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Puccinia areolata, P. treleasiana, and P. gemella on Marshmarigold (*Caltha* spp.) in subalpine habitats in Northwestern United States

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Abstract: Marshmarigolds (*Caltha* spp.) are members of the buttercup family (Ranunculaceae) and inhabit the Cascade and Rocky Mountains in western North America. Several rust species infect *Caltha* species, but only *Puccinia areolata, Puccinia treleasiana*, and *Puccinia gemella* have been reported in the Pacific Northwest. Observations were made on rust incidence and severity on *Caltha* spp. in July through September between 2001 and 2013 in subalpine regions of Colorado, Idaho, Montana, Utah, Washington and Wyoming. Leaves, petioles and flower stalks with rust pustules were examined and the host species identified using morphological characteristics. Plants collected in ID, WA, and MT were identified as *Caltha leptosepala* ssp. *howelli* (syn *Caltha biflora*) and plants in WY, CO, and UT were identified as *Caltha leptosepala* ssp. *leptosepala*. Foci of infection were typically solitary leaves, individual plants or small clusters of infected plants although a 90% incidence of infected rust plants was observed in a 10 m diameter area with no visually distinct foci in WA in 2013. *Puccinia areolata* was the only rust observed in

Washington, *Puccinia gemella* was observed in Idaho, and *Puccinia treleasiana* was observed in Montana and Wyoming. Collections made possible direct comparisons among the three rust species.

Key words: *Caltha, Puccinia areolata, Puccinia treleasiana, Puccinia gemella*, Uredinales, subalpine ecology, diseases of native plants

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Introduction: Marshmarigolds (*Caltha* spp.) are members of the buttercup family (Ranunculaceae) and inhabit wet subalpine meadows and stream banks of the Cascade and Rocky Mountains in western North America. Observations reported herein pertain to multiple occurrences of a rust, *Puccinia spp*. Arthur & H.S. Jackson observed on *Caltha* spp. in these habitats in western United States. Three rust species were observed at varying intensities at five locations in four states from 2001 to 2013 on *Caltha leptosepala* ssp. *howelli* (Huth) P.G. Sm. (syn. *Caltha biflora* DC.) and *Caltha leptosepala* ssp. *leptosepala* DC. A history of species concepts in the *Caltha leptosepala* complex was provided by Morris (1972) and complete synonymies reflecting current taxonomic opinion are available on line (Anonymous 2008, Weinmann et al. 2007).

Several rusts occur on *Caltha* species, but only three rust species have been reported in the Pacific Northwest (Ginns 1986, Hotson 1925, Parmelee et al. 1984). *Puccinia areolata* Dietel & Holw. was described in 1894 (Dietel 1894). It is widely distributed geographically, and has been reported from the Pacific Northwest region of USA, Colorado, and Canada (Cash 1953, Anonymous 1960, Parmalee and Carteret 1984, Shaw 1973). At least five different species of *Caltha* are susceptible to infection by *P. areolata* (Anonymous 1960, Cash 1953, Conners 1967, Ginns 1986). Pycnia, aecia, telia were described by Dietel (1894) and Hotson (1925). Dietel's description did not include observations of uredinia; however they were described as cinnamon brown and hypophyllous by Arthur (1934). Teliospores are reddish brown, two-celled, of variable shape but mostly ellipsoidal. Walls on the distal cell are minutely verruculose but this feature is difficult to detect with light microscopy even under high magnifications (ca. 1000X). Walls are smooth on the base of the basal cell (Parmelee and Carteret 1984). Teliospores are 50 - 80 x 21 - 34 µm, not including the stipe (Dietel 1894).

