Dwarf Lake Iris Recovery and Population Monitoring

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Cover: Flowering dwarf lake iris (*Iris lacustris*) and population monitoring quadrat in Wilderness State Park, Emmet County, Michigan (EO ID 7130) on June 8, 2020. Photograph by Rachel Hackett.

Table of Contents

List of Tables

List of Figures

Abstract

Dwarf lake iris (*Iris lacustris* Nutt.) is a perennial iris endemic to the Great Lakes region with its greatest stronghold in Michigan. The species is listed as threatened throughout its range under the Federal Endangered Species Act, and by every state or province where it occurs. Greatest threats to dwarf lake iris include habitat destruction, habitat degradation, succession, sand dune erosion, invasive species, and effects of climate change.

To update the status and better understand dwarf lake iris population growth and persistence, digital data mining efforts were employed to yield 6 new and update 42 existing populations. Spatial, qualitative, count and/or demographic surveys were conducted at 58 Michigan populations from 2019 to 2020. Fourteen populations underwent rank changes as a result of surveys. For count and demographic surveys, a census of the number of ramets in each stage, flowers, and capsules were counted for each 0.25 m^2 plot. Population estimation methods used by Chicago Botanic Gardens in their Plants of Concern Volunteer program were adapted to provide count-based population data for a population viability analysis following the methods of Dennis et al. (1991). The combination of new count data and existing count data found in literature was able to produce 36 population change increments across 17 [sub]populations, allowing us to reliably project 4 to 7 years in the future. The mean instantaneous stochastic growth rate (μ) was negative, but near zero with large variance (σ^2) that could span positive values: although the populations were likely to decrease between counts, positive growth of the population was inside the realm of possibilities. This was further illustrated by the low extinction rate of 14.2% of 1000 population growth simulations spanning 50 years. Cumulative distribution function predicted that DLI populations with 5,000 or less ramets had a 50% chance of extinction in 25 years. Large area and/or dense populations of dwarf lake iris are likely to persist several decades based on this analysis, but to reliably project 50 years into the future we would need to survey for an additional 8 to 15 years of count survey data; less if we added more populations.

As more reliable population data is collected, efforts should be made to prioritize, conserve, and protect known DLI populations. Twenty-four populations were selected as high priority having representation across the following characteristics: inland and shoreline populations, flower color, natural community, large area, and range-wide geography. Efforts should be made to continue documenting the status of remaining DLI populations to better predict its persistence. Efforts to better understand possible management effects on dwarf lake iris populations is needed to better advise landowners on how to promote the species fitness and persistence.

Introduction

Dwarf lake iris (*Iris lacustris* Nutt.; henceforth DLI) is a perennial iris endemic to the northern shores and few inland areas of Lake Michigan and Lake Huron of the Great Lakes of North America. DLI establishes itself in early successional habitat and some consider it a “gap-phase” species (Bray, 1956; Brokaw, 1987; Van Kley, 1989; Brotske, 2018). The species is listed as threatened throughout its range under the Federal Endangered Species Act, and by every state or province where it occurs. The small, blue, or rarely white DLI flower usually blooms from mid-May to mid-June (Figure 1). Its ramet density, flowering abundance, and fruit set is greatly affected by light levels and litter depth (Van Kley, 1989; Brotske, 2018).

Figure 1. The rare white flower variety grows along the typical blue flower of dwarf lake iris (*Iris lacustris*) at EO 10381, Emmet County, Michigan, on May 22, 2019. Photograph by Rachel Hackett.

Reproduction of DLI has been contributed to mostly asexual means of rhizomatous growth as derived from observation and low genetic diversity among and within populations (Makholm, 1986; Orick, 1992; Simonich, 1992). Recent seed dispersal and germination experiments have increased the likelihood that sexual reproduction is contributing to populations (Brotske, 2018): upwards of 82% of seeds were distributed by *Formica* sp. ants within 24 hours and germination experiments and found no significant difference between seed germination in experimentally disturbed and undisturbed/control sites. The added observation during these experiments that seedlings were unable to be distinguished from older ramets two weeks after emergence may contribute to the underestimation of population contributions from sexual reproduction (Brotske, 2018). A Michigan population has shown variance in ramet number at the permanent plot level over 5-9 years, but variance was statistically insignificant at the site-level for that time period (Ballard and Kowal, 1997).

Genetic studies in the 1990's on Michigan and Wisconsin populations showed little genetic variation among populations in each state (Orick, 1992; Simonich, 1992; Simonich and Morgan, 1994; Hannan and Orick, 2000). More recent and advanced genetic analysis among populations in Michigan and Wisconsin revealed four to five genetically distinct groups aligning geographically with one Lake Huron group, two Lake Michigan groups including the inland Menominee populations, and Brown County, Wisconsin (Cohen, 2021). The southernmost population in Brown County, Wisconsin, was the most distinct from other populations.

Approximately 200 records of DLI have been recorded, 91 of those in Michigan, USA, as determined after recent data mining efforts for this project and submitted Michigan Department of Natural Resources Threatened/Endangered Species Reports in 2020. Michigan contains both inland and shore populations on both Peninsulas with a variety of quality, area, density, and habitat on private, public, and non-governmental organizational land (Table 1; Michigan Natural Features Inventory, 2019). Greatest threats to DLI include habitat destruction, habitat degradation, succession, sand dune erosion, invasive species, and effects of climate change (e.g., extreme drought, variable Great Lake water levels; U S Fish and Wildlife Service, 2013).

Population viability analysis (PVA) was a step in the US Fish and Wildlife Service recovery plan (2013). The two most common methods to conduct PVA are count-based or demographic based. A count-based analysis requires population census data for at least 10 years to predict long-term species viability. Count-based analyses are less intensive from a data collection perspective, but they are not appropriate for all species. Count-based approaches are appropriate for plants that have no extreme fluctuations in population size, are easily identified, can have population turnovers between 10-20 years (e.g., not long-lived trees), whose population growth is not density dependent, are not self-compatible, have infrequent sexual reproduction, have low genetic variation within populations, and lack of large dormant seed banks (Dennis et al., 1991; Morris et al., 1999; Brigham and Thomson, 2003). DLI fits most of these assumptions, thus it would be appropriate to apply a long-term, count-based PVA for this species. Chicago Botanic Garden conducted a count-based population viability analysis while examining local and regional threats on a rare species of similar habit using forked aster (*Eurybia furcata*; Bernardo et al., 2018).

A demographic analysis differs from count-based analysis in that it differentiates between life stages and requires more field intensive work. Census data is gathered for each categorized life stage of the species and fecundity data for at least 2 years, preferably more to capture greater inherent variation and increase reliability (Zeigler et al., 2013). Demographic data is more field intensive, but the data is more informative and can examine or predict effects of management and threats on a population (Morris et al., 1999).

Our efforts focused on updating the status of accessible DLI populations in Michigan and collecting population data to be used in a count-based viability analysis at a select number of sites over the period of the Agreement.

Study areas

The Michigan Natural Heritage Database contained 85 element occurrence (EO) records of DLI at the start of this project (Michigan Natural Features Inventory, 2019; Table 1). All Michigan records occurred along the shore or within 32 km inland from Lake Michigan and Lake Huron (Figure 2). Records stretch west to east in the Upper Peninsula from north of Menominee, to

Drummond Island. Records stretch west to east in the Lower Peninsula from Wilderness State Park to 7 km south of Alpena. Islands with DLI records are Summer, Little Summer, Poverty and St. Martin Islands off the Garden Peninsula; Beaver, Garden, and Hog Islands northwest of Charlevoix; Big St. Martin and St Martin Islands northeast of St. Ignace; Bios Blanc and Round Islands north of Cheboygan; Les Cheneaux Islands south of Cedarville; and Drummond Island; and Thunder Bay Island in Alpena County.

