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Apparent Herbivory and Indigenous Pathogens of Invasive Flowering Rush (*Butomus umbellatus* L.) in the Pacific Northwest

by Nathan E. Harms and Judy F. Shearer

INTRODUCTION: *Butomus umbellatus* L. (Butomaceae; flowering rush) is an invasive aquatic monocot of Eurasian origin, first observed in North America in the Saint Lawrence River in the late 1800s. The earliest U.S. populations were reported from River Rouge, MI in 1918 (Anderson et al. 1974) and subsequently throughout the Great Lakes region (Witmer 1964). Infestations now persist across the northern tier of the U.S., although evidence for multiple introductions from separate source areas exists. Plants in the Northwestern U.S. are mostly triploid, whereas populations in the northeast are more commonly diploid (Lui et al. 2005). Diploid plants are generally quite fertile and abundant seed-producers, while triploid plants are usually sterile (Lui et al. 2005). As such, the relative importance of sexual (seed) or vegetative (bulbils) propagation to spread flowering rush differs between geographic locations.

Few studies have addressed ecosystem impacts associated with flowering rush. Limited data describing economic impacts indicate that dense, monotypic growth negatively affects water delivery in irrigation systems, decreases recreational use of waterbodies (through propeller-fouling and infesting swimming areas) (Boutwell 1990; Rice and Dupuis 2009) and may increase the occurrence of swimmer's itch (cercarial dermatitis) by increasing suitable habitat for pond snails, intermediate host organisms for the swimmer's itch parasite (Parkinson et al. 2010). Additional impacts include competition with native plant species for space and nutrients and colonization of previously unvegetated habitats, which support native fish species (e.g., cutthroat and bulltrout) (Parkinson et al. 2010; Jacobs et al. 2011). The value of flowering rush to wildlife is questionable, though its use as a waterfowl food has been documented (Martin and Uhler 1939).

Currently, available management technologies include mechanical harvesting and mowing, although research into herbicide use and efficacy is ongoing. Early results suggest that application during low water levels may provide limited control (Jacobs et al. 2011). Mechanical methods of removal can be effective, but fragmentation can increase dispersal of bulbils and rhizome fragments, leading to downstream infestations (Jacobs et al. 2011).

Biological control may represent an alternative to traditional management techniques. Surveys for biological control agents in Europe are ongoing. Insect and classical pathogen agents must be host specific to meet the requirements of the Animal and Plant Health Inspection Service permitting processes (Fisher et al. 2007). Since there are no other *Butomus* species in the U.S., this particular plant is a promising candidate for biological control. Most host-specific insect agents' diets are restricted within the host genus, so the probability of identifying suitable, host-specific agents in the host range is high.

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14. ABSTRACT
Butomus umbellatus L. (Butomaceae; flowering rush) is an invasive aquatic monocot of Eurasian origin, first observed in North America in the Saint Lawrence River in the late 1800s. The earliest U.S. populations were reported from River Rouge, MI in 1918 (Anderson et al. 1974) and subsequently throughout the Great Lakes region (Witmer 1964). Infestations now persist across the northern tier of the U.S., although evidence for multiple introductions from separate source areas exists. Plants in the Northwestern U.S. are mostly triploid, whereas populations in the northeast are more commonly diploid (Lui et al. 2005). Diploid plants are generally quite fertile and abundant seed-producers, while triploid plants are usually sterile (Lui et al. 2005). As such, the relative importance of sexual (seed) or vegetative (bulbils) propagation to spread flowering rush differs between geographic locations.

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Prior to initiating a weed biological control program, it is important to conduct surveys in the introduced range of the invasive species to obtain baseline information on associated herbivore fauna and pathogenic diseases. The results of these surveys allow researchers to predict potential interactions between native and introduced agents that could alter management efficacy. To date, no known data have been published describing insect herbivores of flowering rush in the U.S. Additionally, only one pathogen (*Physoderma butomi* Schroeter; Sparrow 1956) has been documented on flowering rush (Farr et al. 1989) in the United States. Sparrow (1956) reported *P. butomi* occurring on flowering stalks and perianths of flowering rush in Michigan.

An initial survey was completed in the Pacific Northwest portion of the flowering rush range to document baseline herbivory and disease on flowering rush in the U.S. To cover the entire U.S. range, out-year efforts should concentrate on infestations in the Upper Midwest and Northeast.

