Establishment and Dispersal of *Puccinia thlaspeos* in Field Populations of Dyer's Woad

Bradley R. Kropp, Dane R. Hansen, and Sherman V. Thomson, Department of Biology, Utah State University, Logan 84322

ABSTRACT

Kropp, B. R., Hansen, D. R., and Thomson, S. V. 2002. Establishment and dispersal of *Puccinia thlaspeos* in field populations of dyer's woad. Plant Dis. 86:241-246.

Dyer's woad rust is being intensively studied as a biocontrol agent for the noxious weed dyer's woad. These studies report on methods to establish this fungus in woad populations and to obtain information about disease incidence and dispersal of the rust subsequent to establishment. Inoculum dosages as low as 1 mg/plant established disease in dyer's woad rosettes. Dosages higher than 1 mg/plant resulted in better levels of infection, and inoculation was equally effective when the inoculum was placed directly on the plants or on the soil adjacent to the plants. The highest incidence of infection occurred when plants were inoculated in the spring, whereas summer or fall inoculations did not result in infections. Large-scale inoculation of woad was effective and resulted in significant levels of disease. However, it was not clear if artificial inoculation was effective in changing disease incidence in populations of woad where the rust was present prior to treatment because there were no significant differences between inoculated and check plots. Woad rust is systemic but apparently it is not seed dispersed, even though some infected plants occasionally produce seed. Two years after inoculation, dyer's woad rust had dispersed an average of 14.6 m from the focus of inoculation. After 3 years, the dispersal distance had not increased and infected plants averaged only 10 m from the point of inoculation. Disease was maintained naturally in woad populations after treatment, but the overall percent infection in the treated plots decreased by 3 years after inoculation.

Additional keywords: biological control, mycoherbicide, noxious weed

Dyer's woad (*Isatis tinctoria* L.) is a noxious weed in the *Brassicaceae* family that was introduced into North America from Europe during the colonial period. It is now present in both the eastern and western United States, although woad is not considered to be a weedy species in eastern North America. Woad is well adapted to arid conditions, produces abundant seed, and is allelopathic (14); therefore, it competes aggressively with the native plant communities in the dry western regions of the United States. Dyer's woad has become a serious problem and is considered a noxious weed in eight western states (3,5,6). Much of western North America is semiarid; therefore, woad has the potential to infest much additional habitat in the western part of the continent (5).

Corresponding author: B. R. Kropp
E-mail: brkropp@biology.usu.edu

This research was supported by the Utah Department of Agriculture, the Utah Agricultural Experiment Station, the USDA Forest Service, and the USDA Rangeland Research Program. This article is Utah Agricultural Experiment Station paper number 7338.

Accepted for publication 15 October 2001.

Control of dyer's woad in accessible areas can be accomplished by using herbicides or other traditional methods (7). However, much of the infested land is either inaccessible to machinery or of low value so that herbicide applications cannot be applied economically. In addition, some of the areas affected by dyer's woad are environmentally sensitive. Many of these areas are near urban centers and are either important as watersheds or receive heavy recreational use. Systematic application of chemicals for weed control in such areas is not allowed or is undesirable because of nonselective damage to the indigenous flora. Biological control provides a very attractive alternative for managing dyer's woad on these lands.

The dyer's woad rust fungus, identified as *Puccinia thlaspeos* C. Schub., has been demonstrated as an effective biocontrol agent of dyer's woad. It is a microcyclic, autecious species that is widely distributed in the Northern Hemisphere (2). *P. thlaspeos* was not known to be a pathogen of dyer's woad until 1978, when it was found on woad in southern Idaho (11). Since its discovery, woad rust has rapidly spread from Idaho into northern Utah, where it now occurs in most populations of dyer's woad. Dyer's woad rust differs from most other rust fungi used in weed biocontrol by causing systemic infections. These infections are usually asymptomatic during

the first year of the woad life cycle, with the fungus overwintering in the tissue of infected plants. Symptoms typically appear during the second season, before the plants bolt. An additional, and very important, difference between woad rust and other rusts used in biocontrol is that a single basidiospore infection by woad rust almost entirely prevents seed production by systemically infected plants (8,9,11). Dyer's woad rust appears to be an ideal biocontrol candidate and holds much promise as a method for reducing the impact of this weed.