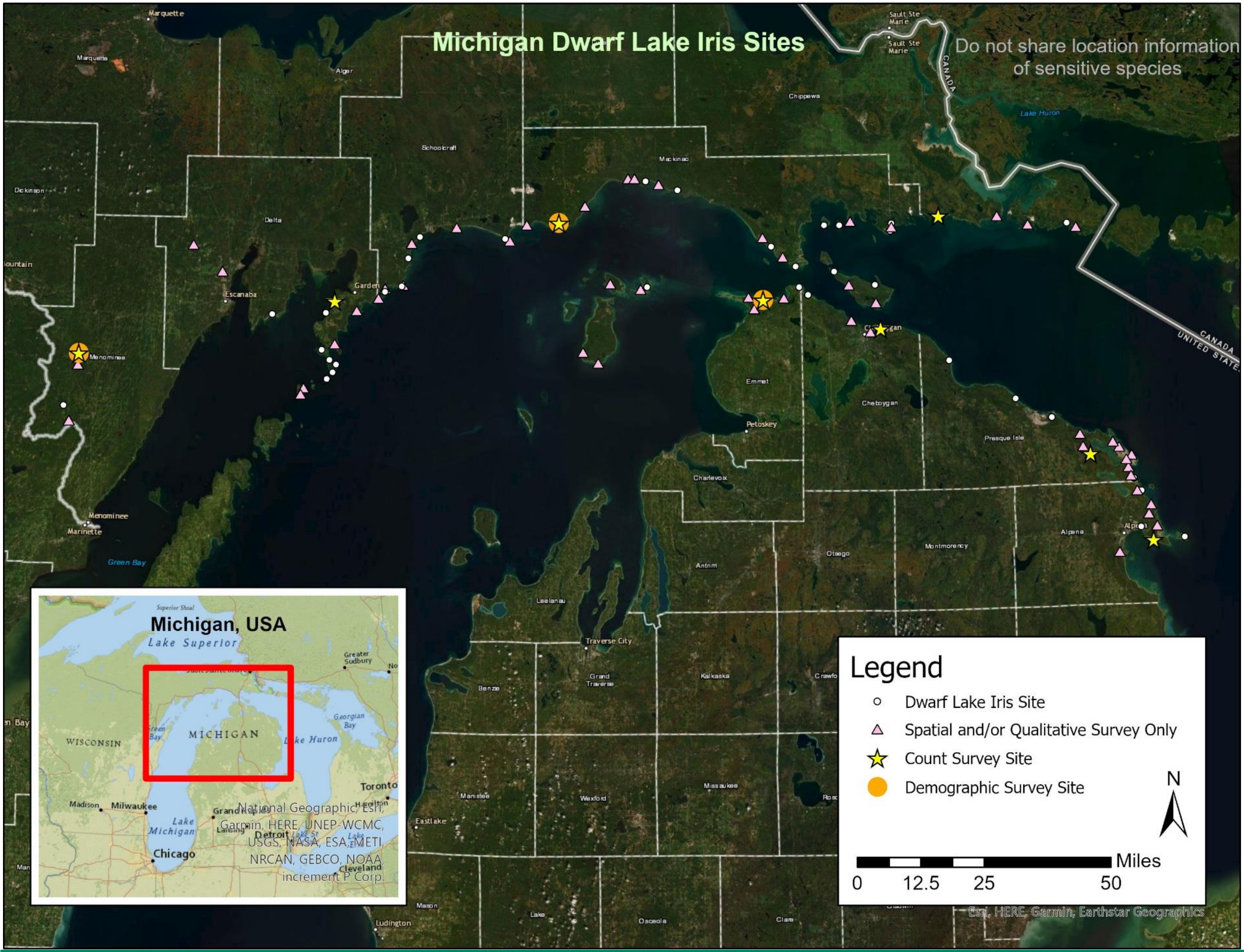

Figure 2. Map of dwarf lake iris (*Iris lacustris* Nutt.) Element Occurrence (EO) records (Michigan Natural Features Inventory, 2021). Sites visited from 2019 to 2021 are marked to represent the type of survey conducted: pink triangle - spatial and/or qualitative habitat survey only; yellow star - count survey; orange circle - demographic survey.

Table 1. List of dwarf lake iris (*Iris lacustris* Nutt.) Element Occurrence (EO) records (Michigan Natural Features Inventory, 2021). EO ID is a unique identifier for each EO record in Michigan's Natural Heritage Database. Rank is a qualitative assessment of estimated viability of species described in Table 2. A record was considered inland if it was greater than 1000 m from a Great Lake coast and appeared to be on post-glacial Lake Nipissing coast. Natural community abbreviations and descriptions can be found in Table 3. Surveys conducted from 2019 to 2021 were marked with a S for spatial survey, Q for qualitative survey, Q_{AS} for qualitative in archeologically sensitive site (i.e., no ground disturbance permitted), C for count survey, and D for demographic sampling. Changes in rank and last observation date due to recent surveys are noted via a strikethrough and listing of new value. Bolded records are categorized as high priority records.

Site	EO ID	Rank	County	Inland or Shore	Natural Communities	Ownership	Last Observation Date	2019-2021 Surveys
[redacted]	**256**	**A**	**Alpena**	**Shore**	**BFT, CFN, LBG, RCS**	**P, S**	**~~2010-08-10~~ 2021-05-26**	**S, Q**
[redacted]	**8385**	**~~BC~~ B**	**Alpena**	**Shore**	**BFT, NFN, RCS, LBG**	**P**	**~~1981-06-02~~ 2021-06-15**	**S, Q**
[redacted]	2837	~~A~~ F	Alpena	Shore	NFN	P, S	~~2002-05-07~~ 2020-06-11	Q
[redacted]	2440	AB	Alpena	Shore	WDS	N, P	~~1905-06-28~~ 2021-06-15	S, Q
[redacted]	3403	A	Alpena	Shore	WDS	N, P	~~1996-06-25~~ 2021-05-25	C
[redacted]	1625	C	Alpena	Shore	WDS	P, S	2002-08-08	
[redacted]	9817	CD	Alpena	Shore	LBS	F	1981-06-07	
[redacted]	6713	~~B~~ F	Alpena	Shore	RCS	P, S	~~2011-05-16~~ 2021-06-14	S, Q
[redacted]	**8775**	**B?**	**Alpena**	**Shore**	**CFN**	**P**	**2010-08-11**	
[redacted]	2472	~~BC~~ F	Charlevoix	Shore	OD	P	~~1999-08-19~~ 2019-06-25	Q
[redacted]	**1369**	**B**	**Charlevoix**	**Shore**	**BFT, CFN, SGB**	**P, S**	**~~1999-05-21~~ 2019-06-26**	**Q**
[redacted]	8033	C	Charlevoix	Shore	WDS	S	1999-08-19	
[redacted]	22194	~~E~~ D	Charlevoix	Shore	BFT	S	~~2000-07-16~~ 2019-06-27	S, Q
[redacted]	18917	CD	Charlevoix	Shore	LBG, RCS	S	~~2012-08-06~~ 2019-06-26	Q_{AS}
[redacted]	**8439**	**B**	**Cheboygan**	**Shore**	**CFN, GLB, WDS**	**N, S**	**~~1999-07-07~~ 2021-06-01**	**S, C**
[redacted]	10464	BC	Cheboygan	Shore	RCS	P	~~1996-08-20~~ 2021-06-16	S, Q
[redacted]	22657	~~E~~ D	Cheboygan	Shore	WDS	N	~~2018-07-19~~ 2020-06-11	S, Q
[redacted]	6907	B	Cheboygan	Shore	BFT, MNF, SGB	P, S	2018-06-20	

[1] Visited subset of area of EO record depending on landowner permissions granted.
[2] New EO record from results of data mining.

Site	EO ID	Rank	County	Inland or Shore	Natural Communities	Ownership	Last Observation Date	2019-2021 Surveys
[redacted]	12375	~~E~~ F	Chippewa	Shore	CFN	P	~~1989-06-08~~ 2019-07-10	S, Q
[redacted]	743	~~E~~ F	Chippewa	Shore	CFN	P	~~1989-06-22~~ 2019-07-10	S, Q
[redacted]	10288	B	Chippewa	Shore	CFN, RCS	P	1998-08-28	
[redacted]	10263	~~C~~ F	Chippewa	Shore	BFT, LBG, LBL	P	~~1990-06-15~~ 2021-06-16	S, Q
[redacted]	4640	~~C~~ F	Delta	Inland	BFT, NFN	P	~~1990~~ 2019-06-19	Q
[redacted] [redacted]	**5552**	**BC**	**Delta**	**Inland**	**ALV, RCS**	**P**	**~~1991-06-02~~** **2019-06-19**	**S, Q**
[redacted]	10711	X	Delta	Shore	LBL	S	1939-05-30	
[redacted] [redacted]	**22191**	**E**	**Delta**	**Shore**	**ALV**	**P, S**	**1998-05-29**	
[redacted] [redacted]	**2811**	**A**	**Delta**	**Shore**	**BFT, LBG, LBL**	**S**	**~~2004-06-02~~** **2021-06-09**	**S, Q$_{AS}$**
[redacted]	**5633**	**A**	**Delta**	**Shore**	**LBG, LBL**	**S**	**~~2008-09-08~~** **2019-06-19**	**Q**
[redacted]	11586	C	Delta	Shore	BFT, LBL	F	1996-06-15	
[redacted]	4466	BC	Delta	Shore	BFT, LBL, LCS	N	~~1981-07-08~~ 2021-05-19	S, C
[redacted] [redacted]	23699	F	Delta	Shore	BFT, LCS	F, P	~~2014-06-22~~ 2021-06-03	S, Q
[redacted] [redacted]	23701	~~E~~ D	Delta	Shore	BFT	F	~~2014-06-22~~ 2021-06-04	S, Q
[redacted]	11928	CD	Delta	Shore	BFT	S	1995-06-23	
[redacted]	3615	H	Delta	Shore	BFT, LBL	P	1968-05-30	
[redacted]	116	H	Delta	Shore	BFT, LBL	S	1968-06-01	
[redacted]	3132	C	Delta	Shore	BFT, WDS	F	2017-08	
[redacted]	7130	~~B~~ C	Emmet	Shore	LBL, LCS, WDS	S	~~2005-08-26~~ 2021-06-11	S, C, D
[redacted]	13051	CD	Emmet	Shore	WDS	P, S	1981-05-14	
[redacted]	**10381**	**C**	**Emmet**	**Shore**	**BFT**	**S**	**~~1991-05~~** **2019-06-17**	**S, Q**
[redacted] [redacted]	3606	B	Emmet	Shore	LSC, WDS	M, P, S	~~2001-06-20~~ 2021-06-18	S, Q
[redacted]	11844	~~H~~ F	Emmet	Shore	CFN	S	~~1966-06-11~~ 2020-06-20	S, Q
[redacted]	**8964**	**A**	**Mackinac**	**Shore**	**BFT, LBG, LBL**	**N, P, S**	**~~2010-08-15~~** **2021-05-20**	**C**
[redacted]	5954	B	Mackinac	Shore	WDS	S	~~2001-06-05~~ 2020-06-17	S, Q