MATERIALS AND METHODS: Nine flowering rush infestations were examined within Washington, Idaho, and Montana (Figure 1) during July 2014. These sites represent the geographic range of northwestern U.S. flowering rush infestations as of 2010.

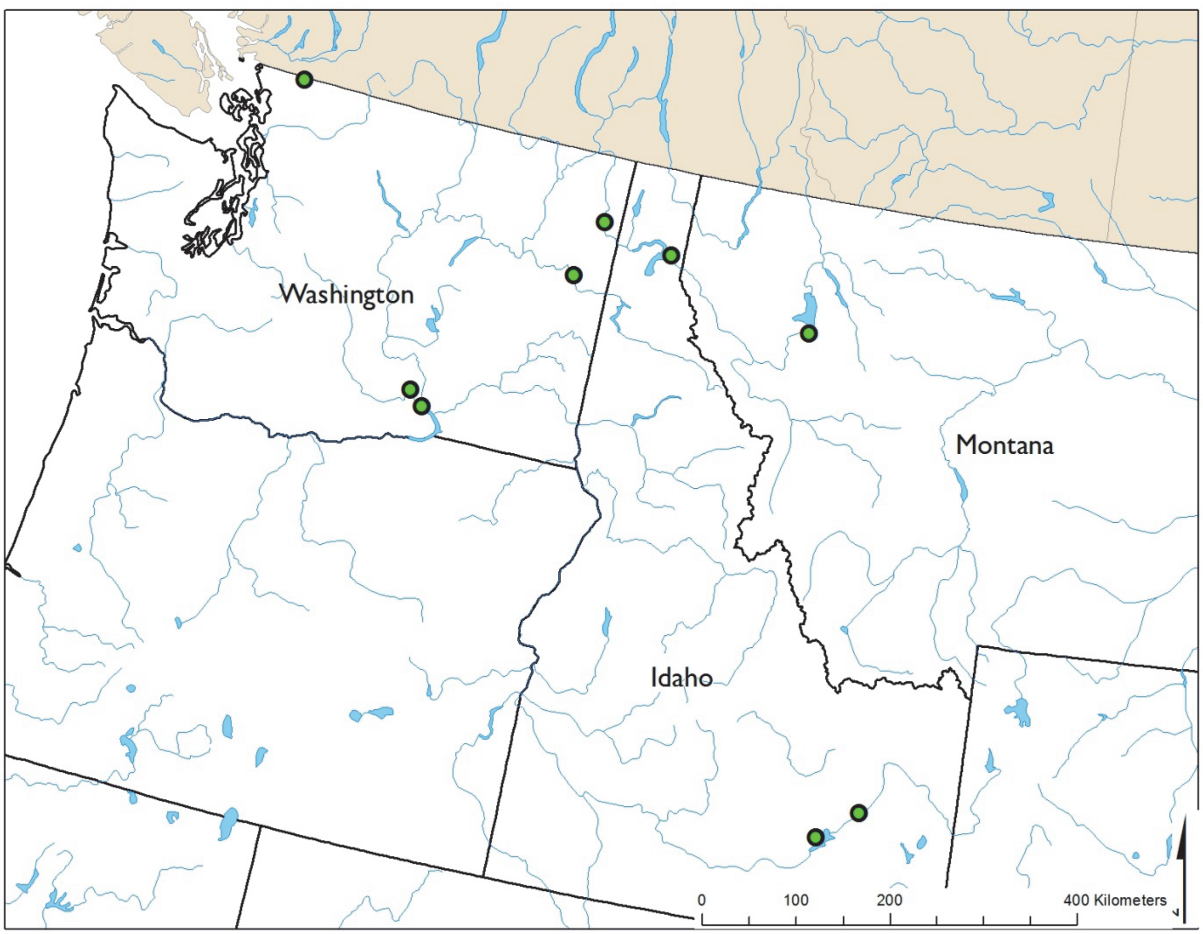


Figure 1. Flowering rush survey sites in 2014.

At each site, plants were visually examined for herbivory and disease symptoms for approximately five minutes. Potential invertebrate herbivores were collected and preserved in 70% ethanol for subsequent identification. Plants suspected to be diseased were collected and diseased parts were excised and shipped overnight to the U.S. Army Engineer Research and Development Center (ERDC), Vicksburg, MS for processing and isolation/identification of pathogens.