Puccinia treleasiana (Pazschke) Arthur & H.S. Jacks. was described by Rabenhorst (1892) and infects at least three species of *Caltha* (Anonymous 1960, Shaw 1973). The geographic distribution of this rust is limited to North America where it has been reported in the Pacific Northwest and the Mountain states of the USA. Only teliospores have been observed. Teliospores have been described by Hotson (1925) as light brown, two-celled ellipsoidal, barely constricted at the septum, or not at all. Walls are slightly rugose, which is difficult to observe and teliospores are 32 - 43 x 21 - 26 (-28) µm. However, an earlier description of *P. treleasiana* indicates that teliospores are longer and narrower than Hotson's description. Arthur and Jackson (1922) describes the teliospores of *P. treleasiana* as chestnut brown. They are fusiform to fusiform-oblong and are 40 - 50 x 20 - 26 µm.

Puccinia gemella Dietel & Holw. was described in 1903 (Sydow et al. 1903) and is known to occur on at least three *Caltha* spp. in Alaska, British Columbia, California, Idaho, Oregon, Montana, and Washington (Blasdale 1919, Cash 1953, French 1989, Hotson 1925, Parmelee and Carteret 1984, Shaw 1973). This fungus also forms only teliospores. Teliospores are yellow-brown to brown, two-celled, ellipsoidal, and

slightly or not at all constricted at the septum. Walls are smooth and teliospores are (37-) 42-56 x 19-23 µm.

Puccinia gemella can be distinguished from *P. areolata* on the basis of teliospore size; those of *P. areolata* are much larger than of *P. gemella*. Hotson (1925) reported *P. gemella* telia to produce a conspicuously ruptured host epidermis, which was not observed in telia of *P. areolata*. Based on the Arthur and Jackson description of *P. treleasiana*, distinguishing between *P. treleasiana* and *P. gemella* is difficult due to the overlap in spore size. Two features that can be used to differentiate between the two species is the size of the umbo, which are 5 - 10 µm and 3 - 6 µm for *P. treleasiana* and *P. gemella*, respectively, and spore shape (Arthur and Jackson 1922).

Information on prevalence of rust infections on *Caltha* spp. is not available from those regions. This paper documents levels of rust incidence and severity on *Caltha* spp. in locations of Idaho, Montana, Washington and Wyoming. Such information can be useful in understanding disease development and patterns in natural habitats and in developing disease management strategies and tactics for agro-ecosystems (Browning 1974).

Materials and Methods: Observations were made on rust disease incidence and conditional severity, defined as mean area of infected leaves and petioles with pustules and associated chlorosis, on *Caltha* spp. in July through September in subalpine regions of Colorado, Idaho, Montana, Utah, Washington and Wyoming in 1993-2013 (Table 1, Figure 1). Subalpine meadows and stream banks investigated were near Timberline Lake in the Rocky National Park in Colorado, Moores Guard Station west of the Gospel Hump National Wilderness in Idaho, Highline Lake in Gallatin National Forest in Montana, Broadhead Meadows near the Uintas Mountains in Utah, Berkeley Park north of Mount Rainier in Washington, and Granite Basin of the Teton Range and Texas Basin in the Wind River Range in Wyoming. *Caltha* populations were observed over a range in elevation of approximately 400 m at each location, including plants ranging from pre- to post flowering. More than 5,000 *Caltha* sp. plants were examined at each location, except at Timberline Lake in Colorado where only 1,000 plants were examined.

Disease incidence and severity estimations were made by slowly walking through *Caltha* populations and counting infected plants. Plants with infected leaves, petioles or flower stalks were counted and the surface areas of plant parts with pustules and associated chlorosis were visually estimated. Total numbers of plants observed were estimated from counts of observed plants in subsections of a population and then estimating the number of plants observed in the population. Values were recorded. Samples of leaves, petioles and flower stalks with rust pustules were collected and transported to the laboratory at Washington State University, Pullman, WA.

Plants were examined with stereoscopic and compound microscopes and identified to species using morphological characteristics. Rust pustules and spores were examined with stereoscopic and compound microscopes to identify rust spore states and to observe and measure spores. Spores were gently scraped from a telium on an infected leaf and mounted in water. Height and width of five spores were recorded. A total of five leaves per collection were examined.