Site	EO ID	Rank	County	Inland or Shore	Natural Communities	Ownership	Last Observation Date	2019-2021 Surveys
[redacted]	15826	C	Mackinac	Shore	CFN,	P	1993-08-10	
[redacted]	12376	AB	Mackinac	Shore	WDS	P, S	~~2002-08-23~~ 2021-06-10	C, D
[redacted].	8201	C	Mackinac	Shore	BFT	P	2001-05-23	
[redacted]	12548	B	Mackinac	Shore	IDW, LBC	P	2001-08-23	
[redacted]	10153	C	Mackinac	Shore	BFT, CFN	S	~~2001-06-06~~ 2019-06-21	S, Q
[redacted]	835	~~C~~ B	Mackinac	Shore	BFT	S	~~2001-06-01~~ 2020-06-18	Q
[redacted]	1885	BC	Mackinac	Shore	BFT	P	1994-09	
[redacted]	**5377**	**AB**	**Mackinac**	**Shore**	**BFT, NFN**	**P**	**1999-06-11**	
[redacted] [redacted]	8623	~~C~~ B	Mackinac	Shore	BFT	S	~~1991-05-31~~ 2020-06-17	S, Q
[redacted] [redacted]	834	C	Mackinac	Shore	BFT, LBL, LCS	P	~~2001-06-04~~ 2021-07	Q
[redacted] [redacted]	8202	C	Mackinac	Shore	BFT	P	~~2001-06-04~~ 2021-07	Q
[redacted] [redacted]	24196	E	Mackinac	Shore	RCS	S	2019-07-12	
[redacted]	12503	AB	Mackinac	Shore	BFT, NFN	P	1999-06-11	
[redacted]	4458	F	Mackinac	Shore	WDS	F	~~1991-09-10~~ 2021-06-10	Q
[redacted]	3635	BC	Mackinac	Shore	CFN, SGB	P	2011-06-08	
[redacted]	10154	BC	Mackinac	Shore	BFT, SGB	F	2008-06-19	
[redacted] [redacted]	12862	A	Mackinac	Shore	BFT, WDS	N, P, S	~~1997-07-11~~ 2020-06-16	S, Q
[redacted]	15825	C	Mackinac	Shore	BFT, NFN	P	1993-08-12	
[redacted] [redacted]	24245	~~E~~ C	Mackinac	Shore	CFN	F	~~2016-07-20~~ 2020-06-15	S, Q
[redacted] [redacted]	**12221**	**AB**	**Mackinac**	**Shore**	**BFT, CFN, LCS**	**P, S**	**~~2002-07-24~~ 2020-06-19**	**S, Q**
[redacted] [redacted]	12547	C	Mackinac	Shore	BFT	P	~~1993-09-15~~ 2020-06-12	Q
[redacted]	**15125**	**BC**	**Menominee**	**Inland**	**BFT, MNF**	**S**	**~~2005-05-20~~ 2021-06-05**	**S, C, D**
[redacted]	**16477**	**AB**	**Menominee**	**Inland**	**BFT, MNF**	**P**	**2005-05-26**	
[redacted]	**5149**	**BC**	**Menominee**	**Inland**	**MNF**	**P**	**~~2010-07-18~~ 2021-06-05**	**S, Q**
[redacted]	**15176**	**C**	**Menominee**	**Inland**	**MNF**	**S**	**~~2005-06-02~~ 2019-06-18**	**S, Q**

[3] New EO record derived from newly submitted Michigan Department of Resources Threatened/Endangered Species Report by GEI Consultants.

Site	EO ID	Rank	County	Inland or Shore	Natural Communities	Ownership	Last Observation Date	2019-2021 Surveys
[redacted]	**2058**	**C**	**Presque Isle**	**Shore**	**ALV**	**P**	**2002-07-11**	
[redacted]	2235	~~B~~ F	Presque Isle	Shore	BFT	P, S	~~1996-06-26~~ 2019-05-28	S, QAS
[redacted]	10481	C	Presque Isle	Shore	BFT, LCS, RCS	P	~~1989-07-13~~ 2019-05-28	S, Q
[redacted]	1854	C	Presque Isle	Shore	BFT	P	1981-06-26	
[redacted]	4553	C	Presque Isle	Shore	BFT	P	1996-06-28	
[redacted]	15944	B	Presque Isle	Shore	LBG	S	~~1996-06-15~~ 2020-06-30	Q
[redacted]	11321	B	Presque Isle	Shore	WDS	P	~~1998-06-10~~ 2019-05-31	S, Q
[redacted]	**5551**	**C**	**Presque Isle**	**Shore**	**CFN, WDS**	**S**	**2004-06-22**	**Q**
[redacted]	**8162**	**B**	**Presque Isle**	**Shore**	**LBG, BFT**	**P, S**	**~~1996-06-15~~ 2021-05-27**	**S, C**
[redacted]	**10080**	**A**	**Presque Isle**	**Shore**	**CFN, WDS**	**S**	**~~2002-05-07~~ 2021-05-28**	**S, Q**
[redacted]	23795	C	Presque Isle	Shore	LBG, LCS	P	2019-05-28	S, Q
[redacted]	**10918**	**A**	**Presque Isle**	**Shore**	**BFT, LBG, NFN, WDS**	**P, S**	**~~2010-08-13~~ 2020-06-30**	**QAS**
[redacted]	10888	B	Presque Isle	Shore	WDS	P	~~2016-06-23~~ 2021-06-16	S, Q
[redacted]	9196	C	Schoolcraft	Shore	BFT, LBL	P	2000-08-09	
[redacted]	6351	C	Schoolcraft	Shore	BFT, LCS, SGB	P	~~2017-08-06~~ 2021-06	S, Q
[redacted]	**12942**	**B**	**Schoolcraft**	**Shore**	**BFT, CFN, WDS**	**P**	**~~2000-08-09~~ 2021-06**	**S, Q**
[redacted]	4465	BC	Schoolcraft	Shore		P	~~1981-06-26~~ 2021-06	Q
[redacted]	1788	C	Schoolcraft	Shore	BFT, WDS	P	2000-08-08	
[redacted][1]	**3589**	**BC**	**Schoolcraft**	**Shore**	**BFT, LBG, LCS**	**P, S**	**~~2016-07-30~~ 2021-06**	**S, Q**
[redacted]	6809	B	Schoolcraft	Shore	BFT, LCS	P	~~2006-08-22~~ 2021-06	Q
[redacted]	8842	C	Schoolcraft	Shore	WDS	P	2000-08-08	
[redacted]	973	C?	Schoolcraft	Shore	BFT	P	2001-08-13	
[redacted]	8015	BC	Schoolcraft	Shore	WDS	P, S	~~1991-06-01~~ 2021-06	Q

[4] With new survey, [redacted] (EO 11321) and [redacted] (EO 10888) no longer have sufficient separation distance and will be combined.
[5] With new survey, [redacted](EO 5551) and [redacted] (EO 10080) no longer have sufficient separation distance and will be combined.
[6] New EO record derived from land manager contact.

Table 2. Definitions of basic EO Ranks for species as defined by NatureServe. Abridged table of that developed by NatureServe (2021).

Rank	Definition
A	**Excellent estimated viability** (species) - Based on current information on E0 rank factors (i.e ., condition, size, and landscape context) for the E0, it is believed to have an excellent probability of persisting, if current conditions prevail, for a defined period of time, typically 20-100 years.
B	**Good estimated viability** (species) - Based on current information on E0 rank factors (i.e., condition, size, and landscape context) for the E0, it is believed to have a good probability of persisting, if current conditions prevail, for a defined period of time, typically 20-100 years.
C	**Fair estimated viability** (species) - Based on current information on E0 rank factors (i.e., condition, size, and landscape context) for the E0, it is believed to have a fair probability of persisting, if current conditions prevail, for a defined period of time, typically 20-100 years.
D	**Poor estimated viability** (species) - Based on current information on E0 rank factors (i.e., condition, size, and landscape context) for the E0, it is believed to have a poor probability of persisting, if current conditions prevail, for a defined period of time, typically 20-100 years.
E	**Verified Extant** (species) - E0 has been recently verified as still existing, but sufficient information on the factors used to estimate viability of the occurrence has not yet been obtained. Use of the E rank should be reserved for those situations where the occurrence is thought to be extant, but an A, B, C, D, or range rank cannot be assigned.
H	**Historical** (species) - There is a lack of recent[7] field information verifying the continued existence of the E0, such as when the occurrence is based only on historical collections data, or when the occurrence was ranked A, B, C, D, or E at one time and is later, without field survey work, considered to be possibly extirpated due to general habitat loss or degradation of the environment in the area.
F	**Failed to find** - E0 has not been found despite a search by an experienced observer at a time and under conditions appropriate for the Element at a location where it was previously reported, but that still might be confirmed to exist at that location with additional field survey efforts. For EOs with vague locational information, the search must include areas of appropriate habitat within the range of locational uncertainty. An F rank, when applicable, supersedes an A, B, C, D, E, or H rank.
X	**Extirpated** - There is documented destruction of the habitat or environment of the E0, or persuasive evidence of its eradication based on adequate survey (i .e. , thorough or repeated survey efforts by one or more experienced observers at times and under conditions appropriate for the Element at that location).
U	**Unrankable** - An E0 rank cannot be assigned due to lack of sufficient information on the occurrence.
NR	**Not Ranked** - An EO rank has not yet been assigned to the occurrence.

