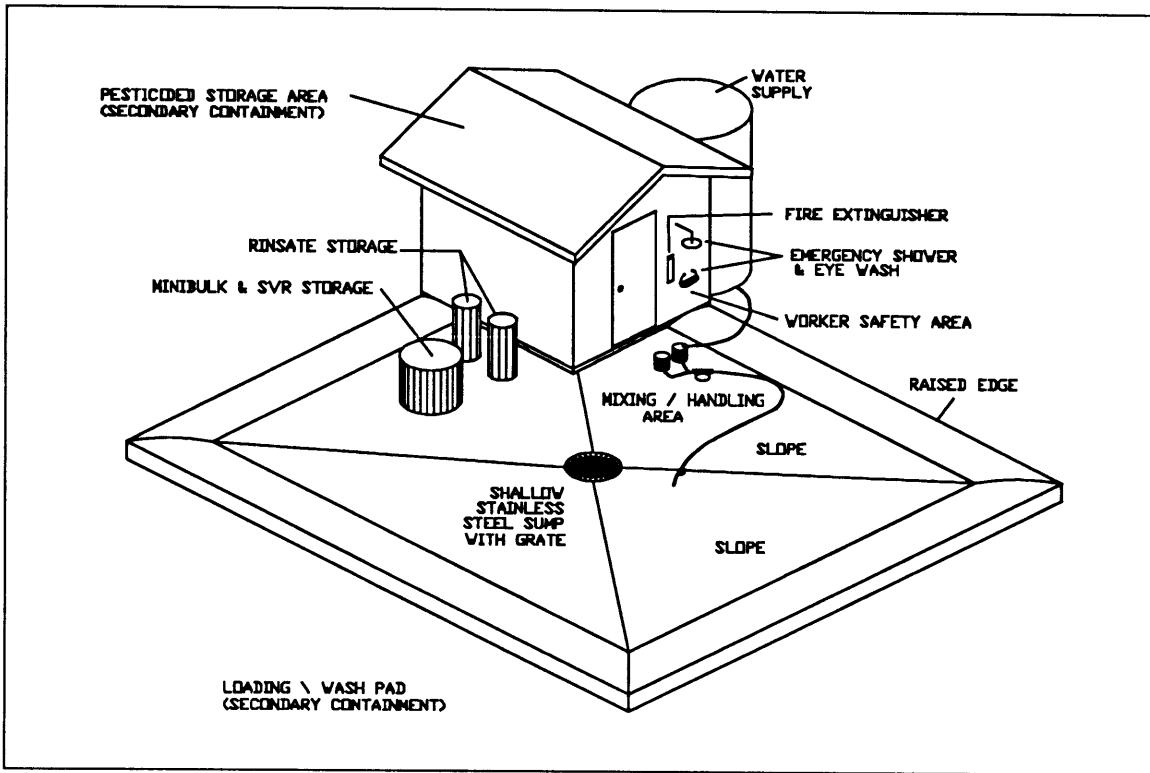


Figure 2 Farm Sized Pesticide Storage, Mixing/loading Facility

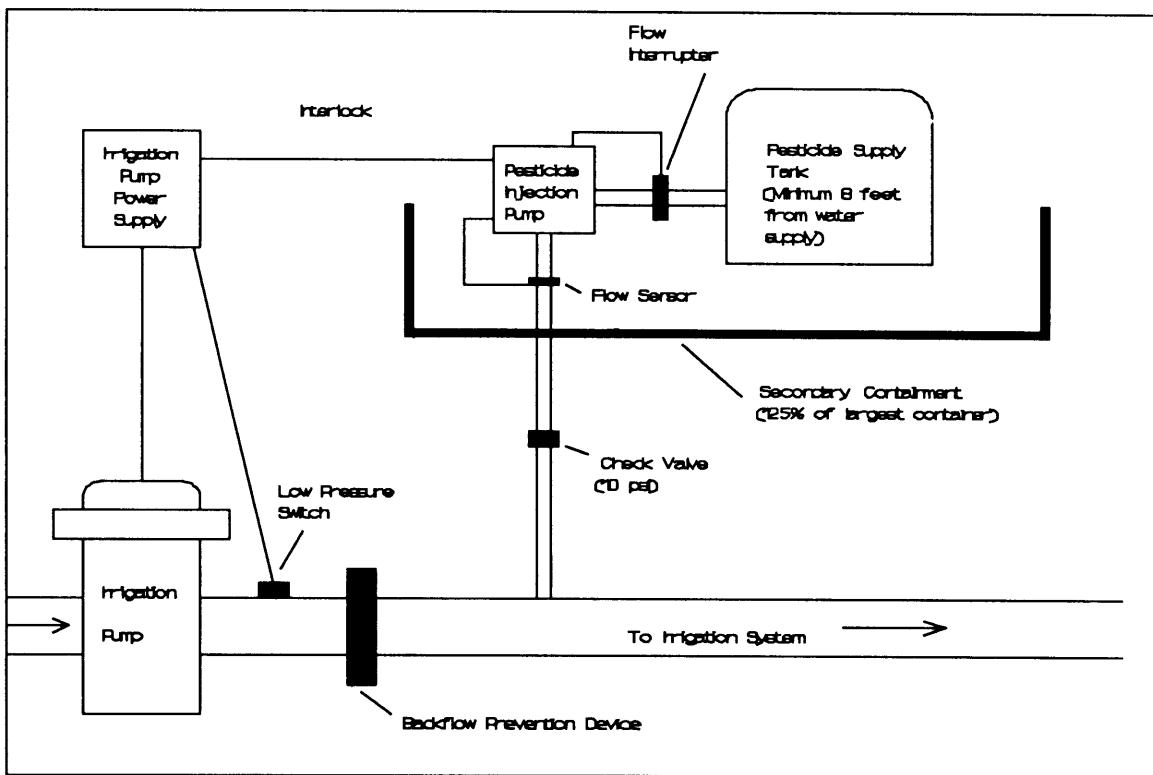


Figure 3 Ag 29 Chemigation Equipment Requirements

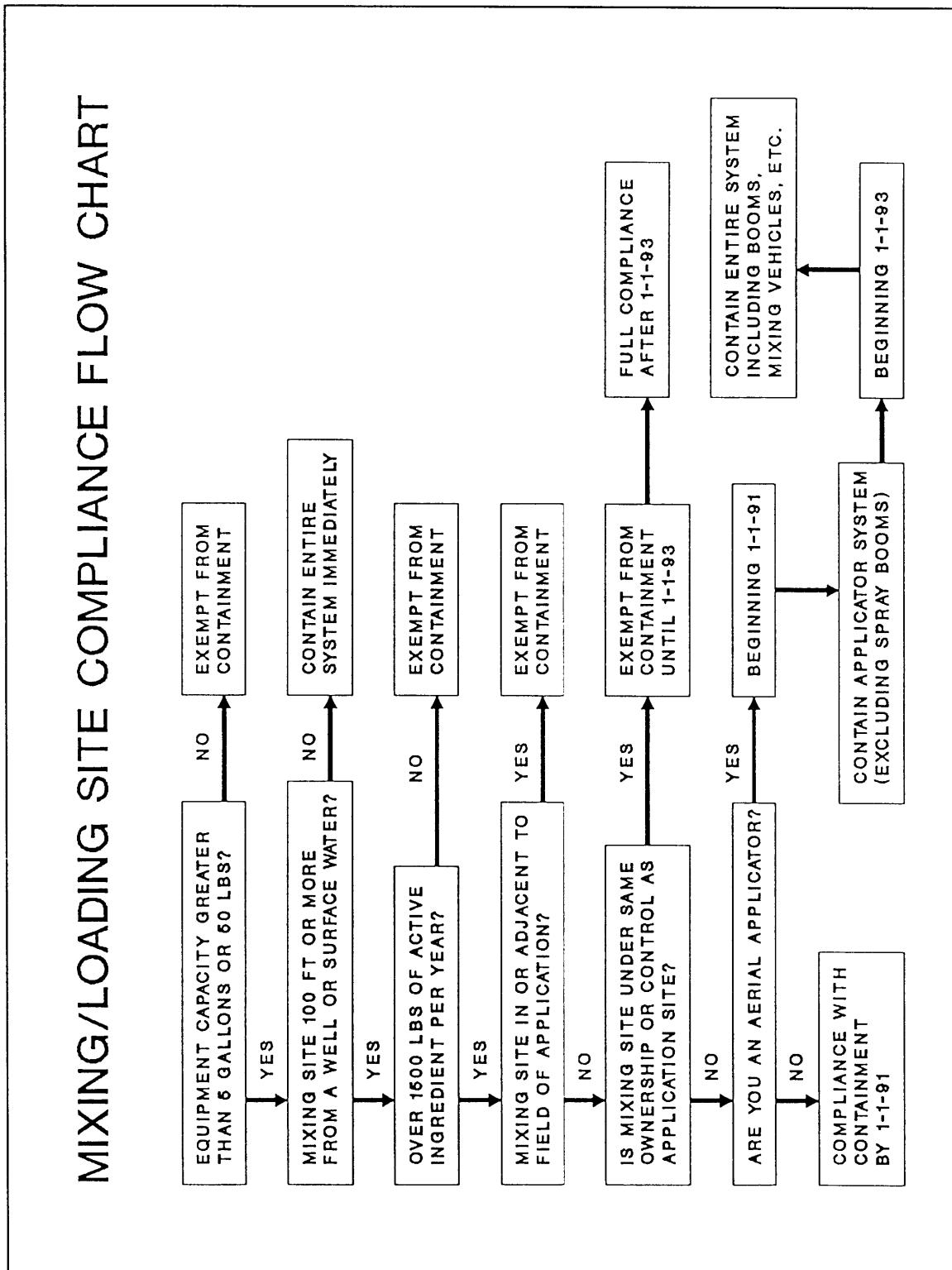


Figure 1 Mixing/loading Site Compliance Flow Chart (WDATCP 1990)

## CONTROL OF OLDFIELD CINQUEFOIL

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Oldfield cinquefoil (*Potentilla simplex* variety *calvescens*) is a perennial weed native to Wisconsin which in recent years has increased in severity in cranberry production fields. It is a prevalent ground layer species of Wisconsin southern dry forest and is believed to have been introduced to cranberry marshes by seeds in sand used for top sanding. Cinquefoil has become a problem since sprinkler irrigation has been practiced in cranberry production. It has the potential of becoming a more troublesome weed in the future due to its growth habit. It spreads by runners in the field and develops a thick plant mass on top of cranberry vines, therefore interfering with cranberry growth and with mechanical harvester operation.

Flooding as a means of weed control was derived from the culture of rice. This method works by keeping oxygen from the roots and leaves. Flooding was used for insect control in cranberries during the early 1900's.

Greenhouse and field studies showed that soil saturation, 2 inch and 3 inches of flooding for 12, 24, 36, and 72 hours did not affect cinquefoil growth significantly. Formation of adventitious roots as a flooding stress response was not observed in these studies. In a growth chamber experiment, four weeks of soil saturation induced the formation of cinquefoil adventitious roots, both at 55° and 72°F day temperatures. At 55° day temperature, soil saturation did not affect cinquefoil growth. However, growth was reduced compared to the plants grown at 72°F. At 72°F day temperature, soil saturation reduced growth in terms of runner development and fresh and dry weight.

Greenhouse herbicide studies showed that GOAL caused injury to cinquefoil during the first week after treatment but the injury decreased with time. Dichlobenil<sup>1</sup> and CLASSIC caused first injury symptoms at two weeks after treatment and the effects increased with time. Small weeds were more susceptible to herbicides as was shown by the greater percentage of injured leaves and higher injury ratings, compared to larger weeds. CLASSIC, dichlobenil and SINBAR significantly reduced the number of leaves, leaf area, and shoot, root, and total fresh and dry weight of cinquefoil at 12 weeks after treatment.

GOAL phytotoxic effect on cranberry decreased with time, and cranberry recovered completely at 9 weeks after treatment. SINBAR effects increased with time, while 2,4-D amine effect was relatively constant until 12 weeks after treatment. CLASSIC at all levels evaluated caused no reduction in cranberry vine growth in terms of the number of branches per plant, height, and fresh and dry weight. Dichlobenil at a high rate did not reduce the number of branches per cranberry plant, but significantly decreased cranberry height and fresh and dry weight.

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1. *Dichlobenil is sold under the trade names of CASORON and NOROSAC.*

The rates of dichlobenil evaluated in these studies were; 1(low) and 4(high) pounds of active ingredient per acre. Control of cinquefoil was greater as the rate increased from 1 to 4 pounds per acre of active ingredient.