Collections examined: USA: Johnson, D. A., 8/9/12, Gospel Hump, Idaho County, ID on *Caltha leptosepala* ssp. *howelli* (WSP 72707). Johnson, D. A., 9/4/13, Gospel Hump, Idaho County, ID on *Caltha leptosepala* ssp. *howelli* (WSP 72708). Johnson, D. A., 8/2/01, Hyalite Lake in Bozeman, Gallatin County, MT on *Caltha*

leptosepala ssp. *howelli* (WSP 72711). Johnson, D. A., 9/13/12, Berkeley Park north of Mt. Rainier, Pierce County, WA on *Caltha leptosepala* ssp. *howelli* (WSP 72706). Johnson, D. A., 9/20/13, Berkeley Park north of Mt. Rainier, Pierce County, WA on *Caltha leptosepala* ssp. *howelli* (WSP 72705). Johnson, D. A., 7/27/07, Granite Basin, Teton County, WY, on *Caltha leptosepala* ssp. *leptosepala* (WSP 72709), Johnson, D. A., 8/05/10, Granite Basin, Teton County, WY, on *Caltha leptosepala* ssp. *leptosepala* (WSP 72710).

Results: Populations of *Caltha* sp. were spatially discontinuous and intermixed in plant communities with many different plant species. Populations differed in degree of plant density within locations. Growth stage of *Caltha* sp. ranged from post-flowering at the lower elevations to flowering to pre-flowering at higher elevations in July and mid- August. Plants generally were past flowering in late August and September.

Plants collected at Gospel Hump Wilderness, ID; Berkeley Park, WA; and Hyalite Lake, MT were identified as *Caltha leptosepala* spp. *howelli* (syn *Caltha biflora*) (Hitchcock et al. 1973, Welsh et al. 1965). Plants collected at Granite Basin, WY; Timberline Lake, CO; Texas Lake, WY; and Broadhead Meadows UT were identified as *Caltha leptosepala* spp. *leptosepala* (*Caltha leptosepala*) (Hitchcock and Cronquist 1973, Welsh, Treshow, and Moore 1965).

Measurements of teliospores from Washington were 20 - 27.5 (32.5) x (45) 47.5 - 62.5 (65) µm, consistent with the description of *P. areolata* (Hotson 1925) (Figure 1). Measurements of teliospores from Idaho, Montana, and Wyoming were 20 - 30 x (32) 35 - 50 µm (Table 2), consistent with either *P. gemella* (Hotson 1925) or *P. treleasiana* (Arthur and Jackson 1922) teliospore characteristics. Teliospores collected from Hyalite Lake, MT and Granite Basin, WY were ellipsoid (Figure 3 and 4) and umbo size was 2.5 - 4.55 - 6.25 µm. The species was determined as *P. treleasiana*. Teliospores collected from Gospel Hump, Idaho were fusiform to fusiform- obtuse (Figure 5) and the umbo size was (2.5) 3.75 -5.45 - 8.75 (10) µm (Figure 6). The species was determined as *P. gemella*. A ruptured epidermis is associated with telia of *P. gemella* (Hotson 1925). In the present study, a conspicuously ruptured epidermis was associated with telia of *P. treleasiana, P. gemella*, and *P. areolata* (Figures 7 - 9).

Puccinia gemella was observed on *Caltha* sp. only in Idaho (Table 1, Table 2). Rust pustules occurred on leaves, petioles and flower stalks, and mostly were observed on plants that had flowered. *Puccinia treleasiana* was the only rust observed in Montana and in two distant locations in Wyoming, the Teton and Wind River Mountain Ranges (Table 2). It was only observed on plants at flowering near Texas Lake of the Wind River Range in Wyoming in 2011. *Puccinia areolata* was the only rust observed at Berkeley Park in Washington.

