ROSE GALL WASPS (CYNIPIDAE: *DIPLOLEPIS*) OF MANITOBA, INCLUDING A NEW PROVINCIAL RECORD

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ABSTRACT

We report the first provincial record of the rose gall wasp *Diplolepis gracilis* (Ashmead) in Manitoba. In addition, we review the distribution and hosts of the 11 *Diplolepis* species previously reported for the province using museum records and citizen science observations through iNaturalist. Our study highlights the value of community science platforms such as iNaturalist for documenting records of otherwise poorly known taxa

INTRODUCTION

Diplolepis Geoffroy (Hymenoptera: Cynipidae) is a Holarctic genus of approximately 50 species, 31 of which are known from North America (Zhang et al. 2020). As with most cynipid taxa, Diplolepis is in need of revisionary taxonomic work. The lack of detailed descriptions for many species and the absence of identification keys make it challenging to identify Nearctic specimens without gall association. Fortunately, there is renewed interest in cynipids and recent (Zhang et al. 2019, 2020) and ongoing work will hopefully clarify species boundaries of the Nearctic fauna.

All species of *Diplolepis* induce galls only on roses (Rosaceae: *Rosa* L.) (Shorthouse 2010; Zhang *et al.* 2014). Most *Diplolepis* species can induce galls on several different species of roses (Shorthouse 2010; Zhang *et al.* 2014; Nastasi and Deans 2021), though comprehensive knowledge of host associations and preference is lacking. The use of ornamental roses as hosts has been reported in other parts of North America, but such occurrences appear to be rare (Shorthouse 2001, J.D. Shorthouse unpublished data).

Diplolepis species reported for Manitoba are univoltine (Shorthouse 2010). The seasonal timing of oviposition varies by species. Adults of some species lay eggs in spring while others do so later in the growing season (Shorthouse 2010). Gall development begins before eggs hatch and larvae are usually surrounded by gall tissue shortly after they begin feeding (Shorthouse 2010). Individuals over-winter in their galls as pre-pupae, and adults emerge the following year to mate (although some species are parthenogenetic) and lay eggs (Shorthouse 2010). Aside from one week as an egg and a week as an adult, a *Diplolepis* wasp spends its entire life within its gall (Shorthouse 2010).

Gall placement and morphology are usually species-specific and can often be used to determine the gall-inducing species (Shorthouse *et al.* 2005). However, the presence of inquilines and parasitoids can alter gall morphology (Shorthouse 2010), making identification of the inducer difficult in some cases. Galls induced by North American *Diplolepis* species host both inquilines (Cynipidae: *Periclistus* Foerster) and numerous parasitoids including Ichneumonidae (*Orthopelma* Taschenberg), Eulophidae (*Aprostocetus* Westwood), Eupelmidae (*Eupelmus* Dalman), Eurytomidae (*Eurytoma* Illiger, *Tenuipetiolus* Bugbee), Ormyridae (*Ormyrus* Westwood), Pteromalidae (*Pteromalus* Swederus), and Torymidae (*Glyphomerus* Foerster, *Torymus* Dalman) (Shorthouse 2010; Zhang *et al.* 2014, 2019).

Four species of wild rose are present in Manitoba: *Rosa acicularis* Lindl., *R. arkansana*, *R. blanda* Aiton, and *R. woodsii* Lindl. Identification to species can be challenging as all are phenotypically variable and can hybridize (Scoggan 1957; Flora of North America Editorial Committee 2014). *Rosa acicularis* is a woodland species that occurs throughout the province except the extreme north (Scoggan 1957). *Rosa blanda* and *R. woodsii* are likewise most often found in woodlands though their occurrence is limited to the southern half of the province (Scoggan 1957). *Rosa arkansana* is a grassland species and is limited to such habitats in the southern portion of Manitoba (Scoggan 1957).

Very little focussed survey work has been done for *Diplolepis* in Manitoba. Joseph D. Shorthouse has done significant work documenting the distribution of *Diplolepis* in Canada, but most of his work was done in Alberta, British Columbia, Ontario, Quebec, and Saskatchewan. Both taxonomic and geographic representatives of his collection are housed, in part, at the Canadian National Collection (CNCI) and the Smithsonian National Museum of Natural History (USNM), yet Manitoba material appears limited to 42 specimens of *D. spinosa* (Ashmead) from a single collecting event at Morden (A.M.R. Bennett, pers. com.). Another significant portion of his collection is at the University of Edinburgh that includes some material from Manitoba in unsorted vials (J.D. Shorthouse pers. comm.). The Manitoba Museum (MM) collection has no *Diplolepis* specimens catalogued, though it is possible (but unlikely) that such specimens are present amongst unsorted/unidentified material (R.D. Mooi, pers. comm.). The material identified to species in the J. B. Wallis / R. E. Roughley collection (WRME) consists of a long series of *D. spinosa* and a few specimens of *D. radicum* (Osten Sacken) (J. Gibbs, pers. comm.).