[7] The term *recent* is generally interpreted as follows: […] For plants or communities, there has been a field survey of the occurrence within the last 20 to 40 years. This higher maximum time limit is based upon the assumption that occurrences of these Elements generally have the potential to persist at a given location for longer periods of time due to plant biology and community dynamics. However, landscape factors must also be considered; thus, areas with more anthropogenic impacts on the environment will be at the lower end of the range, and less-impacted areas will be at the higher end. These time frames represent suggested maximum limits, however the actual time period for historical EOs may vary according to the biology of the Element and the specific landscape context of each occurrence (including anthropogenic alteration of the environment).

Table 3. Natural community types where Michigan DLI populations have been observed. Descriptions have been abridged from Cohen et al. (2015).

Abbr.	Natural Community	Description
ALV	Alvar	a grass- and sedge-dominated community, with scattered shrubs and sometimes trees. The community occurs on broad, flat expanses of calcareous bedrock covered by a thin veneer of mineral soil, often less than 25 cm.
BFT	Boreal forest	a conifer or conifer-hardwood forest occurring on moist to dry sites characterized by species dominant in the Canadian boreal forest. The community occurs primarily on sand dunes, glacial lakeplains, and thin soil over bedrock or cobble. Sand and sandy loam are moderately acid to neutral, but heavier soils and more acid conditions are common.
CFN	Coastal fen[8]	a sedge- and rush-dominated wetland that occurs on calcareous substrates along Lake Huron and Lake Michigan north of the climatic tension zone. The community occurs where marl and organic soils accumulate in protected coves and abandoned coastal embayments and grade to moderately alkaline glacial tills and lacustrine sediments lakeward. Sediments along the lakeshore are typically fine-textured and rich in calcium and magnesium carbonates.
GLB	Great Lakes barrens[8]	a coniferous savanna community of scattered and clumped trees, and an often dense, low or creeping shrub layer. The community occurs along the shores of the Great Lakes where it is often associated with interdunal wetland and open dune.
IDW	Interdunal wetland	a rush-, sedge-, and shrub-dominated wetland situated in depressions within open dunes or between beach ridges along the Great Lakes, experiencing a fluctuating water table seasonally and yearly in synchrony with lake level changes.
LBG	Limestone bedrock glade	an herb- and graminoid-dominated plant community with scattered clumps of stunted trees and shrubs growing on thin soil over limestone or dolomite. Tree cover is typically 10 to 25%, but occasionally as high as 60%. Shrub and herb cover is variable and there are typically areas of exposed bedrock. Mosses, lichens, and algae can be abundant on the exposed limestone bedrock or thin organic soils. Seasonal flooding and summer drought maintain the open conditions.
LBL	Limestone bedrock lakeshore	a sparsely vegetated natural community dominated by lichens, mosses, and herbaceous vegetation. This community, which is also referred to as alvar pavement and limestone pavement lakeshore, occurs along the shorelines of northern Lake Michigan and Lake Huron on broad, flat, horizontally bedded expanses of limestone or dolomite bedrock.
LCS	Limestone cobble shore	occurs along the northern Lake Michigan and Lake Huron shorelines. The community is typically sparsely vegetated, because cobbles cover most of the surface and storm waves prevent the development of a diverse, persistent community.
MNF	Mesic northern forest[8]	a forest type of moist to dry-mesic sites lying mostly north of the climatic tension zone, characterized by the dominance of northern hardwoods, particularly sugar maple (*Acer saccharum*) and American beech (*Fagus grandifolia*). It is primarily found on coarse-textured ground and end moraines, and soils are typically loamy sand to sandy loam.
NFN	Northern fen[8]	a sedge- and rush-dominated wetland on neutral to moderately alkaline saturated peat and/or marl influenced by groundwater rich in calcium and magnesium carbonates. The community is found where calcareous bedrock underlies a thin mantle of glacial drift on flat areas or shallow depressions.
OD	Open dunes[8]	a grass- and shrub-dominated multi-seral community located on wind-deposited sand formations near the shorelines of the Great Lakes. Dune formation and the patterning of vegetation are strongly affected by lake-driven winds.
RCS	Rich conifer swamp[8]	a groundwater-influenced, minerotrophic, forested wetland dominated by northern white-cedar (*Thuja occidentalis*) that occurs on organic soils (i.e., peat). The community is also referred to as cedar swamp.
SGB	Sand and gravel beach[8]	occur along the shorelines of the Great Lakes and on some larger freshwater lakes, where wind, waves, and winter ice cause the shoreline to be too unstable to support aquatic vegetation. These beaches are typically open, with sand and gravel sediments and little or no vegetation.
WDS	Wooded dune and swale complex	a large complex of parallel wetland swales and upland beach ridges (dunes) found in coastal embayments and on large sand spits along the Great Lakes. The upland dune ridges are typically forested, while the low swales support a variety of herbaceous or forested wetland types, with open wetlands more common near the shoreline and forested wetlands more prevalent further from the lake. Wooded dune and swale complexes may encompass several natural communities.

[8] Natural community may be included under the WDS Complex.

Methods

Data mining for Element Occurrence records

Searches for digital occurrence records from preserved specimens and human observations were compiled from the Consortium of Midwest Herbaria (http://midwestherbaria.org), iDigBio (https://www.idigbio.org), and the Global Biodiversity Information Facility (GBIF; https://www.gbif.org) in 2019.

An iNaturalist “Traditional Project” was created to compile and monitor new DLI observations via the community science application (https://www.inaturalist.org/projects/dwarf-lake-iris-iris-lacustris-in-michigan; Figure 3). Alerts of new observations in Michigan were checked against existing EO records and follow-ups with the observer for more information were made as needed. Occasional journal entries were generated to draw interest, but lack of time and resources prevented regular entries.

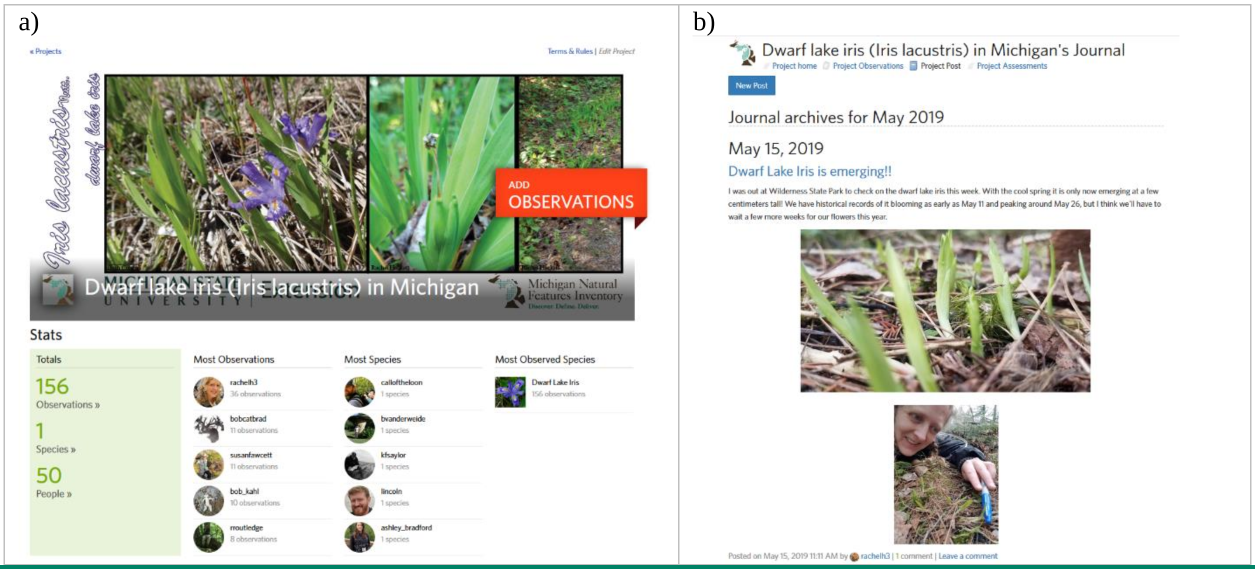

Figure 3. Screenshots of iNaturalist project *Dwarf lake iris (*Iris lacustris*) in Michigan* (*https://www.inaturalist.org/projects/dwarf-lake-iris-iris-lacustris-in-michigan*): a) is the main page and b) is the first journal entry.

Field surveys

Site selection

Sites for surveys and survey types were selected based on the following criteria:

- Cost of access (e.g., ferry costs)
- Previous research
- Survey needs mentioned in record
- Uncertainty of spatial extent of population
- Year since last observation
- Ownership and likelihood to gain access
- Variety of rank
- Both inland and shore locations

Sites where previous quantitative censuses and/or genetic research had been conducted (e.g., Ewert and Scrimger, 1989; Van Kley, 1989; Orick, 1992; Ballard and Kowal, 1997) were prioritized. Within the record, any survey needs that were mentioned, uncertainty of the spatial extent of the population, and time since last observation also weighed into selected sites. Sites were also examined to cover a variety of EO ranks and both inland and shore records. Inland populations were shown to have greater genetic variation within and among populations than shore populations indicating that they are likely glacial relict populations (Orick, 1992).