Twenty whole flowering rush plants — roots included — per site were excavated for microscopic examination and damage assessment. Plants were collected by hand from a boat or by wading in shallow water. In soft substrate, plants pulled easily; in some cases plants, were dug by hand or with a garden trowel. Though flowering rush spreads vegetatively and produces extensive underground rhizome systems, for the purposes of this study, individual plants are considered to be those that pulled free from the sediment independently of others (even though they may have shared root stock). Maximum height of plant, plant growth status (emergent/submersed), presence of flowers, number of leaves per plant, signs of herbivory (chewing/cutting, internal mining, etc.), and any insects collected during examination were recorded and preserved for identification. Additionally, site characteristics were measured or estimated, including infestation extent, water depth, and Secchi depth. Whole plants were examined microscopically, and signs of damage were categorized into cutting/chewing, mining, scraping, clipping/grubbing, and disease (Table 1).

Pathogen examination

Although the primary objective of this study was to identify occurrence of damage (herbivore or disease), suspected diseased plants were examined from several sites to verify pathogen presence. Sections of tissue that appeared to be diseased were excised and surface-sterilized in 10% Chlorox for one minute then rinsed in sterile water. The pieces were subsequently inserted into slits cut into Martin's Agar (Martin 1950) plates and incubated in the dark at room temperature (20-22 C^{o}) for approximately one week. Fungal isolates that emerged from the diseased tissue were transferred to Potato Dextrose Agar (PDA) and Corn Meal Agar (Difco, Detroit MI) slants for preservation. They were also plated onto PDA and Potato Carrot Agar (Dhingra and Sinclair 1995) for identification purposes.

RESULTS AND DISCUSSION: Nine flowering rush populations in Washington, Idaho, and Montana were examined for herbivory and disease (Table 2). Site types varied but included lakes, ponds, rivers, and small creeks and ranged in infestation extent from very low-density, scattered plants to high-density, expansive (>100 hectares). Flowering plants were observed, however, mostly in low numbers — at six of nine sites. The lack of flowering plants is not surprising because most northwestern populations are thought to be triploid, and triploid flowering rush plants rarely flower or put energy into sexual reproductive structures (Lui et al. 2005). Water depth at sample sites ranged from 0 m (moist sediment) to approximately 1.5 m, and water clarity was moderate to high (Secchi visible to bottom) at all but one site (Rose Pond, ID).

Table 1. Damage categories used during flowering rush survey.

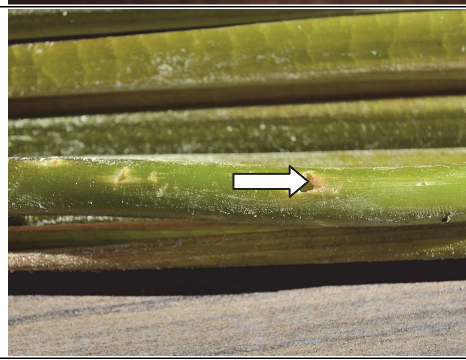
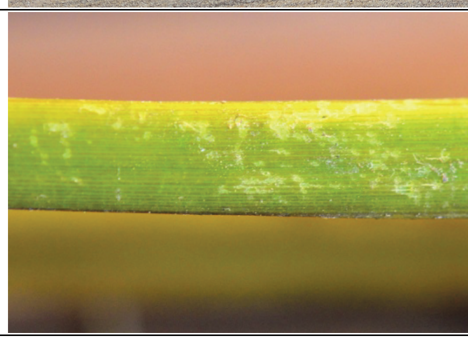
Damage Category	Example	Description
Cutting/Chewing		Removed leaf material, often on leaf margins
Mining		Discoloration of plant epidermal tissue caused by internal feeding, or observable tunneling
Scraping		Removal of epidermal plant tissue with no/limited internal tissue removed
Clipping/Grubbing		Damage that appeared to be caused by vertebrate herbivores, in which plants were uprooted or the upper portion was removed or broken free from the base
Disease		Leaf lesions, discoloration of leaves, or distinct spots

Table 2. Descriptive information for flowering rush herbivory and pathogen survey sites.