The full potential of woad rust as a biocontrol agent will be realized only when we are able to optimally establish disease in wild woad stands. Good progress has already been made toward understanding how to do this. Flint and Thomson (8) found that potted dyer's woad plants placed in the field adjacent to rust infected dyer's woad plants became infected and that these infections occurred most often in the spring when teliospore and basidiospore production occurs abundantly. They were also able to achieve occasional autumn infections by providing inoculum in the form of potted plants bearing teliosori at a time when inoculum is not obviously present. Flint and Thomson (8) showed that woad populations can be inoculated artificially during the spring using crushed, dried leaf material bearing teliosori from diseased plants. However, information is still needed on optimal dosages for inoculation and whether the application of teliosori on crushed leaf material can be used to extend the time frame for inoculation beyond the period of natural basidiospore production. Teliosori and basidiospores are produced during spring and early summer; therefore, it would be very useful to be able to collect rusted leaf material early in the spring to be prepared for applications later in the year. Methods for large-scale inoculation of dyer's woad also need to be developed in order to utilize this rust in managing large areas of woad infestation.

For woad rust to be maximally effective as a biocontrol agent, it should be capable of spreading after inoculation and of causing sustained disease pressure in woad stands from one year to the next. Thus, information is needed on dispersal of the rust fungus after inoculation and on whether disease levels are maintained naturally subsequent to establishment in

the field. Dispersal of dyer's woad rust is only via basidiospores produced on infected plants, as far as we know. However, even though dyer's woad rust severely limits seed production by infected plants, some diseased plants occasionally produce mature seed. These seed often bear teliosori, leading to the question of whether they are capable of germination and of acting to disperse the rust fungus. This article presents results of studies to determine (i) methods for large-scale treatment of woad populations along roadsides, (ii) whether the level of disease obtained by inoculation is maintained in the field over time, and (iii) the dispersal rate and mechanisms of spread of woad rust subsequent to inoculation.

MATERIALS AND METHODS

Inoculum preparation and dosage determination. The inoculum used in all of the experiments was prepared as described earlier (8,9) from naturally infected plants bearing teliosori collected in the field in northern Utah during spring. After the plants were dried, the leaves were stripped from the stems by hand and ground with a Wiley mill into small fragments about 1.0 to 2.0 mm in size for use as inoculum.

Optimal dosages for inoculation were determined using field plots at the Utah State University Greenville Farm in North Logan. Seed from dyer's woad had been collected during the previous summer and the silicles had been removed. The seed were planted in mid-May and allowed to grow until rosettes had formed in mid-June, at which time the inoculations were done. Moisture on the leaves is important in establishing infections (10); therefore, the plants were watered before inoculation to insure that the inoculum was placed on moist plants. Inoculations also were carried out in the evening to take advantage of the cooler temperatures that have been shown to be optimal for teliospore germination (8). Each plot was 1.2 by 1.2 m in size, and three replicate plots were prepared for each inoculum dose being tested. The plots were placed 3 m apart to reduce drift or spread of inoculum and were arranged in a completely randomized design (13). The number of woad plants in each plot was

thinned after emergence to 10 plants/plot. The inoculum dosages tested were 1.0, 5.0, 10.0, 100.0, and 470.0 mg/plant, plus uninoculated controls. In addition, 470 mg of inoculum was placed on the soil within 2 cm of the plants to determine if it was necessary to have inoculum directly on the plants. The inoculum was applied by shaking it by hand onto the moistened leaves of the individual plants in each plot. Percent infection was determined the following April by counting the number of plants with typical symptoms of chlorosis and spermatia on the leaves. Differences in percent infection among treatments were analyzed using standard analysis of variance (ANOVA) procedures.