Spatial surveys
Records of DLI are only as good as their source information. The geographic information available for EO records can be vague, especially for older records, or at sites where resources were not available to allow for the full extent of the population to be determined.

Records with uncertain population extent were prioritized for landowner contact to achieve survey status. Prior to field survey, the aerial imagery and land cover/use maps were used to determine likely extent of population and area to survey. Land access permissions were sought for survey when costs were not prohibitive. In the field, a meander survey in areas of suitable habitat was conducted to assess the extent of the DLI population. Photographs and GPS coordinates were collected at significant transitions and points of interest. Often qualitative surveys were conducted in conjunction with spatial surveys. Spatial surveys could be conducted during any point of the growing season.

Qualitative surveys
Qualitative surveys provide current information and estimates to use in EO ranking for a record. These surveys are quick, provide presence/absence data, produce a density range estimate, generate a current assessment of threats to the population, and can provide qualitative population trends over time. Qualitative surveys can be conducted during any point of the growing season. Soil depth, soil type, litter depth, canopy openness and solar radiation were measured in at least three points for each qualitative survey (Table 4). Soil moisture in each site was categorized into dry, moist, wet, saturated, inundated, or other. Invasive plant species were noted and classified into DAFOR abundance scale (Voss and Reznicek, 2012). Signs of animal impact (e.g., browsing, trampling) were noted and categorized into no impact (0% of DLI population affected), low impact (0 – 50% of DLI population affected), or high impact (51 – 100% of DLI population affected) as used by the Chicago Botanic Garden's Plants of Concern program (Bernardo et al., 2018; Goad et al., 2018).

On lands indicated by landowners or managers to be sensitive to ground disturbance (e.g., of archeological significance), some habitat measurements were waived (Table 4).

Table 4. Description of differences in data collection for each survey type

		Survey Type				
		Spatial	**Qualitative – Sensitive sites**	**Qualitative**	**Count**	**Demographic**
Sampling method		Meander	Meaner	Meander	Transect-quadrat	Transect-quadrat[9]
Data collected	GPS	X	X	X	X	X
	Photographs	X	X	X	X	X
	Presence/absence		X	X	X	X
	Density Range Est.		X	X		
	Threats		X	X	X	X
	Soil Depth			X	X	X
	Soil Type			X	X	X
	Litter depth			X	X	X
	Canopy openness[10]		X	X	X	X
	Solar Radiation			X	X	X
	Soil Moisture			X	X	X
	Invasive Plant Species Density Estimate		X	X		
	Invasive Plant Species percent cover, height, etc.				X	X
	Quadrat (0.25 m^2) ramet counts				X	X
	Specimens collected			X[11]	X	X

Count surveys

Count surveys are more time consuming than qualitative surveys but produce precise data with repeatable methods to be used to predict population trends using population viability analysis derived from other populations or, if collected for at least 10 growing seasons, as a portion of a count-based population viability analysis. Count surveys are best conducted during flowering or fruiting periods.

The methods of Van Kley (1989) were adopted to maintain consistency among usable count census records. Ten random transects were placed approximately perpendicular the shore. At inland sites, the transects were placed perpendicular to the topography. If there was not a colony intersecting the random transect, the transect was conducted at the nearest colony of DLI. A belt transect was used to determine the number of colonies and percent DLI cover for each transect. Each colony that has a ramet that falls within 2 m of the belt transect was considered intercepted. The transect ran until no DLI was intercepted for 40 m. For records with multiple delineated polygons for the same population, one of three courses of action was taken: 1) if the record was a site of previous research, only polygons included in that study were

[9] Quadrats and plants marked for revisitation in 2020.
[10] Began measurements in 2020 field season, because Wisconsin scientists were collecting this measurement.
[11] Only at sites with large populations (i.e., ranks A to BC) or unique features (e.g., white flowers, inland).

surveyed, 2) polygons were selected based on accessible permissions and 3) transects were divided among the polygons in an area-proportional manner with at least one transect in a polygon greater than 1 ha.

Quadrats of 0.25 m^2 area were placed on a randomly selected intercepted colony. At least 10 quadrats were placed at each site. The quadrats were placed a random distance from where the colony first intercepted the transect. The separation distance between colonies was at least 1 m between ramets.

A census of the number of ramets in each stage, flowers, and capsules were counted for each quadrat (Table 5). Soil depth, soil type, litter depth, canopy openness, and either full spectrum solar radiation or categorical sunlight amount (e.g., partial sun) were measured depending on availability and functionality of equipment. Soil moisture in each transect and quadrat was categorized into dry, moist, wet, saturated, inundated, or other. All invasive herbaceous species within the belt transect and quadrat and invasive woody vegetation within 2.5 m was identified (Voss and Reznicek, 2012). Signs of animal impact (e.g., browsing, trampling) were noted and categorized for each belt transect and quadrat into no impact (0% of DLI affected), low impact (0 – 50% of DLI affected), or high impact (51 – 100% of DLI affected) as used by the Chicago Botanic Garden's Plants of Concern program (Bernardo et al., 2018; Goad et al., 2018).

The mean ramet count was used to estimate a number of stems across the population. These estimates were comparable to the methods of the Chicago Botanic Gardens Species of Concern Handbook methods: To estimate DLI counts per colony, the number of plants quadrat were calculated, averaged (i.e., mean), transformed to 1 unit area, and extrapolated across the population area based on the area of the population polygon on record (Goad et al., 2018).

Photographs were taken of the belt transect at the start of each line, toward the population. Photographs were taken to record habitat (Figure 4a). At least one photograph was captured from approximately 1.5 m above the quadrat so that the entire quadrat is contained in the frame (Figure 4b). At least one of the sides of the quadrat was marked in metric units to provide a unit measurement for image comparison.

Table 5. Description of DLI life stages for surveys at/after flowering time

Stage	**Description**
Young Ramet	Ramet less than 5.25 cm tall and lacks sexual reproductive organs (i.e., flower, fruit)
Sterile Adult	Ramet greater than 5.25 cm, but lacks sexual reproductive organs
Reproductive Adult	Ramet has sexual reproductive organs
Dead	No vegetative growth in subsequent year

a) b)

Figure 4. Examples of photographs taken at a quadrat during a count survey at EO 8439, Cheboygan County, Michigan, on June 1, 2019 : a) a photograph of the habitat, b) a photograph of the quadrat and density. Photographs by Rachel Hackett.

Demographic surveys

Belt transects were conducted in the same manner as for the count surveys as for demographic surveys. The quadrats for demographic surveys were permanently marked flags in the two opposite corners of the quadrat. Each ramet received a marked fluorescent tee at its base, labeled with unique sequential numbers. Each ramet was designated a life stage class (Table 5). In subsequent years, the quadrats were located using GPS coordinates and the quadrat flags, ramets linked to nearest marker, and their stage recorded. Demographic surveys were conducted during flowering (preferred) or fruiting period.

Specimen collection

Up to three whole specimens were collected and georeferenced at each count survey site and thriving sites (i.e., ranked A to BC). At sites where specimens were collected, at least three ramets were collected from areas in the population separated by at least 10 m and at least 1 m from a survey quadrat. Flower, leaves, roots, and rhizomes were removed, pressed, dried, and deposited in the Central Michigan University Herbarium for archiving and digitization.

The deposition of preserved plant specimens into a natural history collection increases the legitimacy and longevity of the research (Lane, 1996; Beaman and Cellinese, 2012; Turney et al., 2015; Antunes and Schamp, 2017; Heberling and Isaac, 2017; Rudin et al., 2017; Wilson-Brodie et al., 2017; Yost et al., 2018). Preserved specimens records and their material can prove to be essential for future research in topics such as biogeography, chemical contaminant trends, evolutionary biology, population genetics, and species habitat distribution models

(Chapman, 2005; Polgar et al., 2013; García-Roselló et al., 2015; Juhász et al., 2016; Antunes and Schamp, 2017; Rudin et al., 2017; Soltis, 2017; Mayor et al., 2017; Pacifici et al., 2017; Yost et al., 2018; Folk, Sun, et al., 2018; Folk, Visger, et al., 2018; Funk, 2018).

Count-based population viability analysis

The methods of population estimation used by Chicago Botanic Gardens in their Plants of Concern Volunteer program were adapted to provide count-based population data for a population viability analysis (Bernardo et al., 2018; Goad et al., 2018). For the populations that underwent count surveys, each mapped polygon of the population was called a sub-population and had a separation distance of at least 50 m between DLI colonies. The mean ramet count per quadrat in each polygon during the same year was used to determine plants per 1 m^2 in each polygon. The mapped area was used to extrapolate DLI from population density to the total area of the polygon for estimated total ramets per [sub]population, although it is recognized that DLI was not contiguous throughout the area. Since the area values were consistent for the count years, it is unlikely to affect the end result of the population viability analysis overall.

We followed the methods of Dennis et al. (1991) as described by Elderd et al. (2003) to determine the mean instantaneous stochastic growth rate (μ) and variance of stochastic growth (σ^2). In alignment with this process, we transformed the count data to be described by a linear model of the rate of population change over time verses the length of time using the formulas:

$$x = \sqrt{t_j - t_i}$$

$$y = \frac{\ln(N_j) - \ln(N_i)}{x}$$

where *j* is the later year of the two counts, *i* is the earlier year of the two counts, *t* is year value, and *N* is the population estimate. Using a linear regression on the resulting line with *y*-intercept set at 0, the slope of the line is an estimate of μ and the variance of the individual data points on the line gives variance σ^2.