Incidence of infected plants was low, at < 0.01% and mean severity of infected leaves, petioles and stems (conditional severity, defined as mean area of infected leaves and petioles with pustules and associated chlorosis) was low to moderate with estimates of a trace (less than 3 pustules per leaf) to 20% of the surface area affected at most locations (Table 1). Occasionally, a plant petiole or stem was observed with > 80% severity. However, incidence of infected plants ranged from 10 to 90% and mean conditional severity ranged from a trace to 40% in various sections of the population at Berkley Park, WA in 2013 (Table 1).

Foci of infection were solitary leaves, plants or small clusters of infected plants. Diameters of small clusters of infected plants were usually less than 0.3 m (Table 1). However, an incidence of infected rust plants of 90% was observed in an

area of approximately 10m in diameter with no visually distinct foci at Berkeley Park, WA in 2013.

Discussion: Subalpine habitats are valued natural ecosystems for aesthetics, recreation, watershed, and wildlife. Plant communities contribute to the stability of subalpine ecosystems but can be stressed by relatively short growing seasons, plant diseases, pests, human activities and other factors. Little is known about diseases affecting herbaceous plants in these habitats but their impacts could be important, particularly if exotic plant pathogens become established in fragile natural ecosystems. Another reason for studying plant diseases in these situations is that information concerning diseases in natural habitats may provide insight into developing disease management strategies for agricultural ecosystems.

Of particular interest in studying plant disease ecology is that host density influences disease characteristics both directly and indirectly (Burdon et al. 1982). Direct effects include the decreased interception of airborne spores when distances between hosts increase. On the other hand, low host density can result in increased light and decreased humidity, which may interfere with spore germination (Burdon and Chilvers 1982). Disease severities were low to moderate and disease patterns were aggregated.

Host plants occurred in sparse clusters, with clusters intermixed with non-host plant species. This pattern of distribution may account for the low disease incidence observed in all locations and all years other than at Berkeley Park in 2013. The highly clustered yet diffuse distribution pattern of the host may have favored pathogen spread to small foci. The high disease incidence observed in 2013 at Berkeley Park could have resulted from teliospores accumulating over several seasons, and/or from highly favorable conditions for host development and infection. Fungal diseases are positively density-dependent (Burdon and Chilvers 1982) and host distribution patterns may significantly affect disease incidence and severity, although the effects of clumping have not been well studied (Burdon and Chilvers 1982). Based on host distribution, incidence, severity, and pattern of rust infected plants observed were as expected in natural plant communities.

Plant collections from four states made possible the comparison of teliospore morphology of *P. areolata*, *P.gemella* and *P. treleasiana* by individuals in the same laboratory. Only telia and teliospores were observed on plant material observed in the current study. The inconsistencies in morphological descriptions of fungal species can lead to the misidentification of pathogens. Based on Hotson's description of teliospores, fungi recovered from Hyalite Lake, MT and Granite Basin, WY were initially designated as *P. gemella*. The occurrence of *P. gemella* in WY would have been a first report. However, based on Arthur and Jackson's descriptions of *Puccinia* species on *Caltha*, spore morphology was more in line with *P. treleasiana*, which has previously been described in Wyoming. Acquiring sequence data may help to eventually distinguish among these *Puccinia* species.

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Literature cited:

Anonymous. 1960. Index of plant diseases in the United States. USDA Agriculture Handbook. 165:1-531.

Arthur, J. C. 1934. Manual of the rusts in United States and Canada. Lafayette, Ind.: Purdue Research Foundation.

Arthur, J. C., and H. S. Jackson. 1922. North American Flora. New York Botanical Garden.

Blasdale, W. C. 1919. A preliminary list of the Uredinales of California. University of California Press. http://dx.doi.org/10.5962/bhl.title.26907

Browning, J. A. 1974. Relevance of knowledge about natural ecosystems to development of pest management programs for agro-ecosystems. Proceedings of the American Phytopathological Society.