Herbicide application in a cranberry marsh at cinquefoil bud break did not provide adequate cinquefoil control. Postemergence herbicide application in a cranberry marsh, however, gave significant positive control results. Dichlobenil, CLASSIC or SINBAR significantly reduced the number of weed leaves and weed fresh and dry weight per plot at 12 weeks after treatment. SINBAR reduced cranberry shoot fresh and dry weight significantly. Cranberry fruit fresh and dry weight were not significantly different in Dichlobenil and CLASSIC treated plots compared to the control. However, CLASSIC and SINBAR treated plots had significantly lower cranberry fruit fresh and dry weight compared to Dichlobenil treated plots.

## CONCLUSIONS

Soil saturation and flooding studies have shown that Oldfield Cinquefoil growth was not affected by the treatments up to 72 hours in length suggesting that using flooding as a cinquefoil control in cranberry is limited.

Herbicide studies indicated that dichlobenil and CLASSIC are the two promising herbicides to be used for cinquefoil control in cranberry.

Dichlobenil is labeled for use in cranberry and CLASSIC is being investigated as a IR-4 (minor crop) clearance. Dichlobenil is labeled for use in cranberries at a rate of up to 100 pounds of 4% granular material per acre.

Appreciation is expressed to the Wisconsin Cranberry Board for financial assistance and the the Olson Brothers Cranberry Company of Warrens, WI for use of field research facilities.

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## FERTILIZER GRADES: DETERMINING APPLICATION RATES OF DRY AND LIQUID FERTILIZER MATERIALS

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- A. Fertilizer analysis--Method of indicating the amount of plant nutrients a fertilizer material contains.
- B. Fertilizer grade--The minimum guaranteed amounts of available nitrogen,  $P_2O_5$  and  $K_2O$  in fertilizer material (in that order).

example: 5-10-30  
 $N-P_2O_5-K_2O$   
 5% N, 10%  $P_2O_5$ , 30%  $K_2O$  (by weight)

1. Fertilizer grades for phosphorus and potassium are expressed on an oxide basis rather than an elemental basis.
2. Fertilizer recommendations for phosphorus and potassium are also usually given on the oxide basis (eg., lbs.  $P_2O_5$  and lbs.  $K_2O$  per acre required).
3. There have been attempts to convert the method of reporting fertilizer grades from oxide to elemental basis, but there has been little progress in this direction.
4. Because there is potential confusion between elemental and oxide methods of expressing the amounts of phosphorus and potassium in fertilizer materials, the relationships between these forms are shown below.

Elemental Name	Elemental symbol	Oxide name	Oxide symbol	Plants use
Phosphorus	P	phosphate	$P_2O_5$	$H_2PO_4^-$
Potassium	K	potash	$K_2O$	$K^+$

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Conversions:

$$1 \text{ lb. of } P_2O_5 = 0.44 \text{ lbs. P}$$

$$1 \text{ lb. of P} = 2.29 \text{ lbs. } P_2O_5$$

$$1 \text{ lb. of } K_2O = 0.83 \text{ lbs. K}$$

$$1 \text{ lb. of K} = 1.20 \text{ lbs. } K_2O$$

5. Remember that fertilizer materials do not actually contain P and K as  $P_2O_5$  and  $K_2O$ , and plants do not take up  $P_2O_5$  and  $K_2O$ . The forms are only used to indicate the amounts of phosphorus and potassium in fertilizers.
6. Fertilizer terminology
  - a. Fertilizer ratio--The relative proportion of nitrogen (N), phosphate ( $P_2O_5$ ), and potash ( $K_2O$ ) in a fertilizer material.
 

<u>Grade</u>	<u>Ratio</u>
6-24-24	1:4:4
5-10-30	1:2:6
9-9-9	1:1:1
  - b. Mixed fertilizer--Contains more than one of the three major nutrients: N,  $P_2O_5$ ,  $K_2O$ . (eg. 18-46-0)
  - c. Complete fertilizer--Contains all three of the major nutrients. (eg. 6-24-24)
  - d. Straight fertilizer--Contains only one of the three major nutrients. (eg. 46-0-0)
7. Calculation of the amounts of N,  $P_2O_5$  and  $K_2O$  in liquid fertilizer materials requires information on weight per gallon of the liquid fertilizer.