Other constants for the simulation were derived from the count data. The quasi-extinction threshold was set at 500 ramets; carrying capacity (*K*) at the maximum of population estimates; and starting population at the median of observed population estimates. The simulations ran for 50 years, and 1,000 replicate simulations were run. An extinction probably was calculated from these simulations.

To determine the probability of extinction of a DLI population the cumulative distribution function (CDF) was used (Dennis et al., 1991; Morris and Doak, 2002; Elderd et al., 2003). All constants for the simulation were reused in the CDF except for starting population. DLI populations of various starting ramet estimates were used to illustrate the effects on populations of differing sizes.

Demographic-based population viability analysis

Each year-to-year transition of a marked ramet was used to compute a life-stage transitional matrix (i.e., Leslie Matrix; Table 6). These results were pooled across all demographic quadrats. Marked individuals whose markers were lost or unreadable were not included in the calculation of the life-stage transitional matrix. Individuals that were not associated with a marker in subsequent years were marked as new growth and given a marker. For quadrats that were lost, all individuals marked in the previous year was presumed dead.

For each transitional period of each demographic quadrat, the constant rate of population growth (λ), instantaneous growth rate (r), number of markers documented in both years, number of markers missing in subsequent year, number of newly marked ramets in subsequent year, documented mortality of marked ramets, and survivorship of marked ramets were calculated.

To develop the life-stage transitional matrix, first the ramets marked in both years were counted in the appropriate life stage cell corresponding to a transition between life stages (Table 6; Figure 5). Since much of DLI growth is due to vegetative growth from rhizomes, the new growth can be generated from and to multiple life stages. To account for the new growth in the subsequent year, the new ramets at each stage was contributed proportionally to the survivors of each life stage in the previous year similar to DeWalt (2004). For example, 12 new growth of Sterile Adults would be divided between 3 surviving Young Ramet, 20 surviving Sterile Adults, and 1 Reproductive Adult, 13% (1.56 ramets) would be attributed to the Young Ramet to Sterile Adult transition cell, 83% (9.96 ramets) would be attributed to the Sterile Adult to Sterile Adult transition cell, and 4% (0.48 ramets) would be attributed to the Reproductive Adult to Sterile Adult transition cell. The transitional rate for each stage was calculated by dividing the ramets attributed to each transitional stage by the total number of ramets in the first year, including the non-survivors.

Transitional matrices were calculated at the quadrat-transitional year level, and then life stage matrices pooled and then converted to transitional matrices at the location-year and survey-level. The overall growth rate (λ), sensitivity, and stable stage distributions were calculated from the survey-level transitional matrix.

To project DLI populations 50 years, we used the multiple matrices approach with 1000 iterations. At each time step, a transitional matrix pooled at location-year level derived from demographic surveys was randomly selected to calculate the population of each life stage for the following year. This approach can allow for the inclusion of real life disturbances that may occur and is based on observed data. A limitation of this approach is that only disturbances that affected the population during the years it was observed could be included, thus restricting the possible combinations of vital rates the simulation can generate (Morris et al., 1999). If the total population reached 500,000,000 ramets (K) in an iteration, that iteration stopped and a new one began. The quasi-extinction threshold was set at 500 ramets.

DLI populations were projected using the initial abundances per life-stage in Table 6. Each of the four initial abundances were means derived from the minimum population estimate data collected from the most recent Michigan DLI qualitative, count, and demographic surveys of populations of the corresponding ranks as described by NatureServe (Table 1; Table 2). EOs ranked A and AB were included in calculations for those ranked A; B and BC in B, C and CD in C, and D in D. To determine the proportion of the estimate allotted to Young Ramet, overall mean across all life-stage matrices was used (7%). To determine the proportion of Reproductive Adults, the mode of the categorical density of flowers/fruits in the populations of the rank was used: frequent fruits/flowers were 25% of the total population, occasional were 10%, and rare were 2%. The remaining ramets were assigned Sterile Adults.

To determine the probability of extinction of a DLI population, the mean of 10 quasi-extinction runs of 1000 iterations each were calculated for each rank with different initial abundances (Morris and Doak, 2002; Stubben et al., 2020). All constants for the simulation were reused in the quasi-extinction simulations. DLI populations of different ranks were used to illustrate the effects on populations of differing size and quality.

Analyses were conducted using R version 4.1.1 and R package “popbio” (Stubben et al., 2020).

Table 6. Example transitional matrix.. Columns are life stage in the first year; rows are the life stage in the second year. Values are illustrated in .

	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	$S_{1,1}$	$F_{1,2}$	$F_{1,3}$
Sterile Adult	$G_{2,3}$	$S_{2,2}$	$S_{2,3}$
Reproductive Adult	$G_{3,1}$	$G_{3,2}$	$S_{3,3}$

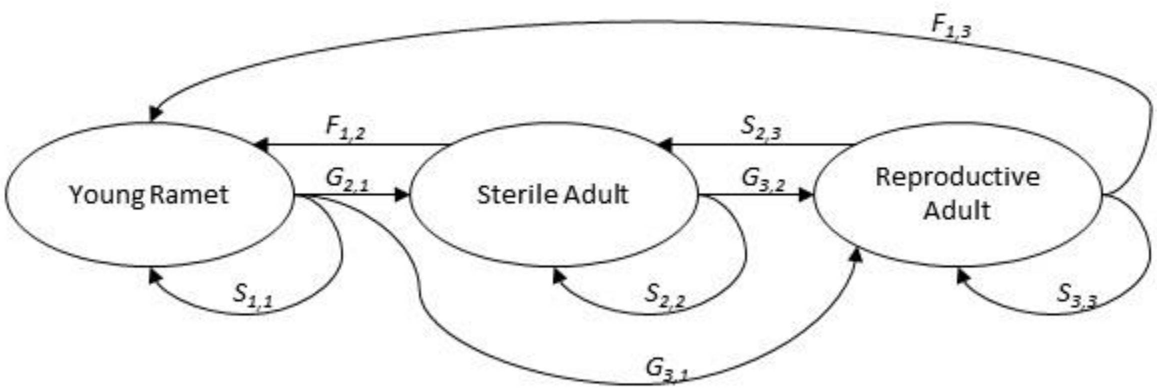

Figure 5. Generalized life-stage cycle model. Each arrow corresponds to the probability of a transition between stages during a transitional period. Variables correspond to those listed in the example of the transitional matrix (Table 6)

Table 7. Initial abundances per life stage used in 50 year DLI projection. Each initial abundance total relates to average population estimates for DLI populations ranked as A, B, C, and D using the NatureServe rubric (Table 2). The mean 7% of the total was assigned to Young Ramet stage; the proportion assigned to Reproductive Adult depended on the mode categorical abundance of the Reproductive Adults in surveyed ranked populations; and the remainder was assigned as Sterile Adult.

Life Stages	Initial Abundance A	Initial Abundance B	Initial Abundance C	Initial Abundance D
Young Ramet	24,833	3,532	189	125
Sterile Adult	294,442	34,311	1,836	1,624
Reproductive Adult	35,475	12,614	675	36
Total	354,750	50,457	2,700	1,785

Results

Element Occurrence records of DLI in Michigan

Michigan Natural Heritage Database contained 85 element occurrence (EO) records of DLI at the start of this project (Michigan Natural Features Inventory, 2019; Table 1). Recent digital data mining and survey efforts have increased number of Michigan records from 85 to 91. Digital data mining efforts in 2019 compiled a total of 593 unique records of DLI, 267 from Michigan, and 23 with unknown locations. Only 66 of the Michigan records were georeferenced and 25 records had obscured coordinates. Documentation was added to EO records from the compilation if the occurrence location could be reliably assessed. Records were also updated with documentation found in literature search. Forty-two existing records were updated with information found with the data mining effort.

The compilation yielded six new EO records: two on St. Martin Island in Grand Traverse Islands of Delta County, Little Summer Island in Lake Superior State Forest, southwest coast of Hog Island near Beaver Island, a population at the Duncan Bay Preserve in Cheboygan, and population on St. Martin Point in Hiawatha National Forest. The two EO records on St. Martin Island were based on literature and flora published in *The Great Lakes Botanist* (previously *The Michigan Botanist*) and follow-up with Green Bay National Wildlife Refuge (Judziewicz, 2001; Judziewicz et al., 2016). The Little Summer Island record was based on a specimen collected in 1998 and deposited in University of Wisconsin-Madison, Wisconsin State Herbarium (WIS, v 0207925 WIS). The Hog Island record was based on the thesis of Van Kley (1989), and a specimen collected in 2000 and deposited in the Central Michigan University Herbarium (CMC, CMC00004124).

From January 11, 2019 to June 28, 2021, the iNaturalist project compiled 156 observations of DLI in Michigan across 50 observers. The population in the Duncan Bay Nature Preserve was documented with an iNaturalist observation by Derek Shiels, botanist and Director of Stewardship at the Little Traverse Conservancy, which owns the property (https://www.inaturalist.org/observations/14521555). The population on St. Martin Point was documented with an iNaturalist observation by Robert Kahl, retired USFWS biologist (https://www.inaturalist.org/observations/6517901), although the uncertainty of the coordinates prevented a visit until a cross-reference in Hiawatha National Forest Current EO feature layer maintained by Hiawatha National Forest was made.