Burdon, J. and G. Chilvers. 1982. Host density as a factor in plant disease ecology. Annual Review of Phytopathology 20:143-166. http://dx.doi.org/10.1146/annurev.py.20.090182.001043

Cash, E. K. 1953. A checklist of Alaskan fungi. Plant Disease Reporter 219:1-70. http://dx.doi.org/10.5962/bhl.title.85799

Conners, I. L. 1967. An annotated index of plant diseases in Canada and fungi recorded on plants in Alaska, Canada and Greenland. Research Branch Dept. Canada Dept. of Agriculture. Publication 1251.

Dietel, P. 1894. Descriptions of new species of Uredineae and Ustilagineae, with remarks on some other species II. Botanical Gazette 19:303-306. http://dx.doi.org/10.1086/327074

French, A. M. 1989. California plant disease host index. California Dept. of Food and Agriculture, Division of Plant Industry.

Ginns, J. H. 1986. Compendium of plant disease and decay fungi in Canada, 1960-1980. Canadian Government Publishing Centre. http://dx.doi.org/10.5962/bhl.title.58888

Hitchcock, C. and A. Cronquist. 1973. Flora of the Pacific Northwest. University of Washington Press, Seattle.

Hotson, J. W. 1925. Preliminary list of the Uredinales of Washington. Preliminary list of the Uredinales of Washington. 273-391.

ITIS report. 2008. *Caltha leptosepala* ssp. *howellii*. Online. Taxonomic Serial No. 523727. Integrated Taxonomic System Information, Smithsonian Institution, Washington, DC.

Morris, M. I. 1972. A biosystematic analysis of the Caltha leptosepala (Ranunculaceae) complex in the Rocky Mountains I. Chromatography and Cytotaxonomy. Brittonia 24:177-188. http://dx.doi.org/10.2307/2805868

Parmelee, J. A. and P. M. D. Carteret. 1984. *Puccinia gemella*. Fungi Canadenses 264:1-2.

Rabenhorst, Winter, and Pazschke. 1892. *Fungi europaei exsiccati*. Hedwigia 3821.

Shaw, C. G. 1973. Host fungus index for the Pacific Northwest. I. Hosts. Bulletin, Washington Agricultural Experimental Station.

Sydow, and Sydow. 1903. Monograph of Urediniales. Lipsiae 1:541.

Weinmann, F., and P. F. Zika. 2007. Washington Flora Checklist: A Checklist of the Vascular Plants of Washington State. University of Washington Herbarium, Burke Museum of Natural History and Culture.

Welsh, S. L., Treshow, M., and G. Moore. 1965. Common Utah Plants. Second Edition. Brigham Young University Press, Provo, Utah.