Large-scale inoculation. Two experiments were performed in naturally established dyer's woad populations along roadsides to test inoculation methods that could be used in large-scale treatments. In 1997, experimental plots were established in five replicate locations in northern Utah named Beaver Dam, Box Elder, Causey, Trenton, and Yacht. Woad rust was already present in these plots; therefore, transects were run through each plot to assess the background disease levels at the time of inoculation (Table 1). A total of four treatments were evaluated and each was replicated once at the five different sites using a randomized complete block design (13). One treatment was inoculum mixed with hydromulch. The hydromulch was used because of its moisture-absorbent properties, which would provide a moist environment for the teliospores to germinate and produce basidiospores over a longer period of time. The inoculum was mixed with the hydromulch in a ratio 626 g of inoculum to 91 kg of hydromulch. The mixture was added to 1,893 liters of water, along with 4.5 kg of tackifier (Alpha Plantago, Canadian Forest Products, Ltd., New Westminster, BC) and sprayed onto the woad plants by a commercial operator at a rate of 2.75 kg/ha. A water suspension of inoculum was also sprayed onto some of the woad plots using a 50-gallon John Bean PCO 50 hand gun sprayer (FMC Corp. San Jose, CA). Dry inoculum also was applied as a treatment at a rate of 2.75 kg/ha by hand because this method previously had been

shown to be effective in establishing disease in the field (8). Uninoculated plots served as controls. The plots varied in size from 93 to 186 m² depending on the area infested by woad. The effect of rust on dyer's woad population was evaluated for 2 years after inoculation by determining the incidence of rusted dyer's woad plants in the roadside areas that were inoculated.

In 1998, a second experiment was conducted near Park City, UT at a location where woad rust was absent (Table 1). For this experiment, some of the inoculated plots were sprayed with inoculum mixed into hydromulch (Mat Fiber Plus, Mat Inc, Floodwood, MN) at a rate of 2.75 kg/ha. The inoculum was mixed with the hydromulch in a ratio of 293 g of inoculum to 34 kg of hydromulch and the mixture was added to 757 liters of water and sprayed onto the woad plants using an Aqua Mulcher 200 (TGMI Inc., Cincinnati, OH) at a rate of 2.75 kg/ha. Uninoculated control plots and plots that were inoculated by hand with dry inoculum at a rate of 2.75 kg/ha also were established for this experiment. The experiment was done at a single roadside location where the treatments were each replicated six times and arranged in a completely randomized experimental design (13). The amount of disease was measured for both the 1997 and 1998 experiments by running transects through the plots 1 and 2 years following inoculation, and the data was analyzed using standard ANOVA procedures.

Inoculation time frame, maintenance, and dispersal. To determine whether the use of artificial inoculation could extend the time frame for treatment beyond spring and to study disease incidence and dispersal after inoculation, a site was chosen in City Creek Canyon near Salt Lake City, UT. This site was chosen because the woad population located at the site was free from natural infection by the woad strain of *P. thlaspeos*. Inoculation was carried out using inoculum prepared as described above from dried, diseased plants. The inoculum was applied dry and, to enhance disease establishment, a heavy inoculum dose of 470 mg/plant was applied.

Inoculations were done on 5 October 1995, 17 May 1996, and 19 July 1996. For each inoculation time, 20 paired plots were established in stands of woad distributed throughout the canyon. Of the 20 plots used for each inoculation date, 10 were treated and 10 were left as untreated controls. Each plot consisted of 10 plants clustered around a stake that had been placed in the ground. After the stake had been placed in the ground, the 10 plants closest to the stake in any direction were chosen for the experiment and the location of each plant was mapped by measuring its distance and direction from the stake. The plants in these plots were tightly clustered around the stake, so that each plot represented a focused inoculation point for dis-

Table 1. Disease incidence after inoculation of roadside populations of dyer's woad^x

Treatment	Northern Utah			Park City		
	1997 ^y	1998	1999 ^z	1998 ^y	1999	2000
Dry	32.2	40.0 a	15.3 a	0	20.1 a	52.9 a
Hydromulch	20.5	40.0 a	9.1 a	0	22.6 a	38.8 b
Water suspension	27.6	39.1 a	15.8 a	...	...	...
None	19.5	36.1 a	12.9 a	0	1.3 b	21.5 b

^x Disease incidence in each year is expressed as the percentage of diseased plants. Percentage of data were arcsin transformed before analysis of variance was performed. Values in columns followed by different letters are significantly different from one another ($P = 0.05$).