Two additional EO records were created during the time of the project unrelated to the digital data mining effort. One was due to landowner contact efforts that resulted in supervised access to Lafarge Holcim/Presque Isle Quarry in 2019. The quarry intersects with polygons of several existing EO records. The other record was the result of a Threatened/Endangered Species Report submitted to the Michigan Department of Natural Resources by a private consulting company in winter 2020.

The current total of EO records for extant Michigan DLI populations is 91, soon to be 89 (Table 1). After spatial surveys were conducted for this project, four pairs of EO records lacked the separation distance of 1000 m or unsuitable habitat between them to be considered separate populations. Two pairs of EO records have been merged into two EO records (EO 11586, EO 12942) due to spatial overlap and expert opinion of EO documentation prior to surveys conducted for this Agreement. The spatial survey for the other two pairs were conducted in

2021 and have not been merged as of the submission of this report. These EO records are marked with footnotes in Table 1.

Survey results

A total of 58 DLI EO records were visited from 2019 to June 2021 (Table 1; Hackett et al., 2021). The dynamic AGOL dataset can be found here for stakeholders that requested their AGOL username to be added to the group: https://arcg.is/145TnW. Spatial surveys were conducted at 39 EO records, qualitative surveys at 50 EO records, count surveys at 8 EO records[12]; and demographic surveys at 3 sites. Among count and demographic sites, 201 quadrat survey events were recorded. Due to high water levels of the Great Lakes in 2019 and 2020, four of the demographic quadrats placed in 2019 were lost. Two demographic quadrats were inundated in 2020 (Figure 6). Inundated quadrats had marker loss, and we were unable to secure new markers in place. Demographic quadrats took an estimated two to four times longer than count survey quadrats depending on DLI density, condition of the quadrat beyond the initial year, and whether new ramets were to be remarked for future survey.

Seventeen EO records underwent rank changes as a result of surveys from 2019 to June 2021 (Table 1). Four records were changed from extant (E) to a valued category (A – D). Two records were changed from E to failed to find (F). Six records were changed from a A to D ranking to a F ranking, although one of these appears to be due to bad coordinates and actually referring to a population within the separation distance of another EO record (EO 2235 >> EO 10080). One record was changed from historical (H) to F (EO 11844). Two records increased in rank from C to B due to new populations counts (EO 835, 8623). One record increased in rank from BC to B due to additional colonies found on Michigan Nature Association property (EO 8385). One record decreased from BC to C due to reduction of population due to high Lake Michigan water levels of 2019 and 2020 and successional shading in remaining occupied areas (EO 7130).

Count-based population viability analysis

The combination of 2019 – 2021 count and demographic survey data and count data available in literature was able to produce 36 population change increments across 17 [sub]populations (Table 8). Using this data, μ was derived to be -0.0657 with σ^2 of 0.114 (Figure 7).

For the simulations, we rounded the maximum estimated population across all counts as K to 500,000,000 ramets and the median estimated population across all counts as the starting population to 7,000,000. Of the 1000 simulations, 14.2% of the simulations went extinct before reaching 50 years (Figure 8). Confidence intervals ranged extensively after 10 years.

The results of the CDF illustrated likely extinction for populations with less than 5000 ramets within 25 years (Figure 9). Populations with 25,000 ramets or more were likely to persist for at least 50 years.

[12] Restrictions in place during the 2020 pandemic reduced the time available to conduct DLI surveys during the flowering period and only 6 sites were visited for count surveys that year.

a) b) c)

Figure 6. Dwarf lake iris ramets persisting underwater during flooding event at EO 12376 Mackinac County, Michigan, on June 10, 2020: a) the red flag of a marked demographic quadrat in knee-deep water, b) flower, c) ramets and buds. Photographs by Rachel Hackett.

Table 8. Mean density of DLI ramets in units of 1 m^2 by year of counts. Unless otherwise stated, data gathered from 2019 to 2021 surveys conducted by MNFI and source feature (SF) polygons derived from survey efforts and mapped in Michigan Natural Heritage Database (Michigan Natural Features Inventory, 2021). In Natural Heritage Database EO ID refers to unique identifier for a population record and SF ID refers to unique identifier of polygon shape mapped. See Table 1 for more information on DLI EO records.

EO ID	SF ID	Area (m^2)	1987	1988	1989	1990	1991	1992	1993	1994	1995	1996	1997	2019	2020	2021
1369	11815	6096[13]	472[13]	378[13]												
3403	66099	60,251												141	180	140
3403	66104	20,395	1 population change increment											145	142	124
4466	66716	24,864												287		261
7130	62359	31,216														
7130	66703	85,349		201[14]										162	98	199
8162	15674	1,399,573												287	267	270
8439	25454	71,279		275[14]										152	180	161
8439	25455	26,544												164	234	380
8439	66299	70,565												225	220	113
8964	27160	163,989												720		216
8964	27162	273,861												307		297
8964	27166	31,029												132		73
8964	66706	11,416												540		228
11321	25462	132[15]			55[15]	49[15]	46[15]	42[15]	54[15]	61[15]	45[15]	41[15]	35[15]	0.2		
12376	66711	137,500		292[14]										168	167	163
15125	66717	2359												315	367	281

[13] Count data and occupied area derived from Van Kley, 1989
[14] Count data derived from Van Kley, 1989
[15] Count data and occupied area derived from Ewert and Scrimger, 1989; Ballard and Lauffer, 1993; Ballard and Kowal, 1997

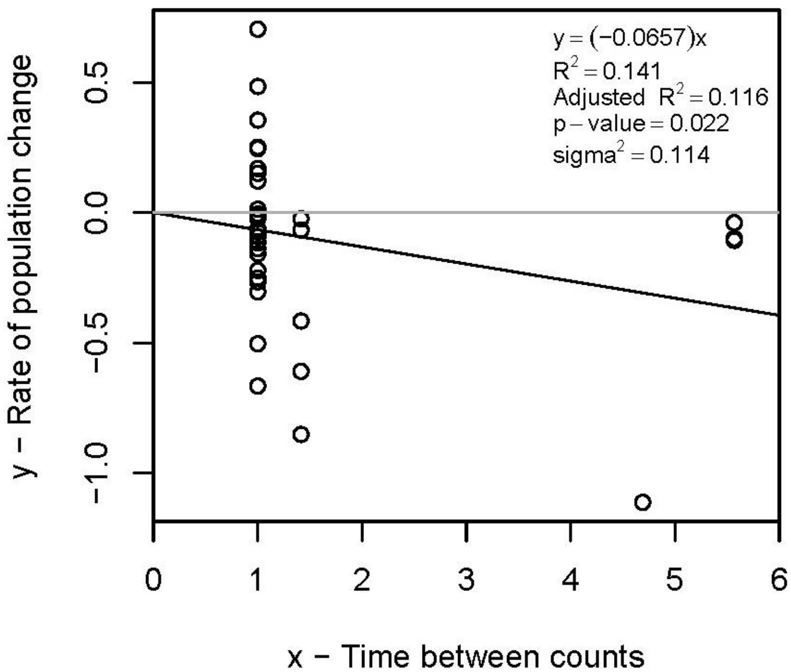

Figure 7. Linear regression of *y* on *x* for DLI count data , where *y* is the natural log transformation of population change between two consecutive counts in a population or subpopulation, and *x* is a transformation of time between those two consecutive counts (equations in text).The slope is an estimate of μ and the variance of the residuals (σ^2) are used for the population viability analysis.

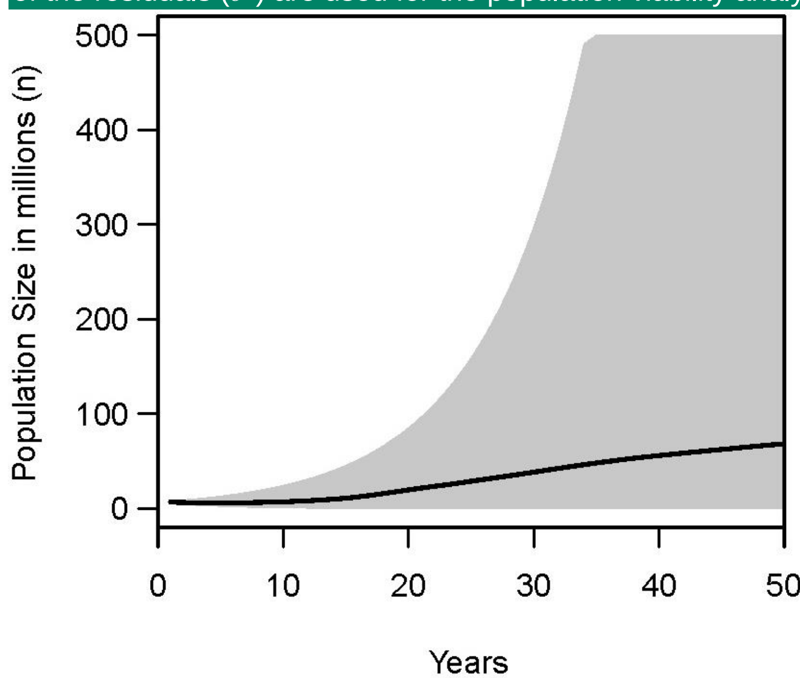

Figure 8. Mean population (black line) and 95% confidence intervals (grey area) of 1,000 iterations of the population simulation to 50 years.