#### Weights per gallon of some liquid fertilizers

<u>Material</u>	<u>Weight</u>
10-34-0	11.7
28-0-0	10.7
12-0-0-26	11.1
7-21-7	11.2
9-18-9	11.7

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# COLOR ENHANCEMENT IN CRANBERRY FRUIT BY USING ENVIRONMENTALLY SAFE NATURAL PRODUCTS

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## 1. Introduction

The commercial value of the cranberry crop is related to the amount of color (anthocyanin) in the fruit. The production of color at the time of fruit ripening appears to be in response to low temperature and light. Thus the fruits exposed on the top of plant canopy change color first and the fruits underneath the plant canopy remain white or blush until quite late in the fall. In order to get good color growers usually try to delay harvest. However, the problems and risks associated with frosts discourage growers to delay harvest.

Research has been conducted to devise chemical means to accelerate and increase color production in the cranberry fruit. Ethephon is an approved chemical for use on cranberries in Wisconsin for increasing anthocyanin content. However, its application has yielded inconsistent results from season to season (Shawa, 1979). Several years ago we initiated research to find environmentally safe chemical means for early color enhancement in cranberry fruit. A summary of this research and progress to date is given below.

## 2. Cranberry Fruit Cuticle (surface) is Essentially Impermeable to Ethrel.

By microscopic examination of live cross-sections of cranberry fruit we were able to demonstrate that the color in the fruit is limited to outer two cell layers (Palta and Stang, 1983). We were also able to show that cranberry has relatively thick cuticle. Later examination of the fruit surface with scanning and transmission electron microscopic showed that fruit cuticle was embedded with waxes (Farag, et al., 1985; Farag and Palta, 1987b). This cuticle contained no pores. We hypothesized that cranberry fruit cuticle was essentially impermeable to Ethrel. This was later confirmed by measuring transport of Ethrel across isolated fruit cuticle (Farag and Palta, 1987a; Farag et al., 1985).

## 3. Overcoming the Permeation Barrier by Modifying the Ethrel Solution

The transport of Ethrel across enzymatically isolated fruit cuticle was studied. For this purpose a transport chamber was designed and fabricated. This chamber allowed the study of transport of Ethrel across the fruit cuticle. Ethrel permeation across the cranberry fruit cuticle was very slow and in most cases the cuticle was essentially impermeable to Ethrel. We then systematically modified the Ethrel solution by adding transport enhancers. These enhancers increased the permeation of Ethrel across the fruit cuticle by over 100-fold. All the enhancers studied were natural products. These enhancers were effective because of one or more of the following properties (Farag and Palta, 1987C):

- (i) Some enhancers were molecules that are constituents of the cranberry cuticle such as ursolic acid. These molecules are able to recognize the cuticle surface, stick to it, and thus allow the cuticle to become permeable to Ethrel.
- (ii) Some enhancers were organic solvents such as ethanol (alcohol). These molecules stick to the waxy surface thus temporarily changing its property. In their presence cuticle became permeable to Ethrel.
- (iii) Some enhancers were highly lipophilic (fat loving) molecules. These molecules are able to get into the waxy surface and take along the Ethrel across the cuticles. Example of these molecules are lysophosphatidylethanolamine and urea.

These transport enhancers were field tested and were demonstrated to be effective in color enhancement. With some of these mixtures we were able to increase anthocyanin content by 50% over the untreated controls in 10-15 days after application.

**4. Development and Large Scale Field Testing of an Ethrel Formulation which is Consistently Effective in Cranberry Fruit Color Enhancement.**

Based on several field trials we selected the cheapest and one of the most effective formulations of Ethrel for further field testing. This formulation consisted of a mixture of Ethrel, ethanol (ordinary alcohol), urea and a spreader (detergent). This formulation has been tested on a large scale in Searls and Stevens cranberries. The results show that we are able to consistently increase the anthocyanin content from 40 to 100% over the untreated controls in about two weeks (Tables 1, 2, 4). These results are based on the last four years of field experiments. Results from last two seasons are shown in Tables 1, 2 and 4. In addition to a dramatic increase in average anthocyanin content all the fruits were improved in color (Tables 3 and 5) which means the fruits underneath the canopy (white and blush stages) were also improved in color. For example, in 'Stevens' no white fruits were present in the treated area.

**5. Our Recommendations.**

Based on our large scale field testing we recommend the following:

- 1 gallon Ethrel/acre
- 1-2 gallon Alcohol (ethanol)/acre
- 1 pint detergent (Tergitol)/acre
- 6 lbs. urea/acre

These chemicals are dissolved and applied as spray at the rate of 200 gallons of solution per acre. This amount of spray solution ensures that the chemical reaches the uncolored berries underneath the plant canopy.

**6. Development of a Procedure for Effectively Delivering the New Ethrel Formulation on the Cranberry Plants.**

In 1989 we devised a simple procedure for precise application of the new formulation on the bed. In this procedure the chemicals are directly injected into an individual irrigation line on the field (figure 1). This approach consisted of mixing the chemicals in a 5 gallon soda tank. By attaching this tank to a pressurized carbon dioxide cylinders we were able to inject the chemicals right into an individual irrigation line in the bed. By adjusting the pressure, it was possible to vary the speed of injection. Total amount of water applied was about 200 gallons/acre.

**7. Use of New Ethrel Formulations for Color Enhancement in Cranberry Fruit: Future Perspectives**

Ethrel is currently marketed by Rhone-Pulenc in USA. This company is not interested in maintaining the Ethrel label for cranberries. However, it is possible to obtain third party label of Ethrel for cranberries with the permission of Rhone-Pulenc. We have had discussions with Mr. Jere Downing of Ocean Spray Inc. and with Dr. John Pickles of the Hopkins Chemical Co. (UAP). There seems to be interest in pursuing this option for Ethrel use in cranberries.

**8. Alternative to Ethrel: Potential use of new (environmentally safe) Natural Lipids for Improvement of Cranberry Fruit Color and Improvement of Keeping Quality of Fruit During Storage.**

In the last two years we have investigated the use of natural lipids such as lysophosphatidylethanolamine (LPE) for improvement of fruit color in cranberries (Farag and Palta, 1989). This lipid is known to be present in all biological systems and is currently purified from egg yolk.

We have found that LPE is able to promote natural production of ethylene in fruit which in turn stimulates color production. We do not know at present the exact mechanism by which these natural lipids stimulate fruit ripening. The most interesting property of these lipids is that they are able to improve color as well as keeping quality of the fruit. During 15 weeks of cold storage after harvest the cranberry fruits sprayed with lipids had much less soft and rotted fruits compared with controls. We plan to conduct further research on the use of these lipids for fruit color enhancement and for improving fruit storability.

**9. Conclusions**

- (i) Cranberry fruit surface (cuticle) is essentially impermeable to Ethrel.
- (ii) In the presence of transport enhancers the Ethrel is able to permeate across the fruit cuticle. These enhancers are environmentally safe natural products.
- (iii) A new formulation of Ethrel containing ethrel, urea and spreader (detergent) has proved to be consistently effective in improving fruit color.
- (iv) A simple system utilizing 5 gallon soda-tanks and pressurized carbon dioxide has been devised to deliver new formulation effectively in the cranberry field.

(v) Some natural lipids show potential for improving fruit color and fruit storability. The use of the chemicals as an alternative to Ethrel is currently being investigated.

10. **Acknowledgements**-- We would like to acknowledge Whittlesey and Dubay Cranberries for providing facilities for this work and Brian Bowan, Assistant Superintendent Hancock Experiment Station, for help with field application by chemigation.

11. **Literature cited.**

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Shawa, A.Y. 1979. Effect of ethephon on color, abscission, and keeping quality of 'McFarlin' cranberry. HortSci. 14:168-169.

Table 1. The effect of field applications of new ethephon formulation on the anthocyanin content of Searles cranberry fruit. Chemicals were applied by chemigation method on Sept.18, 1989 and harvested on Oct.6, 1989.

Table 2. The effect of field application of new ethephon formulation on the anthocyanin content of Stevens cranberry fruit. Treatments were applied by chemigation method on Oct. 1st, 1989 and harvested on Oct.13, 1989.