Site	Waterbody Type	County	State	Latitude	Longitude	Infestation	Other species present	Depth (m)	Secchi depth (m)	Flowering?	Comments[1]
Silver Lake	Lake	Whatcom	WA	48° 58.033	122° 04.183	Moderate density	*Potamogeton amplifolius* Tuck. *Potamogeton praelongus* Wulfen *Nitella* sp. *Chara* sp.	0.5-2	6.5	None	First observed in 1997, already well established. Treated with diquat previous 2 years.
Yakima River	Flowing River	Benton	WA	46° 22.764'	119° 25.844'	Low density, Scattered	*Heteranthera dubia* (Jacq.) MacMill. *Ceratophyllum demersum* L. *Potamogeton nodosus* Poir. *Potamogeton crispus* L. *Myriophyllum spicatum* L. *Elodea* sp.	0-1	> 3	Yes, few	WA DOE first observed flowering rush in 2008, but likely present since at least 2003.
Columbia River	Flowing River	Benton	WA	46° 14.782'	119° 13.391'	Low density, Scattered	*Hetheranthera dubia* (Jacq.) MacMill. *Potamogeton crispus* L. *Myriophyllum spicatum* L. *Typha* sp. *Zannichellia palustris* L. *Potamogeton* sp.	1.5	>2	None	First surveyed by WA DOE in 2010. All plants submersed.
Lake Spokane	Lake	Spokane	WA	47° 48.070'	117° 33.310'	Low density, Scattered	*Stuckenia pectinata* (L.) Börner *Potamogeton Richardsonii* (Benn.) Rydb. *Nymphoides peltata* (S.G. Gmel.) Kuntze *Schoenoplectus* sp.	0-2	>4.1	Yes, few	First observed in 2010.
Pend Oreille River	Flowing River	Pend Oreille	WA	48° 21.706'	117° 17.078'	Scattered patches	*Sagittaria* sp. *Ranunculus* sp. *Potamogeton illinoensis* Morong *Zosteriformis* sp.	1.2	>1.2	None	First observed in 2010, population source is Flathead Lake, MT.

[1] Historical information from J. Parsons (Washington Department of Ecology), B. Bluemer (Bonner County Weed Superintendent), V. Dupuis (Salish Kootenai College)

Table 2. Descriptive information for flowering rush herbivory and pathogen survey sites.

Site	Waterbody Type	County	State	Latitude	Longitude	Infestation	Other species present	Depth (m)	Secchi depth (m)	Flowering?	Comments[1]
Lake Pend Oreille	Lake	Bonner	ID	48° 10.793'	116° 14.037'	Moderate density, >100 acres	*Ranunculus* sp.	1	>1	Yes, few	Site in driftwood yard. Infestation first documented in 2007.
Flathead Lake	Lake	Lake	MT	47° 41.793'	114° 04.231'	High density, expansive	*Utricularia* sp. *Typha* sp. *Lemna* sp. *Ricciocarpus* sp. *Potamogeton* sp.	1	>3	Yes, many	Old infestation, at least 30 years.
Aberdeen Canal	Flowing Creek	Bingham	ID	42° 57.044'	112° 49.742'	Moderate density, expansive	*Typha* sp. *Sagittaria* sp. *Elodea* sp.	1	>1	Yes, many	Approximately 15% plants were flowering.
Rose Pond	Static Pond	Bingham	ID	43° 14.822'	112° 18.927'	High density, expansive	*Typha* sp.	0.5	<0.5	Yes, few	Approximately 1% plants flowering.

Apparent Herbivory

Plants at all sites displayed at least one type of herbivore damage. Despite this, no flowering rush populations displayed extensive damage, such that further examination was warranted. The exception to this was damage from clipping/grubbing. At several sites (e.g., Yakima River), small stands of plants were nearly destroyed by apparent feeding of vertebrate herbivores. Herbivory by vertebrates, such as waterfowl, may increase fragmentation and spread flowering rush. The implications of this are important because it is believed that seed production rarely occurs in triploid populations and clonal spread is the primary reproductive mechanism (Rice and Dupuis 2009; Lui et al. 2005).

Overall, a high percentage of plants (112 out of 180 plants examined, 62%; Figure 2) displayed damage that is probably attributable to invertebrate herbivory. In addition, 29% (52) of plants displayed one category of damage; 26% (46) displayed two; and 8% (14) of plants displayed three damage categories.