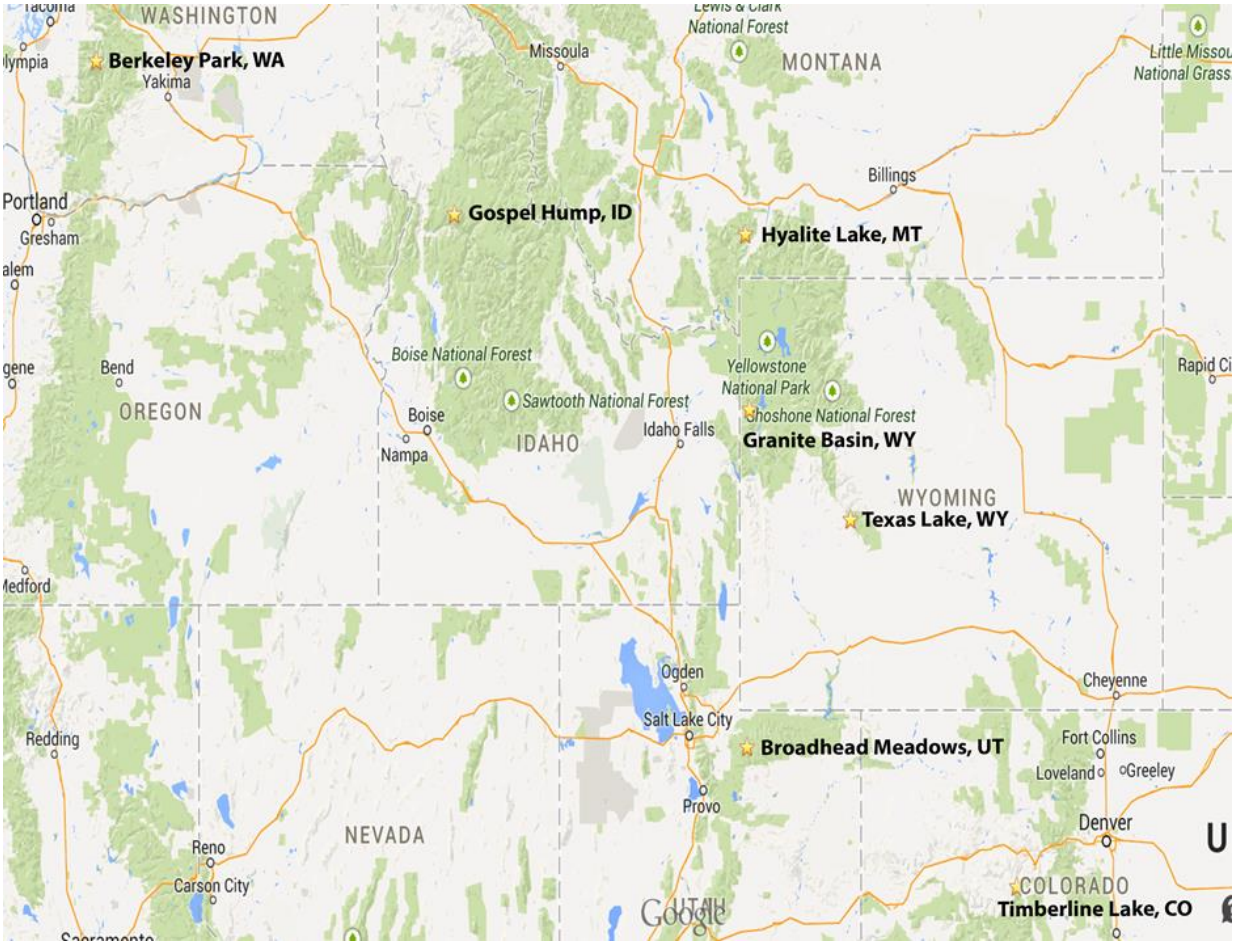
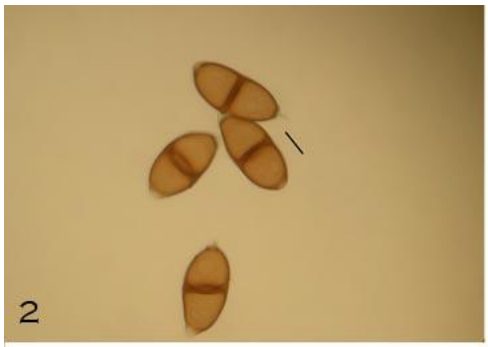


Figure 1. Study site locations in WA, MT, ID, UT, CO, and WY.