Thirteen native species and two introduced species of *Diplolepis* are known from Canada (Shorthouse 2010; Zhang *et al.* 2019). Shorthouse (2010) explicitly included Manitoba in the range of one species, *D. polita* (Ashmead), and implicitly included the province in the range of nine others. It is unclear which, if any, of these reports are based on actual specimens or gall observations as opposed to inferring presence in Manitoba based on the general distribution of species. Nastasi and Deans (2021) include Manitoba in the range of seven of the ten species included by Shorthouse (2010) based on that work and observations recorded on the citizen science platform iNaturalist (www.inaturalist.org). In addition to these 10 species, there is a specimen labelled as *Diplolepis tuberculator* (Cockerell) at the American Museum Natural History. Thus, there are 11 nominate species reported for Manitoba (Table 1). Morphological characteristics and recent phylogenetic work suggest *D. nebulosa* (Bassett)

is a junior synonym of *D. ignota* (Osten Sacken) (Zhang *et al.* 2019, 2020) and *D. tuberculator* is synonymous with the older name *D. spinosa* (Shorthouse 1988; Y.M.Z. unpublished data), though we await confirmation of this through the revisionary work now underway. Nonetheless, in this paper we will treat these as two species-pairs rather than four individual species, in which case there are nine species reported for Manitoba prior to this paper. Note that we are treating *D. rosaefolii* (Cockerell) and *D. fusiformans* (Ashmead) as two separate species given their distinct gall morphology (leaf blister vs. stem swelling), despite the two having relatively low COI divergence of 2.4% (Zhang *et al.* 2019).

MATERIALS & METHODS

In this paper, we report one additional species for Manitoba, *D. gracilis* (Ashmead), and provide a summary of what is known about the distribution and host use of all *Diplolepis* species in the province. The distribution and host use information presented here comes from two sources: 1) opportunistic surveys conducted by C.F. in 2020 and 2021, the results of which have been uploaded to iNaturalist, and 2) observations uploaded to iNaturalist by other users through 27 October 2021. All observations were reviewed by Y.M.Z. to confirm the identification of the inducer based on Shorthouse (2010) and the gall voucher collection housed at USNM. C.F. has reviewed most observations to determine, where possible, the identity of the host plant using Scoggan (1957) and the Flora of North America (Flora of North America Editorial Committee 2014).

RESULTS

This is the first report of *Diplolepis gracilis* in Manitoba (Fig. 1). Galls were observed by C.F. on the underside of *R. woodsii* leaves in a remnant balsam poplar (*Populus balsamifera* L. (Salicaceae)) woodland in Warren (50.12928N, 97.54083W) on 19 September 2021 (Fig. 1 A–C). It has since been observed elsewhere in Warren on *R. woodsii* growing under bur oak (*Quercus macrocarpa* Michx. (Fagaceae)). Several galls were collected for rearing adults.

Prior to this report the distribution of *D. gracilis* in the prairie provinces was restricted to Alberta and western Saskatchewan (Shorthouse 2010). It has also been reported in southern Ontario (Beutenmuller 1914) and Minnesota (Olson 1964), so its occurrence in Manitoba is expected. Its range likely includes most of southern Manitoba, though it has been noted as rare in other parts of its range so it may not be abundant here (Shorthouse 2010). Previously reported hosts of *D. gracilis* include *R. woodsii*, *R. acicularis*, and *R. blanda* (Beutenmuller 1914; Shorthouse 2010; Looney and Eigenbrode 2011). Subsequent examination of Cynipidae material at WRME revealed a *D. gracilis* gall collected at Erickson on 30.viii.1997 along with the adult *Diplolepis* wasp that emerged from it (JBWM0210963).

Adult *D. gracilis* (Fig. 1 E–F) are active from mid-June through mid-July, with galls maturing in autumn and dropping with their host leaves (Shorthouse 1998). Eggs are laid and galls are induced on the underside of leaves. Shorthouse (1998) noted there is often a patch of red cells on the upper surface of leaves at the point of gall attachment; this discoloration of the upper leaf surface was also observed by C.F. on many of the galled leaves found in Warren.