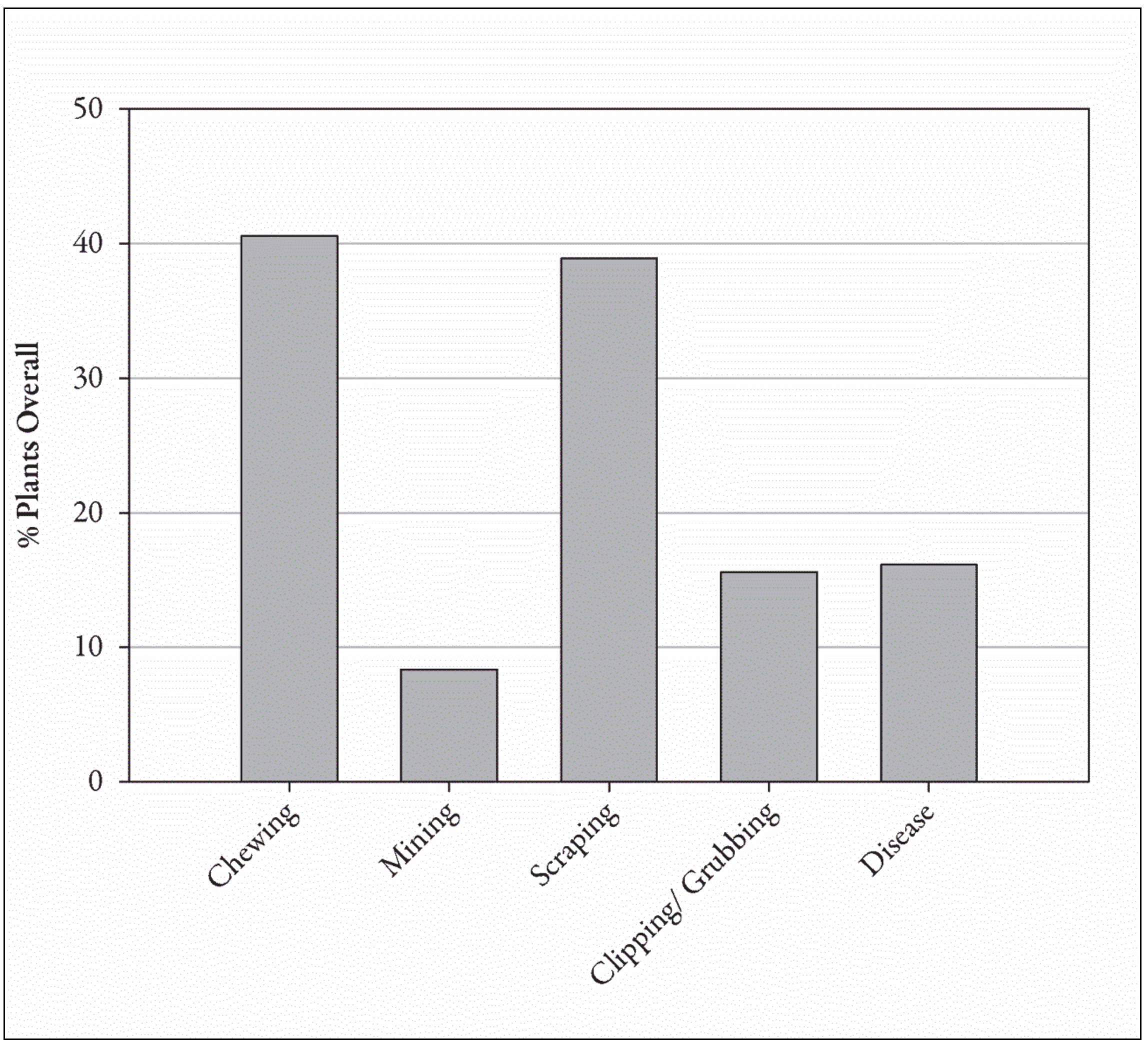


Figure 2. Percentage of plants from all sites exhibiting various types of herbivore damage.

Cutting/chewing damage was observed on plants at every site; 40% of plants overall displayed cutting/chewing damage. This damage was mainly observed on submersed leaves and appeared to be the result of generalist caddisfly or caterpillar herbivores. Various caddisfly and caterpillar genera are known to shred vascular leaf material for food and case-building materials (Merrit et al. 2008; Wiggins 2004). Larvae of *Ylodes* sp., *Ceraclea* sp., and *Oecetis* sp. were recovered within their cases on flowering rush plants during this survey. One unidentifiable early-instar caterpillar was collected from Rose Pond, ID. There are several species of aquatic/subaquatic caterpillars found in the Northwest, including several with extreme generalist diets (e.g., *Synclita* spp.). Although each of these taxa contains herbivorous members, the extent of damage caused by these specific taxa was indeterminable.