Figures 2-9 (next page). 2. Ellipsoid teliospores of *P. areolata* from Berkeley Park, WA collected in 2013. Scale = 15 µm. 3. Ellipsoid teliospores of *P. treleasiana* from Granite Basin, WY collected in 2007. Scale = 20 µm. 4. Ellipsoid teliospores of *P. treleasiana* from Hyalite Lake, MT collected in 2001. Scale = 20 µm. 5. Fusiform to fusiform obtuse teliospores of *P. gemella* from Gospel Hump, ID collected in 2012. Scale = 20 µm. 6. Fusiform to fusiform obtuse teliospore of *P. gemella* from Gospel Hump, ID collected in 2013. Arrow pointing to umbo. Scale = 10 µm. 7. Telium of *P. areolata* and ruptured epidermis of *Caltha leptosepala* ssp. *howelli* collected from Berkeley Park, WA. Scale = .05 mm. 8. Telia of *P. treleasiana* and ruptured epidermis of *Caltha leptosepala* ssp. *howelli* collected from Hyalite Lake, MT. Scale = .05 mm.9. Telium of *P. gemella* and ruptured epidermis of *Caltha leptosepala* ssp. *howelli* collected from Gospel Hump, ID. Scale = 0.2 mm.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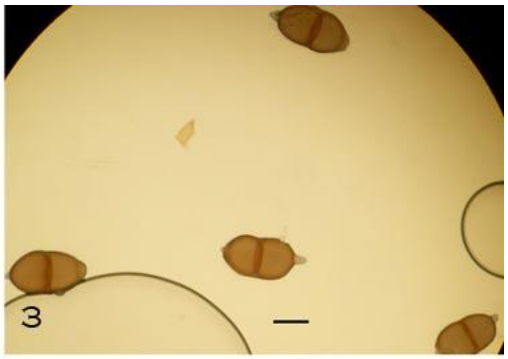
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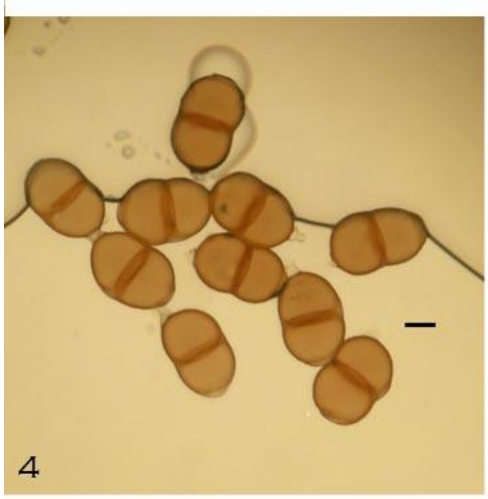
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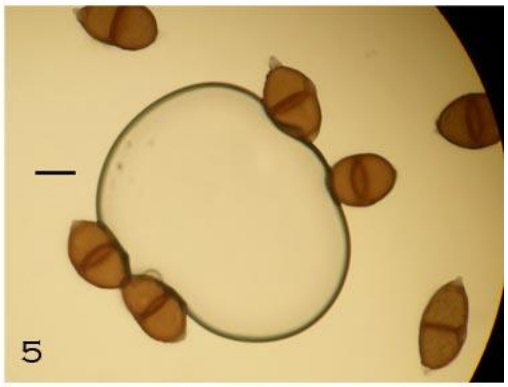
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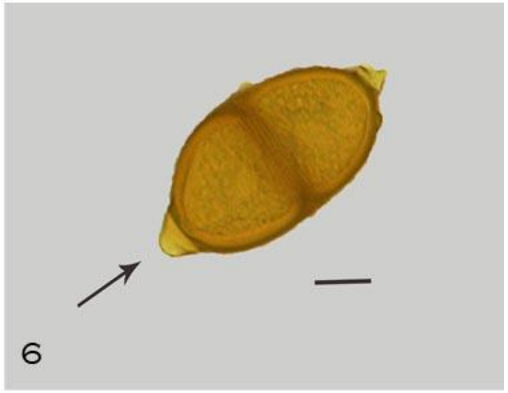
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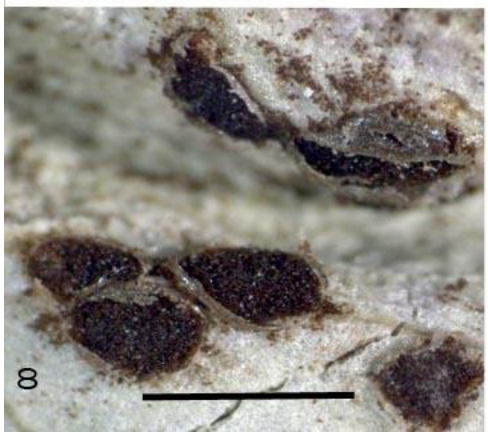
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Table 1. Location, date of observation, rust incidence and conditional severity, and pattern of *Caltha* sp. infected with *P. gemella*, *P. areolata,* and *P. treleasiana* in western North America[a].