^y 1997 data for northern Utah and 1998 data for Park City represent baseline percent disease levels present at the time of inoculation.

^z In 1999, only four northern Utah sites were included in the analysis because no plants were present at the fifth site during that year.

persal studies. The results of the inoculations were determined as the percentage of the plants showing rust symptoms in the spring following treatment.

The 10 plots inoculated in May 1996 were used to study dispersal of the rust and maintenance of disease levels after treatment. To learn whether disease is maintained in the field after inoculation, the number of symptomatic plants within a 4-m diameter circle around the stake was determined in the spring of 1998 and again in the spring of 1999. By spring 1998, three of the plots had been lost due to construction or vandalism and therefore the measurements were completed on the remaining seven plots. By spring 1999, six of the original plots remained. Maximum dispersal distance was determined for each undisturbed plot during both years by measuring the distance between the stake at the center of the plot and the diseased plant most distant from the stake.

Seed dispersal. Seed were collected from both healthy and diseased plants during July and August of 1998 to determine whether seed from rust-infected plants can disperse the rust fungus. The seed from diseased plants were divided into three categories: seed bearing obvious teliosori collected from symptomatic branches (Fig. 1A), seed lacking teliosori collected from symptomatic branches, and seed obtained from asymptomatic branches produced by diseased plants. The seed were stored at room temperature until the silicles were removed, after which they were stored at 4°C until being planted in October 1998.

In all, 160 seed with teliosori collected from symptomatic plants and 100 seed from each of the other categories were grown to determine whether the rust could be seedborne. The seed were individually planted in random order in individual cells of flats containing Redi-Earth potting mix (Scott's Sierra Horticultural Products Co., Marysville, OH). One seed was planted per cell and the seeds were watered as needed until emergence. After emergence, the percent germination was determined and the seedlings were each transplanted into individual 100-mm plastic pots filled with Redi-Earth that were completely randomized. The plants then were grown at ambient greenhouse temperatures using supplemental halogen lighting to extend the photoperiod to 16 h. The plants were grown under these conditions and watered and fertilized as needed until September 1999. At that time, they were placed in the dark at 4°C for 6 weeks to stimulate bolting. Following the cold treatment, the plants were again grown in the greenhouse until June 2000, when the experiment was terminated. The plants were observed for symptom expression for the duration of the experiment.

RESULTS

Inoculation and dosage. During the spring following artificial inoculation,

symptoms of infection were visible in the field plots at all dosages of inoculum tested (Fig. 2). Dose level was significant at the $P = 0.014$ level, meaning that disease incidence increased with greater inoculum dosages. The best level of infection, 31.4%, was obtained when 470 mg of inoculum per plant was applied to the soil adjacent to the plants rather than directly on the plants. Applying 470 mg of inoculum to each plant resulted in about 40% lower infection, and this level was not significantly different from the treatment where inoculum was applied to the soil. At inoculum levels less than 100 mg, less than 10% of the plants became infected. No statistically significant differences were detected between any of the inoculum levels below 100 mg and none of them differed significantly from the control.

Large-scale inoculation. The results of roadside inoculations carried out during 1997 in northern Utah indicate that, on average, there was no significant increase in infection due to our inoculation treat-

ment (Table 1). Inoculation using dry inoculum and inoculum either suspended in water or mixed with hydromulch resulted in disease incidences of between 39.1 and 40.0% that were only marginally higher than the uninoculated controls. For this experiment, the background disease incidence at the time of inoculation was relatively high, averaging 26.4% for all five plots. By 2 years after inoculation, the overall disease incidence, regardless of treatment, had declined substantially in the northern Utah plots (Table 1). By 1999, the inoculated plots showed between 9.1 and 15.8%, while 12.9% of the plants in the uninoculated controls were infected. None of these values differed significantly.