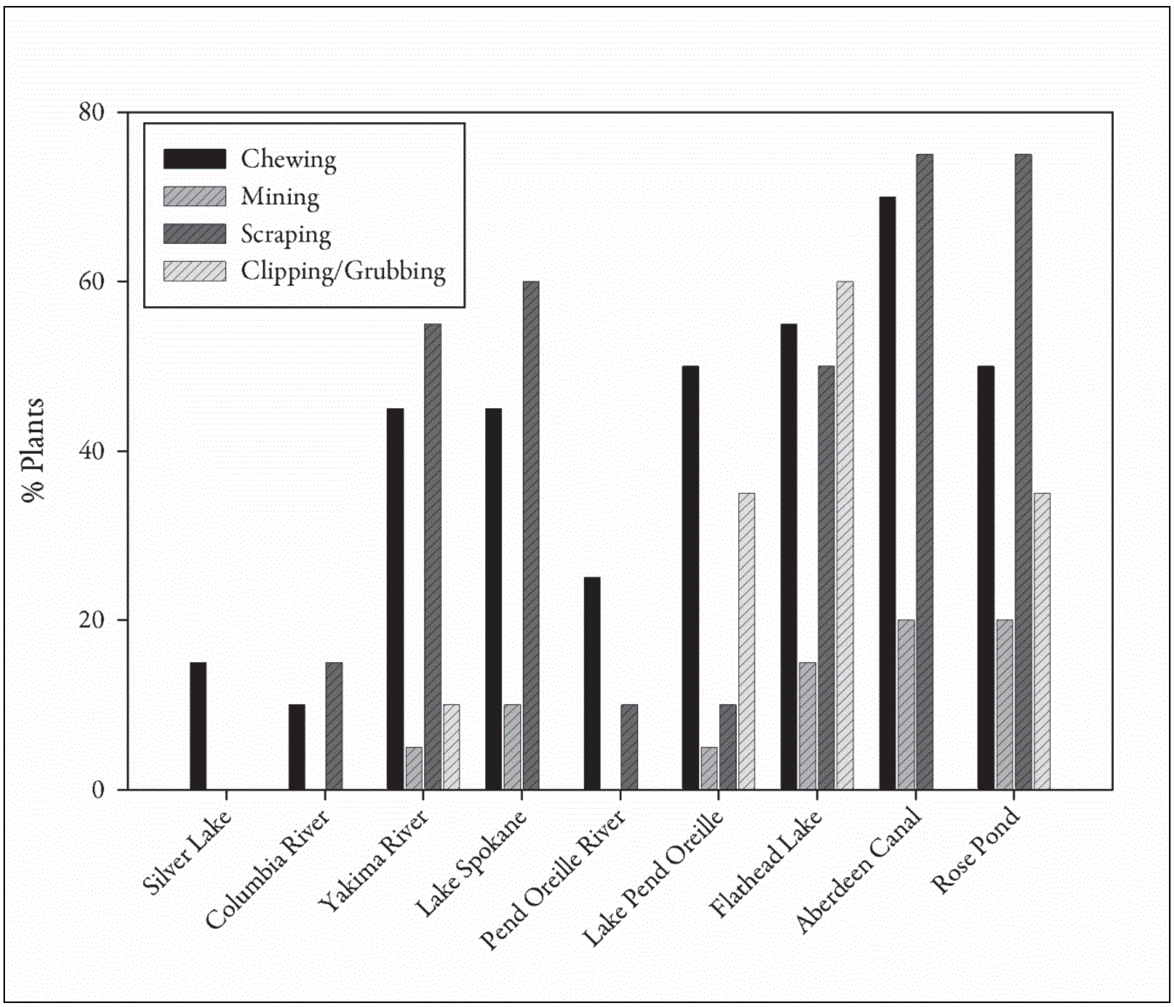


Figure 3. The percentage of plants at sites displaying herbivore damage.

Mining damage was uncommon on plants during this survey, with 8% of plants displaying this type of damage. Mines were probably produced by chironomids. Some species of chironomids are known to mine plant tissues for retreat-building; others cause sufficient damage to be considered as biocontrol agents (e.g., *Cricotopus lebetis* Sublette as an agent of *Hydrilla verticillata* L.f. Royle; Cuda et al. 2011).