Location[a]	Date	Host[b]	Incidence (%)	Rust Conditional Severity (%)	Pattern of infect plants, notes
Colorado					
Timberline Lake	8/22/12	Cl.l	0	0	No rust observed
Idaho					
Gospel Hump	8/9/12	Cl.h	0.0002	Trace to 5	Two infected leaves on one plant – a focus
	9/4/13	Cl.h	< 0.0001	Trace to 5	Four infected leaves on one plant – a focus
Montana					
Hyalite Lake	8/2/01	Cl.h	<0.01	Trace to 10	Solitary leaves and infected plants in small foci.
Utah					
Broadhead Meadows	8/15/12	Cl.l	0	0	No rust observed.
Washington					
Berkeley Park	9/13/12	Cl.h	<0.001	Trace to 10	Infected plants in small foci.
	9/20/13	Cl.h	10 to 90	Trace to 40	No distinct foci, general epidemic with rust infected plant distributed throughout *Caltha* populations
Wyoming					
Granite Basin	7/27/07	Cl.l	< 0.001	Trace to 10, one stem at 80	Infected plants in small foci. Foci scattered over large populations of *Caltha* sp. intermittently covering several miles.
	8/5/10	Cl.l	0.001	Trace to 20	Solitary plants to infected plants in small foci.
	7/18/12	Cl.l	<0.001	Trace to 10	Infected plants in small foci.
Texas Lake	8/25/11	Cl.l	<0.001	Trace to 20	Small foci, plants were in bloom.

[a] *Puccinia gemella* was observed in Idaho, *P. areolata* in Washington, and *P. treleasiana* in Montana and Wyoming.

[b]Cl.h = *Caltha leptosepala* spp. *howelli* (syn *Caltha biflora*), Cl.l = *Caltha leptosepala* spp. *leptosepala*

Date = month, day, year

Conditional severity = mean area of infected leaves and petioles with pustules and associated chlorosis.

Table 2. Location, date, and teliospore measurements of rusts isolated from *Caltha* spp.

		Teliospore Measurements (µm)				
Location	Collection Date	Height	Width	Umbo	Teliospore shape	*Puccinia* species
Colorado						
Timberline Lake	8/22/2012	ND	ND			
	8/9/2012	ND	ND			
Idaho						
Gospel Hump	8/9/2012	(37.5) 40 - 45 (50)	(20) 22.5 - 27.5	3.75 - 5.45 - 8.75	fusiform, fusiform obtuse	*P. gemella*
	9/4/2013	(35) 37.5 - 45 (50)	(20) 22.5 - 27.5 (30)	2.5 - 5.4 - 10	fusiform, fusiform obtuse	*P. gemella*
Montana						
Hyalite Lake	8/2/2001	(35) 40 - 42.5 (45)	(20) 24.8 - 27.5	2.5 - 3.8 - 5	ellipsoid	*P. treleasiana*
Utah						
Broadhead Meadows	8/15/2012	ND	ND			
Washington						
Berkeley Park	9/13/2012	(47.5) 50 - 60 (65)	22.5 - 27.5 (32.5)		variable	*P. areolata*
	9/20/2013	(45) 47.5 - 55 (65)	(2.5) - 4.55 - (6.25)			*P. areolata*
Wyoming						
Granite Basin	7/27/2007	(37.5) 40 - 47.5 (50)	(20) 22.5 - 25 (30)	2.5 - 4.55 - 6.25	ellipsoid	*P. treleasiana*
	8/5/2010	(32.5) 37.5 - 45 (50)	(20) 22.5 - 27.5 (30)	2.5 - 4.55 - 6.25	ellipsoid	*P. treleasiana*
	7/18/2012	ND	ND			
Wyoming						
Texas Lake	8/25/2011	ND	ND			

ND- rust was not observed