Treatments	Fruit Anthocyanin Content (mg/100 g fr.wt) at 10 Locations									
	1	2	3	4	5	6	7	8	9	10
Control	19.83*	25.05	25.17	24.85	22.42	28.09	21.72	21.96	26.66	29.71
	22.73	23.50	25.83	25.40	22.18	27.67	22.74	21.50	27.86	29.14
	21.66	20.14	20.02	26.46	24.13	28.85	22.85	22.65	22.10	27.14
	21.21	20.76	19.79	26.56	23.64	28.55	22.44	23.18	23.60	26.85
Mean	21.36	22.36	22.70	25.82	23.09	28.29	22.44	22.32	25.06	28.21
<b>Overall Mean ***24.17±0.80</b>										
Ethephon+	39.42	46.93	39.49	44.59	44.68	36.50	35.96	30.44	31.92	
Tergitol+Urea	41.64	46.99	41.76	41.57	40.99	34.84	32.20	30.79	34.08	
+Ethanol	39.26	44.81	39.10	45.20	36.79	35.22	40.61	35.80	32.04	
	39.29	41.35	43.88	45.77	36.50	45.87	42.37	34.32	31.82	
Mean	39.90	45.02	41.06	44.28	39.74	38.11	37.79	32.84	32.47	
<b>Overall Mean ***39.02±1.46</b>										

\* Anthocyanin content (mg/100 g fresh weight)

\*\* Mean of 40 separate observations±SE.

\*\*\* Mean of 36 separate observations±SE.

Table 3. Percentage of degree of coloration of Searles cranberry fruit at harvest as influenced by field application of the new ethephon formulations using the chemigation method.

Treatments	Extent of Fruit Coloration (%)		
	White to 10% Blush	Medium	100%
<b>Water</b>			
Fruit Number	13.17	57.53	29.30
Fruit Weight	11.77	58.02	30.21
<b>Ethepron+Tergitol +urea+ethanol</b>			
Fruit Number	1.04	16.84	82.12
Fruit Weight	0.98	15.83	83.18

Table 4. The effect of preharvest spray of new ethephon formulation on anthocyanin content of Searles cranberry fruit. Treatment was done by injecting the chemicals into the irrigation line. Chemicals were applied on Sept. 28, 90 and fruits were harvested on Oct. 7, 90. Location of the experiment: Whittlesey Cranberries, Wisconsin Rapids, WI. Anthocyanin analyses were done by Ocean Spray Cranberries Inc., Babcock, WI.

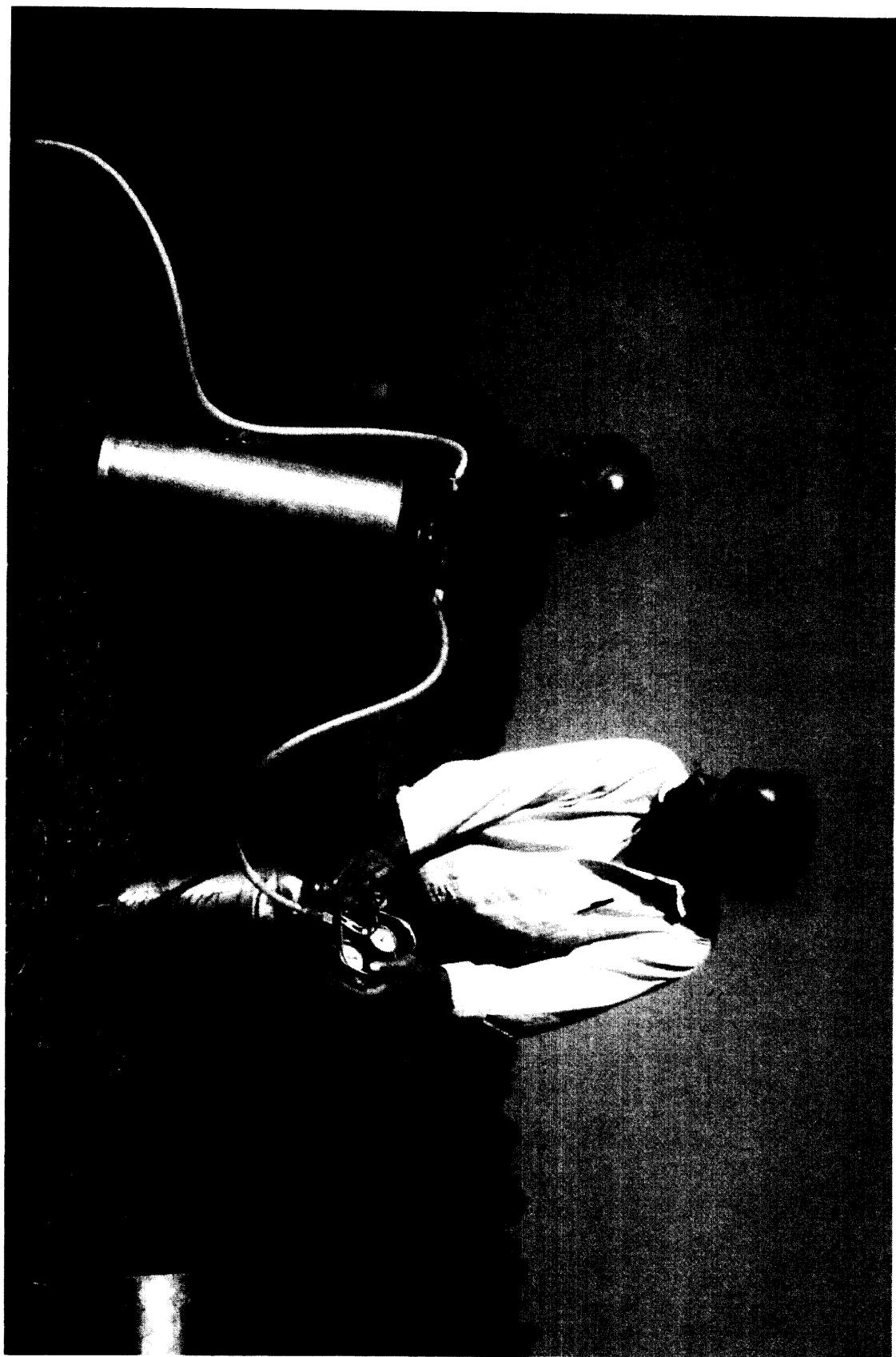
Treatments	Anthocyanin Content (mg/ 100 g)
Water	16.4 ± 0.8
Ethrel+Tergitol+Ethanol+Urea	25.1 ± 1.5

Table 5. Distribution of fruit anthocyanin content at various locations in the field as influenced by the application of new ethephon formulation at Whittlesey Cranberries in 1990 season.

Treatments	% of the Fruits in Various Classes of Anthocyanin Content (mg/ 100 g)							
	13-15	16-18	19-21	22-24	25-27	28-30	31-33	34-36
Water	45.4	27.3	27.3	0	0	0	0	0
E+T+U+EtOH	0	0	36.3	0	27.3	27.3	0	9.1

Abbreviations: E, Ethrel; T, Tergitol; U, urea; EtOH, ethanol.

Figure 1.



## CRANBERRY TIPWORM: PRELIMINARY RESULTS OF 1990 SANDING STUDIES

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Cranberry tipworm, *Dasineura oxycoccana*, is a tiny insect which damages the growing tips of cranberry uprights. Damage late in the season kills the fruiting bud for the following year, thereby reducing yield. Information on the biology and control of this insect was presented by Mahr and Kachadoorian in the "Proceedings: 1990 Wisconsin Cranberry School".

One on the approaches to tipworm control is the use of winter sanding to cover up the overwintering tipworm. Wisconsin growers have had varying success with sanding, which prompted a research project to evaluate this practice. This paper presents first-year results of the effects of sanding on tipworm populations. I also present data on fruiting status of first-year regrowth uprights from previous tipworm injury.

### Methods Used in the Study

The study was conducted at the Meadow Valley Division of Northland Cranberry Company, 4 miles northwest of Mather, WI. The bed of Howes where the study was conducted had a past history of significant tipworm injury. The width of the bed was divided into four approximately equal strips. The center two strips were sanded by conventional means during the winter of 1990-91, with 1/2 - 3/4 inch of sand being applied. The two outer strips were left unsanded. Each strip was divided in half length-wise, resulting in four sanded plots and four unsanded plots.

Sampling for tipworm activity was conducted twice, on 20 June and 6 July. These dates coincided with approximate times of first and second generations, as determined in other studies. However, there is some overlap of generations. In each of the eight plots, four sites were arbitrarily chosen, each at least four feet from the edge of the plot. At each site, a handful of at least 25 adjacent uprights were removed for laboratory analysis. In the laboratory, 25 uprights were arbitrarily selected from each sample site and microscopically examined for tipworm presence and damage. Therefore, the total sample for each of the eight plots consisted of 100 uprights.

In addition to tipworm infestation, I also assessed the impact on flowering of the previous year's (1989) tipworm damage. Each of the 100 terminals per plot (800 terminals total) from the 6 July sample, were evaluated for 1989 tipworm damage, subsequent regrowth of side branches, and flowering characteristics of undamaged terminals vs. regrowth terminals. For each terminal, records were kept of branching and combined total of buds, flowers, and set fruit (alled "flower units").