Scraping damage was observed at 89% of sites (39% of plants overall) but was most abundant at Rose Pond and Aberdeen Canal in eastern Idaho. Seventy-five percent of sampled plants at these two sites were damaged by scraping, although no substantial impacts were noticeable.

Clipped/grubbed plants were observed at four sites (Yakima River, Lake Pend Oreille, Flathead Lake, Rose Pond) but were most abundant at Flathead Lake, MT, with 60% of plants displaying signs of grubbing. Clipping/grubbing appears to be the result of vertebrate feeding; flowering rush has been reported as a valuable duck food (Martin and Uhler 1939); and heavy waterfowl grazing has been observed previously (B. Bluemer[1], personal communication). In fact, the uprooting feeding style of some waterfowl may be partially responsible for the spread of propagules downstream of infestations.

Invertebrate Taxa Observed

The goal of this study was to assess occurrence of herbivore damage on flowering rush, not to construct a comprehensive list of invertebrate herbivores using flowering rush as a food resource. However, several potentially herbivorous taxa were observed during plant examination, including *Lymnaea stagnalis* L. and *Physa* sp., *Chironomus* spp., *Triaenodes* sp., *Ylodes* sp., *Oecetis* sp., *Ceraclea* sp., and an immature aquatic caterpillar (possibly *Synclita* sp.).

Pathogens

Sixteen percent of all plants displayed disease symptoms consisting of leaf discoloration, lesions, or spots (Figure 2). Ten fungal isolates were obtained from tissues. Of these, five could not be identified because they did not sporulate. Four were moniliaceous (hyaline hyphae) Ascomycetes, and one was a dematiaceous (dark hyphae) Ascomycete. The remaining isolates were determined to be *Pestalotiopsis guepinii* (Desm.) Steyaert, *Virgaria nigra* (Link) Nees, *Hansfordia ovalispora* S. Hughes, *Fusarium oxysporum* Schlecht., and a *Phoma* sp. *Pestalotiopsis guepinii* has been reported to cause a dieback on pistachio (Gore et al. 2010) and a twig blight on hazelnut and walnut in Turkey (Karaca and Erper 2001). It also invades rhododendrons, producing zonate gray-brown leaf spots (Barr 1975). *Virgaria nigra* occurs on the wood and bark of various trees in tropical and subtropical regions (Ellis 1971). *Hansfordia ovalispora* was isolated from Fraserfir during an inventory in forests of the Great Smoky Mountains National Park in 2004 (Baird et al. 2007). Both *F. oxysporum* and *Phoma* have broad host ranges, which may limit their desirability as biocontrol agents.

All the fungal isolates from the current study represent new host associations. Their efficacy as pathogens for *Butomus* is dubious, considering that the majority of them have been tested on other plant species (hydrilla and Eurasian watermilfoil) with little success (Shearer, personal observation). Species of *Fusarium* and *Phoma* have been researched as mycoherbicides: *F. equiseti* (Corda) Sacc. for management of barnyard grass (Motlagh 2011) and *P. exigua* Desm. for management of yellow starthistle (Laurent et al. 2004). Within *F. oxysporum,* many *formae speciales* (an informal taxonomic grouping allowed by the International Code of Nomenclature for algae, fungi, and plants that is applied to a parasite that is adapted to a specific host) are recognized. Before a *forma speciales* could be applied to flowering rush, Koch's postulates would have to be executed to prove *F. oxysporum* was

[1] Bluemer, B. 2014. Personal communication with Nathan E. Harms. Bonner County Weed Control, Sandpoint, ID.

a disease-causing organism and could be reisolated from tissues that were infected (Agrios 2005). The same would be true for *Phoma*. Additionally, species identification is needed for the *Phoma* sp.

CONCLUSION: Herbivory and pathogens were identified on flowering rush in the Pacific Northwest, but the levels of damage were nearly always minimal; neither herbivory or disease appeared to be sufficient to provide population-level impacts in the Pacific Northwest. The lack of natural enemies in introduced areas is often cited as a chief reason why species become invasive and provides the basis for searching in the native range of the target species. The lack of damaging enemies in the U.S. provides additional justification for continued overseas exploration and importation of biological control agents of flowering rush. Additionally, although signs of herbivory/ disease were common, the extent of each was minimal, so there does not appear to be any likelihood of negative interaction with native herbivores or diseases, should agents be imported and released in the U.S. in the future.

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