The experiment done near Park City, UT showed that inoculating a woad population lacking background disease can significantly increase disease incidence (Table 1). For this experiment, inoculation resulted in a disease incidence of up to 22.6%. Very low disease incidence occurred in the uninoculated plots, which probably can be

Fig. 1. A, Teliosori (arrows) on the silicles of seed produced by a diseased dyer's woad plant. **B,** Germination of dyer's woad seed with teliosori (arrows) present on the integument surrounding the seed inside the silicle.

attributed to drift of inoculum from the treated plots. In this case, disease incidence had increased by 2 years after inoculation (Table 1). By spring 2000, the significant differences between the hydromulch-inoculated and uninoculated treatments had disappeared, probably as a result of natural spread of the rust fungus.

Inoculation time frame, maintenance, and dispersal. When artificial inoculation was performed at different times of the year, the only plots that developed disease were those inoculated in spring. Plots that were inoculated in May showed 66.3% infection. None of the plants inoculated in July or October developed symptoms.

Dispersal of the rust fungus from the disease foci established in the spring of 1996 in City Creek readily occurred, although variability was high among the plots (Table 2). The fungus dispersed an average of 14.6 m from the infection focus by spring 1998 but, by spring 1999, the fungus had not advanced further and the dispersal distance averaged only 10 m. The level of disease established by artificial inoculation of the plants was 66.3% 1 year

after treatment. However, subsequent to that, the amount of disease due to natural infection declined (Table 2). The number of diseased plants for 1998 and 1999 did not differ significantly from one another based on a paired *t* test ($P = 0.05$). The level of infection in 1997 was obviously higher but was not compared statistically with disease levels in 1998 and 1999 because the infections were initiated differently (i.e., by direct inoculation of a specified number of plants).

Seed transmission. Seed collected from healthy and diseased woad plants all germinated normally (Table 3, Fig. 1B) and all plants appeared healthy. Of the seed from healthy plants, 59% germinated and about the same ratio (63%) of the seed from asymptomatic branches of diseased plants germinated. When seed from symptomatic branches of diseased plants were collected, the seed on which teliosori were found germinated at a slightly lower percentage (41%) than those lacking teliosori (64%). Based on a χ^2 test, seed germination was independent of the source of the seed. There were no differences in

germination of seed attributable to disease. This supports the results obtained in an earlier study (9), where rust infection was also not found to affect seed germination on filter paper. However, in this study, the plants were grown after seed germination to learn whether disease transmission occurred. None of the plants grown from seed in this study showed symptoms of rust infection, indicating that dyer's woad rust is not spread via seed even when teliosori are present on the seed.

DISCUSSION

The results presented in this article show that woad rust can successfully be established in populations of dyer's woad that lack naturally occurring rust using methods appropriate for large-scale applications. Relatively low dosages are needed to initiate disease and, once established, the fungus is able to reproduce and disperse on its own, although augmentation may be needed to maintain constant high disease pressure in woad stands.

Natural infections by woad rust usually occur during the spring because both the teliospores and basidiospores of dyer's woad rust are produced in spring. Artificial inoculation also can be done successfully in spring using inoculum prepared from dry leaf material (8). Inoculum consists of leaves bearing teliospores that are collected in the field; therefore, it must be prepared when the teliosori are naturally produced. In cases where large volumes of inoculum are required, it would be difficult to both prepare the inoculum and apply it during a single spring season. Flint and Thomson (8) showed that a low percentage of infection could be achieved in autumn by exposing healthy woad rosettes to potted diseased woad plants purposely raised to produce basidiospores at that time of year. Thus, because it is possible to produce disease in autumn, it would be useful to be able to extend the time frame for artificial inoculation of woad populations to midsummer or fall. Unfortunately, the experiment carried out on time of year of

Fig. 2. Effect of inoculum dosage on disease establishment in dyer's woad. Dose level was significant ($P = 0.014$). Columns with different letters are significantly different from one another ($P = 0.05$). There was no significant difference in disease incidence resulting from applying 470 mg of inoculum on the plants and on the adjacent soil.