## Results of Sanding

Sanding significantly reduced the amount of immature stages (eggs, larvae, pupae, and spent cocoons) of tipworm found (Fig. 1) and number of terminals damaged (Fig. 2). In the unsanded plots, 33% of the terminals were infested or damaged in the first generation, as compared to only 4% in the sanded plots. Infestation levels increased in second generation, but the sanded plots had only half the level of infestation of the unsanded plots.

Although it would be natural for some increase between first and second generations, even in the sanded plots, the observed increase from 4% in first generation to 36% in second generation is somewhat problematical, and could result in part from the narrowness of the plots and close proximity of the large population in the adjacent unsanded strips. The tiny winged adult midge is capable of flight and being borne on wind currents, and reproductive females undoubtedly moved from the unsanded strips into the sanded strips. A study using entire beds as plots would be necessary to resolve this question.

## Effects of Tipworm Damage on Regrowth and Flowering

Undamaged terminals rarely spontaneously branch from previous year's growth. Of the 800 terminals examined on July 6, 562 (70%) had been undamaged the previous year. Of these, only 13 (2%) had branched from previous year's growth; 11 were singly branched and 2 were doubly branched.

The majority of the terminals damaged in 1989 developed a single regrowth terminal. Of the 238 damaged 1989 terminals, 209 (88%) had a single regrowth terminal, 28 (12%) were doubly branched, and 1 (<1%) was triply branched. Therefore, the 238 terminals which were damaged in 1989 gave rise to a total of 268 terminals in 1990.

In summary, undamaged terminals which branched spontaneously resulted in a net increase in terminal density of 2.7%. In contrast, regrowth from damaged terminals resulted in a net increase in terminal density of 12.6%.

Although tipworm damage resulted in an increase in terminal density, of more importance is the fruiting productivity of the regrowth terminals. Therefore, the total numbers of flower units (buds, flowers, or fruits at the time of sampling) was compared between the undamaged terminals from 1989, and the regrowth terminals resulting from 1989 damage. Of the 562 undamaged terminals from 1989, 196 (35%) produced flowers. In contrast, of the 268 regrowth terminals from 1989 damage, only 3 (1%) produced flowers. A total of 459 flower units were produced on the 562 undamaged terminals, an average of .82 flowers per terminal. In contrast, the 268 regrowth terminals produced only 6 flowers total.

## Implications for Tipworm Management

The results reported above, and the following interpretations apply to the set of conditions in this study. Tipworm damage to other varieties, or in combination with other crop or pest management practices may result in a different outcome. However, the general implications should apply in similar situations. It is evident that 1/2 - 3/4 inch of winter-applied sand significantly reduces first generation

tipworm the following spring, with an equally significant reduction in damage. In the research reported herein, second generation tipworm populations were about 1.8 times higher than first generation in the unsanded plots. In the sanded plots, the second generation population was about 16.7 times higher than first generation. This suggests that there may have been significant migration of reproductive females from the unsanded plots into the sanded plots. This has important implications for managing sanding as a tipworm control method. Instead of sanding randomly or arbitrarily chosen beds in any given year, an effort should be made to sand contiguous beds. Sanding all beds in a block in one year will reduce the rate of reinfestation from adjacent beds.

Prof. Marucci (cranberry entomologist retired from Rutgers University and the New Jersey cranberry experiment station) reported that early season tipworm damage created regrowth terminals that eventually became fruiting uprights. In the study reported herein, regrowth uprights in their first full year produced virtually no flowers. Therefore, without control of tipworm, it would be expected that such vegetative terminals would continue to be damaged, resulting in an overall reduction of fruiting uprights. The fact that only 35% of the undamaged uprights in this study produced flowers supports this conclusion. The ultimate conclusion is that damaging levels of tipworm should be reduced by pest management practices.

#### Acknowledgements

*I thank John Stauner of Northland Cranberry Company for providing the research site and for helping to coordinate this study. Richard Ness conducted the laboratory counts. This research was funded, in part, by a grant from the Wisconsin Cranberry Board, Inc.*

FIGURE 1. NO. OF TIPWORM  
PER 100 TERMINALS

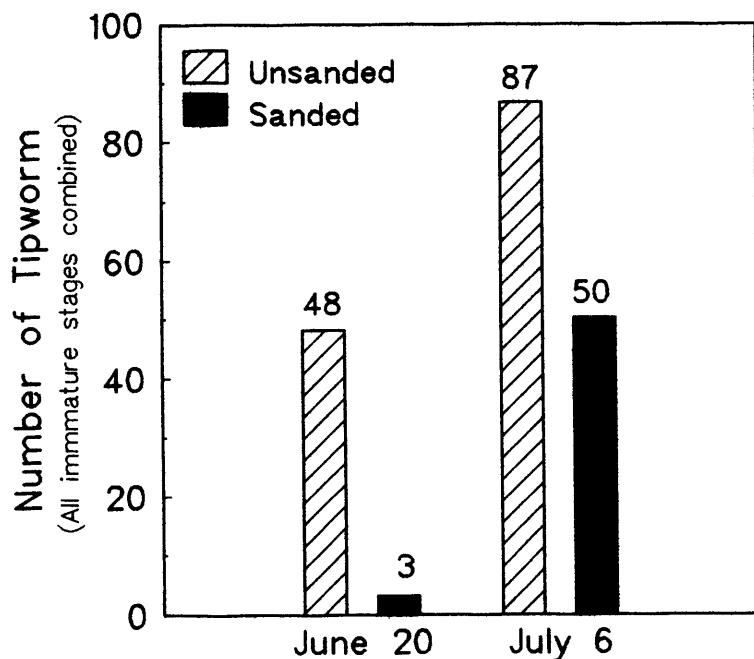
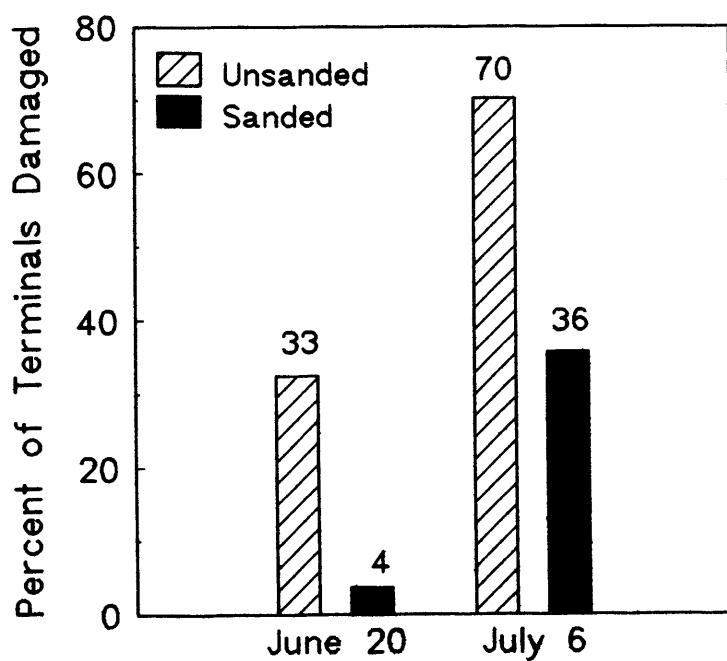


FIGURE 2. PERCENT OF TERMINALS  
DAMAGED BY TIPWORM



## Field Evaluation of Thermal Models to Predict Blackheaded Fireworm Egg Hatch

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Blackheaded fireworm remains a major pest in many of the cranberry growing regions in the United States and Canada. In both annual generations, adults and larvae can be monitored in an IPM program, but the very young larvae are difficult to detect. Since these larvae are the desired targets, a model of egg hatch would be useful to anticipate the optimum time of control measures.

Rose Kachadoorian and Daniel Mahr have developed models for the hatch of overwintering and summer generations based on laboratory studies. The purpose of this paper is to evaluate the linear degree-day models with field data. Specifically, the objectives are (1) to determine what information can be gathered in a commercial cranberry marsh for the model, and (2) to evaluate the degree-day models for both generations.

### Overwintering Eggs

As cranberry marshes are flooded to protect the plants from desiccation, flooding alters the environment for overwintering eggs as well. To understand the environment surrounding overwintering eggs, temperature was monitored in a marsh from the time of egg laying until the end of hatch. Daily maximum and minimum air or water temperatures were recorded, depending on which matched the environment of the eggs. By scientific convention, all measurements in this and other experiments were made in degrees Celsius. Values in degrees Celsius and their equivalents in degrees Fahrenheit are presented at the end of the text (Appendix A).