In total, 179 observations of *Diplolepis*-induced galls were uploaded to iNaturalist through 27 October 2021, 166 for which the inducer could be determined with confidence (there are no observations showing adults). Twelve of these observations are of *Periclistus*-modified *D. nodulosa* (Beutenmeuller) galls for which the iNaturalist identification is *Periclistus*; unmodified *D. nodulosa* galls are inconspicuous stem swellings that are easily overlooked whereas galls modified by *Periclistus* are much larger (Brooks and Shorthouse 1998). There are no observations of *D. fusiformans* (Ashmead) galls in Manitoba on iNaturalist. Observations of conspicuous galls, such as those induced by *D. polita* (n=24) and *D. spinosa* (n=78), are relatively more abundant on iNaturalist compared to galls induced by *D. rosaefolii* (n=9, all by C.F.) which may not be recognized as galls by most iNaturalist users, or *D. radicum* (n=4) which occur at or just below ground level. However, this does not appear to be the case for *D. bicolor* (Harris), which produces showy galls yet only four observations have been uploaded to iNaturalist, perhaps indicating that it is uncommon in Manitoba.

Table 1. Species of Diplolepis reported in Manitoba. Species reported in Manitoba by previous authors are indicated by numbers: Shorthouse 2010 = 1, Deans and Nastasi 2021 = 2. SWPP = Spruce Woods Provincial Park, RMNP = Riding Mountain National Park.

Species	Exemplary iNaturalist Observation	Documented Distribution in Manitoba	Recorded Host(s) in Manitoba	Gall Location on Host
D. bicolor ^{1,2} (Harris)	90383041	Whiteshell; Grand Beach; Hecla; SWPP	R. woodsii	leaf
D. fusiformans ¹ (Ashmead) n/a	n/a			stem
D. gracilis (Ashmead)	95416841	Warren; Erickson	R. woodsii	leaf (underside)
$D. ignota^1$ (Osten Sacken) + $D. nebulosa^1$ (Bassett)	<u>95589462</u>	Winnipeg; Warren; St. Ambroise; Treesbank; SWPP; St. Lazare	R. arkansana	leaf (underside)
D. nodulosa¹ (Beutenmuller)	99142370 (only Periclistus-modified galls observed)	southern Manitoba north to R. woodsii RMNP	R. woodsii	stem
D. polita ^{1,2} (Ashmead)	30460026	southern Manitoba; disjunct observations at The Pas and Churchill	R. arkansana R. acicularis leaf (upperside)	leaf (upperside)
<i>D. radicum</i> ^{1,2} (Osten Sacken)	31204644	Winnipeg; Oak Hammock; Hadashville; Brokenhead; Erickson	R. acicularis R. woodsii x R. rugosa	adventitious shoots

Warren; Portage Sandhills; R. arkansana, R. woodsii Treesbank; SWPP; RMNP R. acicularis southern Manitoba north to R. woodsii
Hecla and RMNP
Warren; St. Lazare

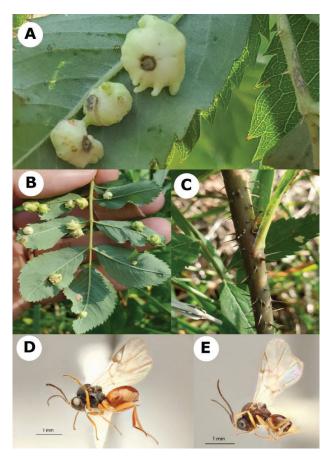


Figure 1. A-B) Gall of *Diplolepis gracilis*; C) Host plant *Rosa woodsii* showing the characteristic infrastipular prickles with enlarged bases; D) Lateral habitus of adult female *D. gracilis*; E) Lateral habitus of adult male *D. gracilis*.

DISCUSSION

A significant priority for future work on all *Diplolepis* species in Manitoba is to better understand and document their distribution. The northern extent of their ranges in Manitoba is unknown. Only *D. polita* has been recorded north of the line between Riding Mountain National Park and Hecla (51.2° latitude). Records of most *Diplolepis* species in Manitoba is based on the presence of their galls. Except for *D. spinosa* (CNCI, WRME), *D. gracilis* (WRME), and *D. radicum* (WRME), adult specimens of *Diplolepis* from Manitoba are either unidentified or not present in the collections checked. At least one additional species (*D. rosae* L.) may occur in Manitoba but has yet to be reported in the province. This introduced species from Europe is widely distributed in North America, but records are scarce in the northern great plains (Nastasi and Deans 2021).