Table 2. Disease incidence and dispersal of woad rust in dyer's woad subsequent to inoculation

Season	Disease incidence (%) ^z	Distance dispersed (m)	
		Range	Average
1997	66.3	...	...
1998	22.3	5.6–28.7	14.6
1999	12.7	3.2–17.5	10.0

^z Disease incidence for the 1998 and 1999 seasons did not differ significantly from one another (paired *t* test, $P = 0.05$). The values for the 1997 season were higher but were not compared statistically with the other 2 years because disease was established the first year by direct inoculation of a specified number of plants.

Table 3. Germination and disease incidence of seed from healthy and infected dyer's woad plants

Seed source ^x	Germination (%) ^y	Disease incidence ^z
Diseased plants		
Sori	41	0
No sori	64	0
Asymptomatic	63	0
Healthy plants	59	0

^x Seed from diseased plants on which sori were either present or absent were compared with one another and with seed from asymptomatic branches of diseased plants and seed from healthy plants.

^y Percent germination was independent of seed source (χ^2 test, $P = 0.05$).

^z Disease incidence was monitored for 20 months after planting.

inoculation indicates that spring is the optimal time for inoculation and that inoculation may not be easily extended into other seasons. When one considers the biology of woad rust along with the climate in the western United States where woad is currently established, it is not surprising that spring inoculations are successful and that the summer inoculations failed. The optimal temperature for teliospore germination in this rust is 15°C and leaf infection is facilitated by free moisture on the leaf surfaces (8,10). Spring temperatures of around 15 to 20°C are usual throughout the region and precipitation is often highest at that time of year, creating nearly ideal conditions for infection. In contrast, summer precipitation is scarce and temperatures much above optimum may be expected. However, conditions in October, when our autumn inoculations were done, seem appropriate for the rust fungus, yet our autumn inoculations were unsuccessful. Autumn temperatures are often near optimum for spore germination and dew forms almost nightly on the woad. Dew was present on the leaves of the plants when our inoculations were carried out, although it might have been that the period of leaf wetness was too short to allow infection. The rosettes that were inoculated in autumn would, in most cases, be several months older than those inoculated in spring and might be more resistant to infection. Another possibility is that the physiological state of the plants, which are ready to lie dormant through winter, may be somehow less conducive to infection than in the spring.

The experiment to determine optimal inoculum dosages showed that the amount of disease increased as dosages increased and that the highest disease levels were obtained when plants were inoculated with relatively heavy doses of inoculum. The best results were obtained when 470 mg of inoculum was applied either to the soil adjacent to plants or directly on the plants themselves. The use of 470 mg of inoculum per plant would be prohibitive in practical terms because inoculum must be prepared by collecting diseased plants in the field, drying them, and then removing and grinding the leaves. This is time consuming and expensive and would act as a bottleneck to inhibit treatment of large areas of land infested with woad. However, levels of inoculum as low as 1.0 and 5.0 mg/plant were effective in establishing disease in the field plots. Uninoculated control plots also developed low levels of disease. This was unexpected because a distance of approximately 3 m separated the plots, but it can be attributed to drift of spores from inoculated plants. In any case, the appearance of disease in the uninoculated controls and the ability to establish disease using low levels of inoculum indicate that heavy applications of inoculum

are not necessary to use woad rust as a biocontrol agent. Once the disease is established in field populations of woad, even at low levels, it can be expected to spread on its own. It is also important to note that inoculum need not be applied directly to the foliage to be effective. Any efficient large-scale inoculation method would inevitably result in much of the inoculum landing on the ground between plants but, in this study, high rates of infection were seen when the inoculum was applied to the soil near the plants. This might be explained by moisture in the soil that would keep the inoculum moist for longer periods after inoculation and prolong basidiospore production.