Judging from the stability of the temperatures measured during the winter (Fig. 1), ice protects eggs from exposure to extreme cold or untimely warmth. In spring when the ice melts, eggs are exposed to air temperatures suitable for development. The date when all the melted ice is drained, what can be called the Water Free Date, becomes a logical time to begin measurement of air temperatures for a hatch model. If the marsh is reflooded, that period of reflood is disregarded in degree-day calculations.

We monitored egg hatch for the overwintering generation at four commercial cranberry marshes in 1990. After the flood was removed, 30 to 100 eggs were located and daily maximum and minimum sheltered air temperatures were recorded near the eggs. Eggs were checked approximately three times per week until all had either hatched or desiccated.

Because of differences in management and climate between sites, the distribution of hatching was unique in each marsh (Fig. 2). On all marshes, hatching lasted for many weeks.

A linear model for hatch of overwintering eggs generated from laboratory results predicted the date of 50% hatch. Degreedays were computed with the sine wave method using a lower threshold of 10°C (50°F) and an upper threshold of 31°C (88°F).

In general, the actual date of 50% hatch occurred much sooner than the predicted date of hatch (Table 1). Probably the low temperatures during the spring fell within the area of poorest fit for a degree-day model. Also, simply indicating a date of hatch is not useful for an activity completed over several weeks. A non-linear distribution model based on laboratory results may fit the data better.

As an alternative to laboratory-generated models, a model of the distribution of hatching can be constructed directly from field data. Such a model was constructed and is presented with the data (Fig. 3). In the future, we will test if this alternative model is a better predictor than the laboratory model has shown to be.

### Summer Eggs

To evaluate the summer model, we established groups of eggs to sample. Egg-laying moths were caged over uninfested cranberry vines for 24 hours. Three groups of eggs were started a week apart, then 30 to 100 eggs from each group were located and monitored as in the other experiment.

In the summer generation, hatch occurred within a few days because of the warm temperatures (Fig. 4). There were differences in time of hatch for the three groups of eggs because of differences in temperature between the three weeks.

We evaluated the linear degree-day model for summer eggs by comparing the predicted date of hatch to the date of 50% hatch. The summer egg model had a base temperature of 8°C (47°F), an upper threshold of 31°C (88°F), and required 79 Celsius Degreedays (174 Fahrenheit Degree-days) until hatch. There was a small deviation from the predicted date of hatch and the date of 50% hatch (Table 2). On average, the deviation was one day, which means the model is as reliable as possible.

### Conclusions

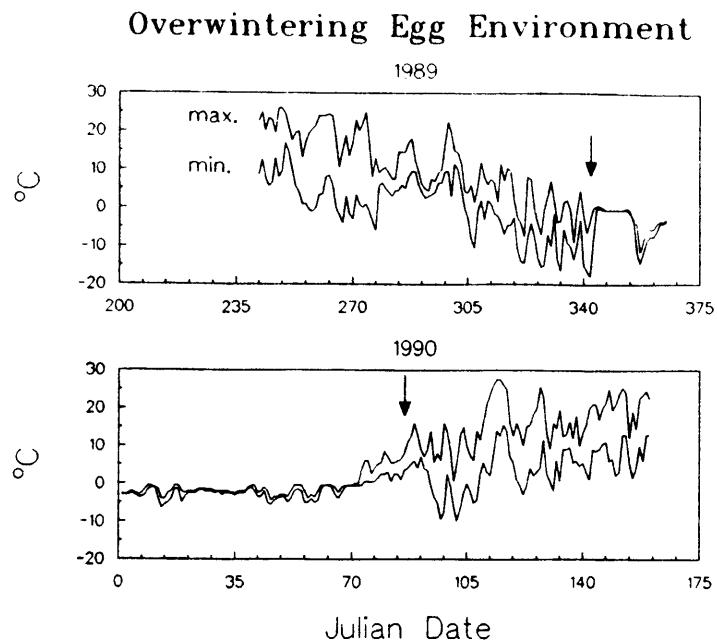
The maximum and minimum daily temperatures after the water is removed in the spring are the most practical measurements to use in a model of egg hatch. A laboratory model for summer egg hatch works very well. Practical application of the summer model will depend on a companion model that correlates egg laying and pheromone trap catches. A field-generated model for hatch of overwintering eggs will be evaluated in 1991 in Wisconsin and other cranberry growing regions.

### Acknowledgments

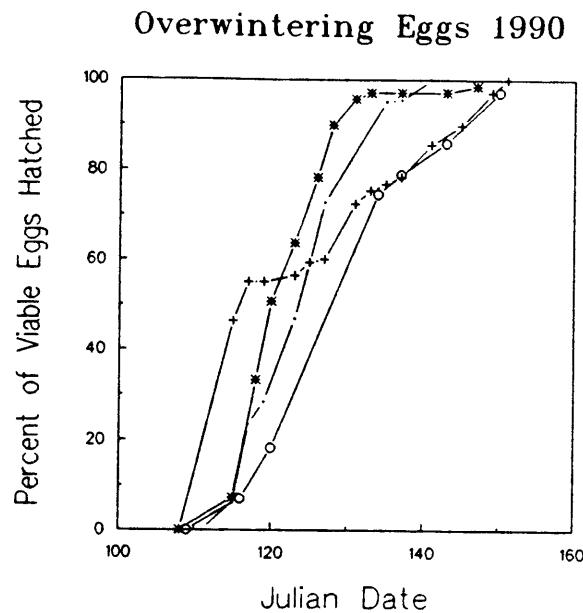
*We thank Charles Strozewski, Bob Duckart, Harold Mezera, and Carl Plaza for allowing us to do research on their marshes. We thank Sara Ott, Marc Gehl, and Richard Ness for field assistance. This research was supported in part by a grant from Ocean Spray Cranberries, Inc.*

**Appendix A.** Temperatures in degrees Celsius and their equivalents in degrees Fahrenheit.

Degrees C	Degrees F	Degrees C	Degrees F
-20	-4	5	41
-15	5	10	50
-10	14	15	59
-5	23	20	69
0	32	25	77
		30	86



**Figure 1.** Daily maximum and minimum air or water temperatures in a cranberry marsh when overwintering blackheaded fireworm eggs are present. Julian dates are the order of days in a year starting with January 1. Arrows indicate the start of winter flood and removal of flood.



**Figure 2.** Percent hatch of overwintering eggs at four marshes in 1990. Data for each marsh is represented by a different symbol. Julian date 110 is April 20, 120 is April 30, 130 is May 10, and 140 is May 20.

Table 1.

COMPARISON OF DATE OF 50% HATCH FOR FOUR OVERWINTERING COHORTS  
AND PREDICTED DATE OF MEAN HATCH.

SITE	DATE OF 50% HATCH 1	PREDICTED HATCH DATE 2	DEVIATION (DAYS)
MONROE Co.	4 MAY	8 MAY	4
WOOD Co.	26 APRIL	25 MAY	29
PORTAGE Co.	30 APRIL	14 MAY	14
RUSK Co.	8 MAY	23 MAY	15
MEAN			15.5

1 ESTIMATED BY EXTRAPOLATION.

2 DEGREE-DAY MODEL, KACHADOORIAN AND MAHR.

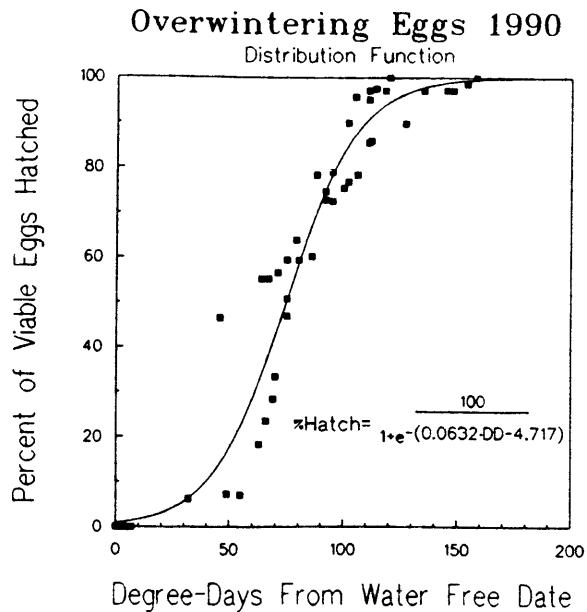


Figure 3. Alternative model for hatch of overwintering eggs generated from field data. The equation is an s-shaped curve, which approximates the measurements of egg hatch represented by squares. Degree-days are in degrees Celsius and are calculated the same as the laboratory-generated model.