The community science platform iNaturalist has proven valuable for documenting the presence and distribution of *Diplolepis* galls in Manitoba. Cynipid adults are small and inconspicuous and would rarely be observed and recorded by naturalists; the galls, however, are often noted even by those who are not looking for them. Additionally, the iNaturalist platform has provided many opportunities to engage with amateur naturalists to facilitate

information sharing about rose galls and increase general interest in looking for and documenting them. Indeed, there is an active community of gall enthusiasts on iNaturalist that identify observations and act as a resource for those wishing to learn more about these systems. The website www.gallformers.org provides additional gall identification resources.

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REFERENCES

- Beutenmuller, W. 1914. Notes on the genus *Rhodites* with descriptions of new species. Bulletin of the Brooklyn Entomological Society 9(5): 87–90.
- Brooks, S.E. and J.D. Shorthouse. 1998. Developmental morphology of stem galls of *Diplolepis nodulosa* (Hymenoptera: Cynipidae) and those modified by the inquiline *Periclistus pirata* (Hymenoptera: Cynipidae) on *Rosa blanda* (Rosaceae). Canadian Journal of Botany. 76(3): 365–381.
- Flora of North America Editorial Committee. 2014. Flora of North America North of Mexico. Vol. 9. Magnoliophyta: Picramniaceae to Rosaceae. Oxford Univ. Press, New York, USA.
- Looney, C. and S.D. Eigenbrode. 2011. Landscape-level effects on cynipid component communities of "orphaned" native shrubs. Journal of Insect Conservation 15(5): 695–706.
- Nastasi, L.F. and A.R. Deans. 2021. Catalogue of rose gall, herb gall, and inquiline gall wasps (Hymenoptera: Cynipidae) of the United States, Canada and Mexico. Biodiversity Data Journal 9: e68558.
- Olson, E. 1964. A preliminary study of Minnesota rose gall insects. Minnesota Academy of Science 32: 26–29.
- Scoggan, H.J. 1957. Flora of Manitoba. Department of Northern Affairs and National Resources, Ottawa, Canada.

- Shorthouse, J.D. 1988. Occurrence of two gall wasps of the genus *Diplolepis* (Hymenoptera: Cynipidae) on the domestic shrub rose, *Rosa rugosa* Thunb. (Rosaceae). The Canadian Entomologist 120(8-9): 727–737.
- Shorthouse, J.D. 1998. Role of *Periclistus* (Hymenoptera: Cynipidae) inquilines in leaf galls of *Diplolepis* (Hymenoptera: Cynipidae) on wild roses in Canada. *In* The Biology of Gall-Inducing Arthropods. *Edited by* G. Csóka, W.J. Mattson, G.N. Stone and P.W. Price. USDA, Forest Service, North Central Forest Experiment Station, St. Paul, MN, USA. 61–81 pp.
- Shorthouse, J.D. 2001. Galls induced by cynipid wasps of the genus *Diplolepis* (Cynipidae, Hymenoptera) on cultivated shrub roses in Canada. Acta Horticulturae 547: 83–92.
- Shorthouse, J.D., J.J. Leggo, M.D. Sliva, and R.G. Lalonde. 2005. Has egg location influenced the radiation of *Diplolepis* (Hymenoptera: Cynipidae) gall wasps on wild roses? Basic and Applied Ecology 6(5): 423–434.
- Shorthouse, J.D. 2010. Galls induced by cynipid wasps of the genus *Diplolepis* (Hymenoptera: Cynipidae) on the roses of Canada's grasslands. *In* Arthropods of Canadian Grasslands (Volume 1): Ecology and Interactions in Grassland Habitats. *Edited by* J.D. Shorthouse and K.D. Floate. Biological Survey of Canada, Ottawa, Canada. 251–279 pp.
- Zhang, Y.M., M.W. Gates, and J.D. Shorthouse. 2014. Testing species limits of Eurytomidae (Hymenoptera) associated with galls induced by *Diplolepis* (Hymenoptera: Cynipidae) in Canada using an integrative approach. The Canadian Entomologist 146(3): 321–334.
- Zhang, Y.M., Z. László, C. Looney, A.L. Dénes, R.H. Hanner, and J.D. Shorthouse. 2019. DNA barcodes reveal inconsistent species boundaries in *Diplolepis* rose gall wasps and their *Periclistus* inquilines (Hymenoptera: Cynipidae). The Canadian Entomologist 151(6): 717–727.
- Zhang, Y.M., M.L. Buffington, C. Looney, Z. László, J.D. Shorthouse, T. Ide, and A. Lucky. 2020. UCE data reveal multiple origins of rose gallers in North America: Global phylogeny of *Diplolepis* Geoffroy (Hymenoptera: Cynipidae). Molecular Phylogenetics and Evolution 153: p.106949.