Our results indicate that large-scale applications of woad rust inoculum can be effectively accomplished along roadsides. The clearest effect of inoculation was obtained when applications of inoculum were made in the Park City, UT area where the rust fungus was not present prior to inoculation (or present at a very low incidence). In this experiment, inoculum applied as a mixture with hydromulch was not more effective than dry inoculum applied by hand. However, the inoculation treatments were done during unusually wet weather and this might have negated the potential advantage of using hydromulch to maintain moisture levels of the inoculum. In the northern Utah studies where the rust fungus was present prior to inoculation, there was no significant effect attributable to inoculation. The inability to demonstrate a significant inoculation effect at these sites might have been due to the spread of the rust basidiospores for greater distances than anticipated from the inoculated to the uninoculated plots. Alternatively, natural infection sources contributed more inoculum than anticipated under the conditions of the test. Despite the lack of an experimental effect of inoculation on percent disease in the northern Utah sites, when the incidence of infected plants is examined, it is clear that the rust causes high levels of infection and has the potential to greatly reduce seed production. For example, at the Trenton location in northern Utah, the incidence of infected plants before inoculation was 75% but, in the year after inoculation, it had increased to 92% and, 2 years after inoculation, there were very few dyer's woad plants at the site. Most of the sites showed the same trend, although the incidence of infection was lower. Average disease incidence prior to inoculation for the northern Utah experiment ranged from around 20 to 30% and the average disease incidence for the northern Utah experiment increased for all treatments between 1997 and 1998, then dropped in 1999 (Table 1). Unfortunately, we don't know whether these population fluctuations were a direct effect of our inoculation or were caused by varying environmental conditions. It will take several more years of monitoring and

inoculation to reach a conclusion about the effectiveness of inoculating woad populations where rust is already present.

Effective use of woad rust as a biocontrol agent is dependent on the ability of the fungus to maintain disease pressure on the population over time and to disperse through the population after being applied. Dispersal from the disease foci that were established by artificial inoculation occurred but was slower than we had anticipated. In fact, the average maximum dispersal distance from the plot center was slightly lower after the third year than it was after the second year; perhaps in response to less favorable environmental conditions. Most of the dispersal observed in these plots was very localized. Initially, dispersal was over a relatively short distance and subsequent disease spread seemed to be mostly between adjacent plants within the infected population. Given this scenario, one might anticipate that the area infected by woad rust would expand slowly from year to year and that rapid long distance dispersal is unlikely. Yet, in a little over 20 years, woad rust has spread from the original source to as far south as Salt Lake City, UT, which is roughly 300 km from the site where it was first discovered in southern Idaho. To accomplish this, the fungus would have had to spread an average of 15 km/year, which is much more than one would anticipate from our dispersal data. The fungus apparently makes occasional long-distance jumps and then slowly disperses within the newly infected population. Long-distance dispersal of dyer's woad may occur by seed and it is reasonable to assume that the fungus might also be dispersed via seed from systemically infected plants. However, our results indicate that woad rust is not seedborne. Thus, long-distance dispersal of woad rust probably occurs relatively rarely under ideal conditions via basidiospores carried on air currents, or by other means. In contrast, long-distance dispersal of another well-studied rust, *P. chondrillina* Bubák & Syd., which has been used in biocontrol of *Chondrilla juncea* L., is rapid. It was reported to have spread as much as 320 km in a 9-month period following release in Australia, although dispersal after release in the United States was slower (1,4). Woad rust is microcyclic and thus can complete only one cycle per year, as opposed to a fungus like *P. chondrillina*, which can rapidly produce successive generations of uredospores. Artificial inoculation, especially at the expanding margins of the woad infestation, would compensate for this lack of rapid long-distance dispersal.