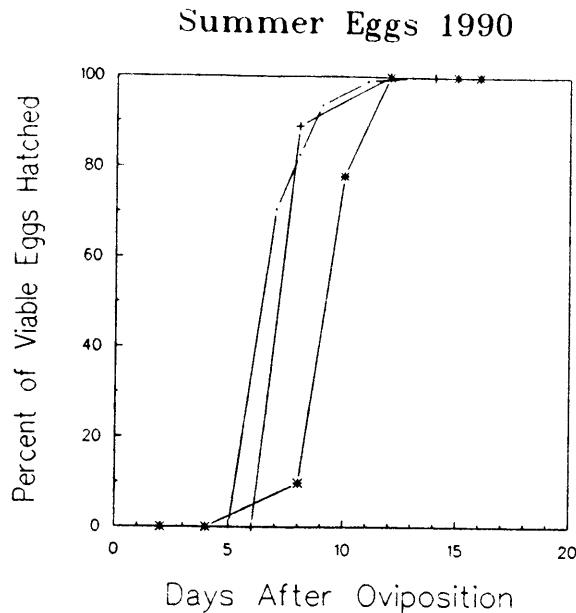


Figure 4. Percent hatch of summer eggs starting from the day of oviposition. Three groups of eggs are represented by different symbols.

Table 2.

COMPARISON OF DATE OF 50% HATCH FOR THREE SUMMER COHORTS AND PREDICTED DATE OF MEAN HATCH.

OVIPOSITION DATE	DATE OF 50% HATCH 1	PREDICTED HATCH DATE 2	DEVIATION (DAYS)
28 JUNE	4 JULY	3 JULY	1
6 JULY	13 JULY	13 JULY	0
10 JULY	19 JULY	17 JULY	2
MEAN			1

<sup>1</sup> ESTIMATED BY EXTRAPOLATION.

<sup>2</sup> DEGREE-DAY MODEL, KACHADOURIAN AND MAHR.

## EFFECT OF TWO OVERWINTERING TEMPERATURES ON EGG VIABILITY AND HATCHING TIME OF BLACKHEADED FIREWORM (*Rhopobota naevana*)

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### INTRODUCTION

The cultural practice of flooding cranberry beds during the late fall affects both the cranberry plant and pests. This project examined if the winter flood used to protect the buds of the cranberry plant may also protect blackheaded fireworm (BHFW) eggs from unfavorably low temperatures. We assessed (1) if egg mortality is increased when eggs are exposed to below freezing temperatures for extended periods of time (2) if egg hatch is delayed by low overwintering temperatures and (3) if prolonged post-freezing chilling affects egg survival (i.e., if a lengthy period of cold spring weather is detrimental to BHFW eggs).

### METHODS

We collected 2,000 BHFW eggs from a Monroe County marsh in October, and stored them at 39°F for 3 months. In mid-January half of the eggs were placed at 32°F (0°C), and the other half at 14°F (-10°C). The eggs were held at these temperatures for various lengths of time and then returned to 39°F ("post-freezing chilling") for different lengths of time. After the freezing/chilling treatments, eggs were transferred to 73°F and hatch recorded.

Different groups of eggs were held at the two freezing temperatures for 1, 2, 4, 6, 8 and 10 weeks. Subgroups were then chilled 1, 2, 4, or 6 weeks before transferal to 73°F.

### RESULTS AND DISCUSSION

#### NUMBER OF EGGS HATCHING

##### Freezing Time

Exposure to 32°F simulated temperatures beneath the ice, and the 14°F temperature reflected colder conditions that might occur without the winter flood. Significantly fewer eggs held at 14°F hatched than eggs exposed to 32°F (Figure 1) except for the eggs held at 14°F only one week. Short term exposure to this colder temperature did not negatively affect hatch as did long term exposure.

The length of time eggs were held at 14°F significantly influenced egg viability. As length of freezing time increased, percent egg hatch decreased (Figure 2). In contrast, the length of exposure time at 32°F (0°C) did not significantly influence egg hatch, although there was a slight downward trend as freezing time increased.

### Post-freezing chilling

Chilling at 39°F simulated the post-freezing chilling that would occur during cold spring weather. For eggs held at the 14°F temperature, as length of post-freezing chilling increased, the percent egg hatch decreased. Significantly fewer eggs hatched after being refrigerated for 6 weeks, than at 1, 2 or 4 weeks at all freezing time periods. Embryos stressed by cold temperatures often developed to a stage in which the black head capsule and body of the larva were visible microscopically within the egg. But the embryo would die instead of the egg hatching.

Eggs originally held at 32°F were slightly affected by post-freezing chilling, but not as severely as eggs held at 14°F. This suggests that a prolonged cold spring could be detrimental to egg viability, especially if eggs were previously exposed to unfavorably cold temperatures.

### HATCHING TIME

In most treatments, eggs frozen at 32°F took significantly less time to hatch than those frozen at 14°F. This suggests some type of development occurs at 32°F and not at 14°F, or that eggs are somehow stressed. Table 1 indicates the average number of days to egg hatch.

**Table 1.** Average number of days needed for blackheaded fireworm egg hatch at 73°F after various freezing and chilling regimes. These are selected examples. Comparisons should be made between rows (between 32° and 14°F)

		Weeks Frozen/Weeks Refrigerated						
Temp		1/1	1/2	2/1	2/2	4/2	6/1	10/2
32°		6.9	6.5	5.1	6.2	5.4	5.4	5.2
14°		8.7	7.0	7.1	7.7	7.4	8.4	7.3

Eggs frozen for only 1 week at either 32°F or 14°F tended to take longer to hatch than eggs frozen longer. This indicates eggs must be exposed to cold temperatures for a certain minimum period of time to break dormancy.

Most of the eggs exposed to the 32°F freezing temperature hatched within 3 to 5 days of being transferred to 73°F (Figure 3). In contrast, most of the eggs exposed to 14°F required 6 to 8 days to hatch. Very few eggs from either freezing temperature required longer than 9 days to hatch.

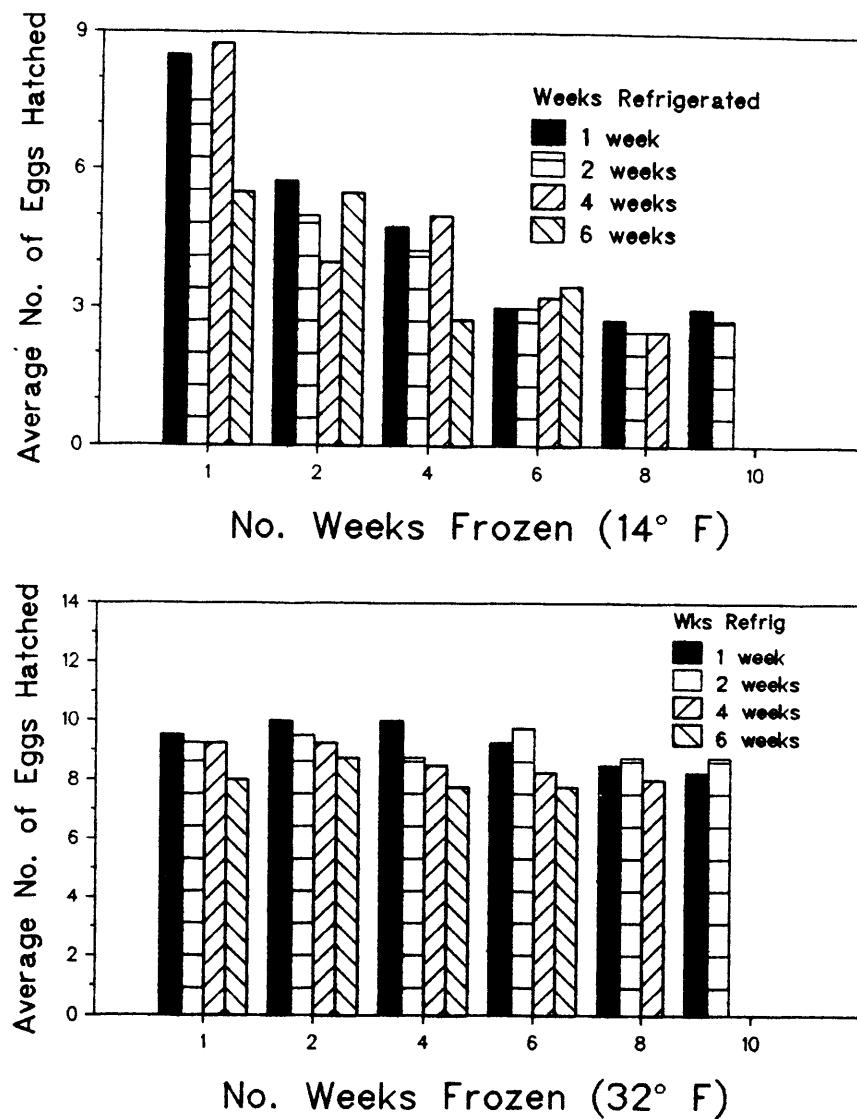


Figure 1. Fewer eggs hatched after exposure to the overwintering temperature of 14° F than eggs exposed to 32° F.