These studies show that dyer's woad rust is capable of maintaining disease pressure once established in the field by artificial inoculation. However, long-term work would be needed to evaluate the impact that inoculation with rust has on dyer's

woad populations. Our inoculation treatments and dispersal of the fungus can easily be evaluated by measuring disease incidence. To determine whether the fungus is really effective in decreasing woad populations, data need to be gathered over a period of years on seed production and recruitment of new plants in diseased stands. If disease levels in inoculated woad stands were maintained at the relatively low levels that our data indicate, the majority of the plants in the population would continue to produce seed and the population probably would never be completely eliminated. In this case, the system management approach (12) is a suitable model for woad and relies on the pathogen to reduce fitness of the weed rather than kill it. The fungus would thus weaken woad populations so that indigenous vegetation might be able to compete successfully. A more aggressive alternative would be to maintain higher disease levels in woad stands in accessible areas by repeated annual inoculations. A single inoculation may significantly reduce seed production, but escapes and new seedlings will develop and produce a new seed crop. We probably can achieve a higher level of biological

control of woad by using repeat or sequential applications of woad rust over several years. Multiple years of high infection rates could potentially reduce the available seed bank.

ACKNOWLEDGMENTS

We thank S. Durham for help with the statistical analyses.

LITERATURE CITED

1. Adams, E. B., and Line, R. F. 1984. Epidemiology and host morphology in the parasitism of rush skeletonweed by *Puccinia chondrillina*. *Phytopathology* 74:745-748.
2. Arthur, J. C., and Cummins, G. B. 1934. Manual of the Rusts in the United States and Canada. 2nd ed. Hafner Publishing Co., New York.
3. Callihan, R. H., Dewey, S. A., Patton, J. E., and Thill, D. C. 1984. Distribution, biology and habitat of dyer's woad (*Isatis tinctoria* L.) in Idaho. *J. Idaho Acad. Sci.* 20:18-32.
4. Cullen, J. M., Kable, P. F., and Catt, M. 1973. Epidemic spread of a rust imported for biological control. *Nature* 244:462-464.
5. Dewey, S. A., Price, K. P., and Ramsey, D. 1991. Satellite remote sensing to predict potential distribution of dyer's woad (*Isatis tinctoria*). *Weed Technol.* 5:479-484.
6. Evans, J. O. 1991. The importance, distribution, and control of dyer's woad (*Isatis tinctoria*). Pages 387-396 in: *Noxious Weed Plants*. L. F. James, J. O. Evans, M. H. Ralphs, and R. D. Child, eds. Westview Press, San Francisco.
7. Evans, J. O., and Chase, R. L. 1981. Dyer's woad control. *Utah State Univ. Ext. Bull.* EL-199.
8. Flint, K. M., and Thomson, S. V. 2000. Seasonal variation in infection of the noxious weed, dyer's woad, by a potential biological control rust, *Puccinia thlaspeos*. *Plant Dis.* 84:753-759.
9. Kropp, B. R., Hansen, D., Flint, K. M., and Thomson, S. V. 1996. Artificial inoculation and colonization of dyers woad (*Isatis tinctoria*) by the systemic rust *Puccinia thlaspeos*. *Phytopathology* 86:891-896.
10. Kropp, B. R., Hooper, G. R., Hansen, D. R., Binns, M., and Thomson, S. V. 1999. Initial events in the colonization of dyers woad by *Puccinia thlaspeos*. *Can. J. Bot.* 77:843-849.
11. Lovic, B. R. 1991. Two potential biocontrol strategies: *Puccinia thlaspeos* Schub. for *Isatis tinctoria* (dyers woad), and sugar beet associated bacteria for *Phoma betae*. M.S. thesis, Utah State University, Logan.
12. Muller-Scharer, H., and Frantzen, J. 1996. An emerging system management approach for biological weed control in crops: *Senecio vulgaris* as a research model. *Weed Res.* 36:483-491.
13. Steel, R. G. D., and Torrie, J. H. 1960. Principles and Procedures of Statistics. McGraw-Hill, New York.
14. Young, J. A., and Evans, R. A. 1971. Germination of dyer's woad. *Weed Sci.* 19:76-78.