## CONCLUSION

At some point below 32°F, cold temperatures increase the mortality rate of overwintering BHW eggs. Prolonged cool weather after freezing has a detrimental affect on eggs especially if the eggs were previously exposed to unfavorably low temperatures. We also know from data collected from the UWEX Cranberry IPM Program that if temperatures rise rapidly in the spring and eggs hatch prematurely, significant larval mortality will occur because of freezing night temperatures.

It is probable that some development occurs at 32°F but not at 14°F. The possibility of some type of development taking place underneath the ice or development being inhibited by cold temperatures complicates the use of models to predict egg hatch.

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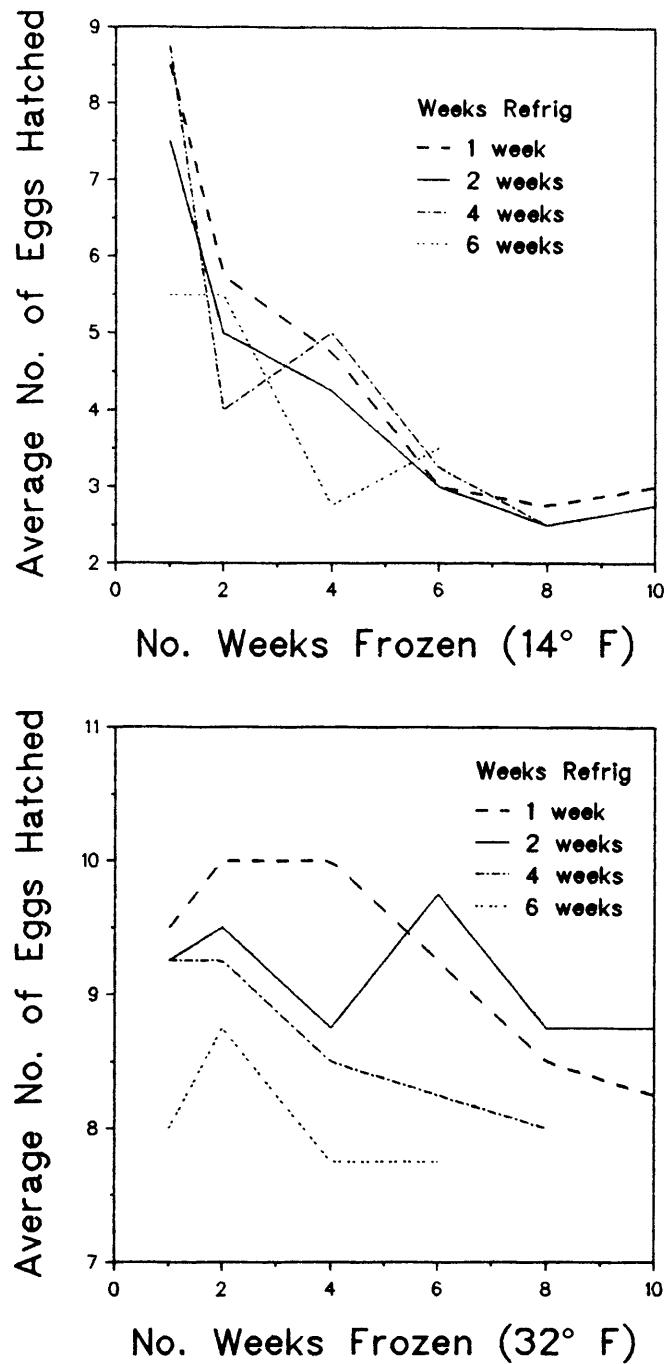


Figure 2. The length of time eggs were held at 14° F influenced egg viability. As the length of freezing time increased, percent egg hatch decreased. The length of time eggs were held at 32° F did not significantly influence egg hatch.

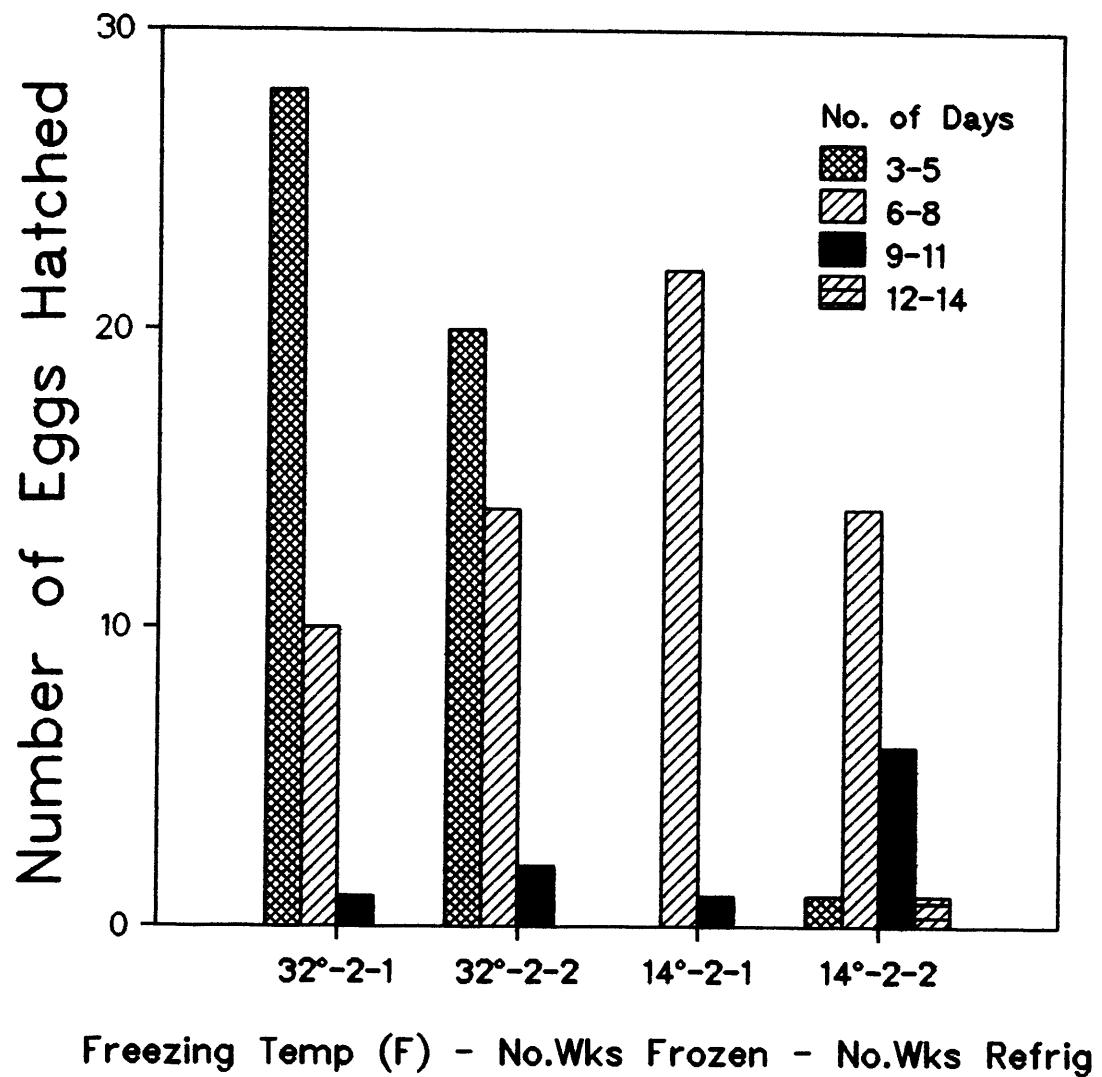


Figure 3. Most eggs exposed to 32° F freezing temperature hatched within 3 to 5 days of being transferred to 73° F. In contrast, most eggs exposed to 14° F required 6 to 8 days to hatch. Very few eggs from either freezing temperature required longer than 9 days to hatch.