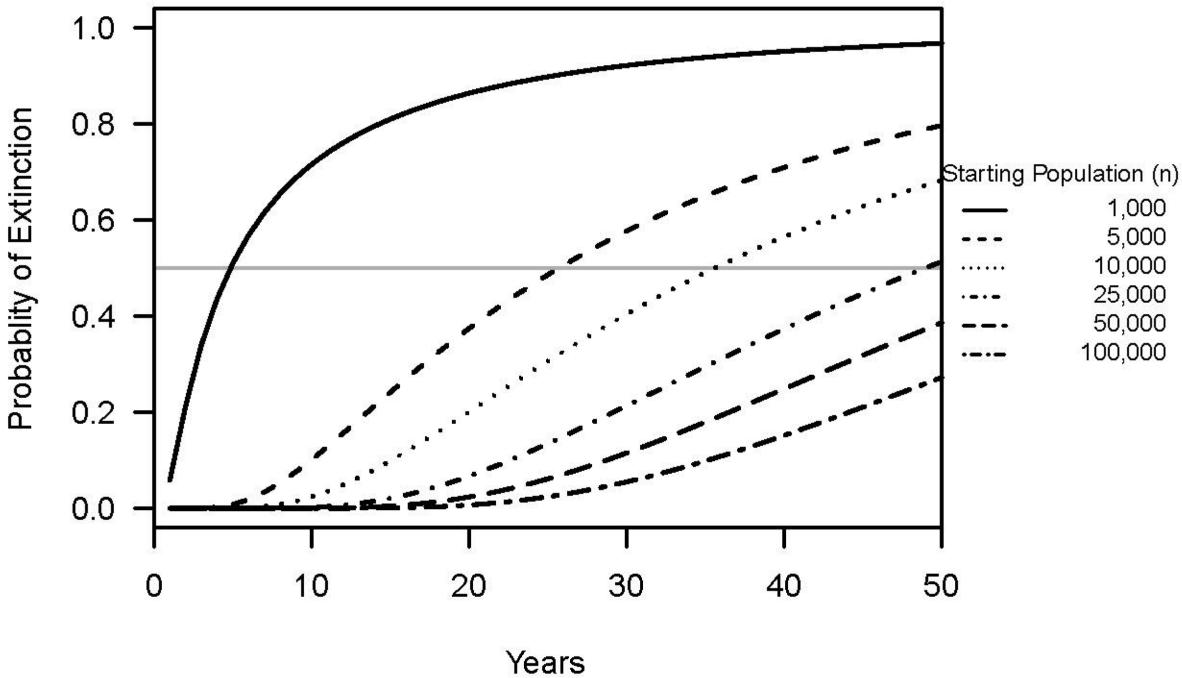

Figure 9. Cumulative distribution function (CDF) for the probability of extinction for DLI at various starting populations (Morris and Doak, 2002). Each style of black line represents the probability of extinction for a population of starting populations listed in the legend. The 0.50 probability is marked with a grey line.

Demographic-based population viability analysis

There were 29 year-to-year transitions of demographic quadrats across three DLI populations from 2019 to 2020. One demographic quadrat was documented only from 2020 to 2021. Four demographic quadrats were lost from 2019 to 2020 (Figure 10), and one quadrat was inundated in 2020 and unable to be properly recorded (Figure 6a).

The λ recorded at each location-year were almost all greater than 1 (Table 9). The greatest number of missing or unreadable markers in the subsequent year was 71, with a mean of 14 (SD 19). The quadrat-transitions with the greatest missing or unreadable markers were those quadrats lost during a year of high water level for the Great Lakes. All except 7 transitions had more growth than mortality. The life-stage counts and life-stage transitional matrices for each quadrat-transition can be found in Appendix A.

When all quadrat-transitional matrices were pooled, the resulting life stage transitional matrix had positive overall growth (λ = 1.04), and the Sterile Adult stage was a relatively stable stage (Figure 11). The stable stage distribution for each life stage was 0.092, 0.83, and 0.076 for Young Ramet, Sterile Adult, and Reproductive Adult, respectively, and. The species was most sensitive to disturbances or threats that cause decline of Sterile Adults. All life stages had approximately the same mean probability of mortality (0.44, 0.30, 0.39, respectively).

For the simulations based on transitional matrices at the location-year level, the mean populations of each rank increased overall (Figure 13). Confidence intervals ranged extensively after 10 years. The simulation using abundances derived from rank A/AB did not have any simulations that reached extinction or quasi-extinction (Figure 12a; Figure 12b; Figure 13);

minimum population ever reached was 2447 ramets. Although no simulation of the rank B/BC, C/CD, or D populations reached 0 ramets, 0.1%, 13.6%, and 22.1% of simulations, respectively, reached below the set quasi-extinction threshold (500) before year 50 (Figure 13).

Table 9. Transition summary of marked DLI ramets. EO referred to the Element Occurrence number assigned to population in Michigan's Natural Heritage Database (Table 1). Marked ramets 'Missing 2nd year' were missing or unreadable markers. 'Survivorship' was the number of marked ramets not classified as "Dead" next year. New growth were ramets without a marker in the 2nd year. Mortality was the number of markers without a corresponding ramet in the 2nd year. Constant rate of population growth is λ. Instantaneous growth rate is r

EO	Quadrat	Years	Marked in both years	Missing 2nd year	Survi-vorship	New Growth	Mortality	λ	r
7130	1	2019-2020	43	2	38	22	5	1.44	0.36
7130	1	2020-2021	62	3	41	10	21	1.26	0.23
7130	3	2019-2020	26	1	23	10	3	1.33	0.29
7130	3	2020-2021	36	0	11	9	25	1.25	0.22
7130	4	2019-2020	19	1	17	13	2	1.65	0.50
7130	4	2020-2021	30	3	17	8	13	1.21	0.19
7130	5	2019-2020	17	7	15	32	2	2.08	0.73
7130	5	2020-2021	35	15	27	32	8	1.36	0.31
7130	6	2019-2020	-	17	0	0	17	0.00	0.00
7130	8	2019-2020	-	68	0	0	68	0.00	0.00
7130	10	2019-2020	-	71	0	0	71	0.00	0.00
7130	13	2020-2021	58	20	28	5	30	0.88	-0.13
12376	1	2019-2020	7	10	6	0	1	0.41	-0.89
12376	1	2020-2021	5	2	4	5	1	1.43	0.36
12376	2	2019-2020	-	38	0	0	38	0.00	0.00
12376	3	2019-2020	42	0	38	11	4	1.26	0.23
12376	3	2020-2021	52	1	40	13	12	1.23	0.21
12376	5	2019-2020	83	8	78	32	5	1.29	0.25
12376	5	2020-2021	68	49	39	8	29	1.08	0.08
12376	6	2019-2021	32	0	30	12	2	1.38	0.32
12376	6	2020-2021	44	0	35	10	9	1.25	0.22
15125	1	2019-2020	95	20	80	11	15	0.92	-0.08
15125	1	2020-2021	100	6	71	50	29	1.52	0.42
15125	4	2019-2020	15	4	14	7	1	1.16	0.15
15125	4	2020-2021	17	5	16	18	1	1.68	0.52
15125	5	2019-2020	46	7	40	15	6	1.15	0.14
15125	5	2020-2021	50	11	40	23	10	1.23	0.21
15125	7	2019-2020	30	18	29	46	1	1.58	0.46
15125	7	2020-2021	59	17	52	39	7	1.34	0.29

a)

b)

c)

d)

e)

f)

Figure 10. Photographs from selected lost quadrats in 2020: a) Flag from a quadrat in EO 12376, Mackinac County, while b) unmarked DLI ramets were found near GPS coordinates; c) - e) unmarked ramets near GPS coordinates for quadrats in EO 7130, Emmet County. In f) no ramets were found near GPS coordinates, so quadrat was likely lost. Photographs by Rachel Hackett

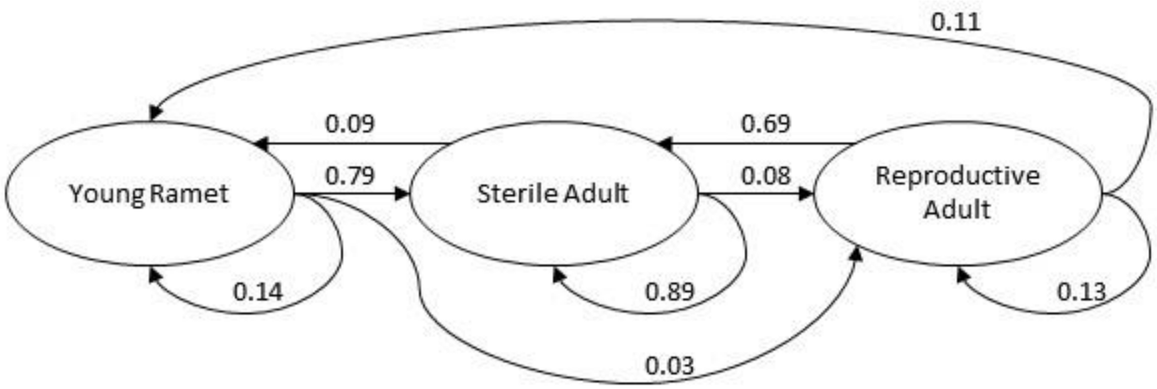

Figure 11. Illustrated life stage cycle of the pooled life stage transitional matrix. The numbers are the probability that a ramet will transition between life stages as indicated by the arrows.

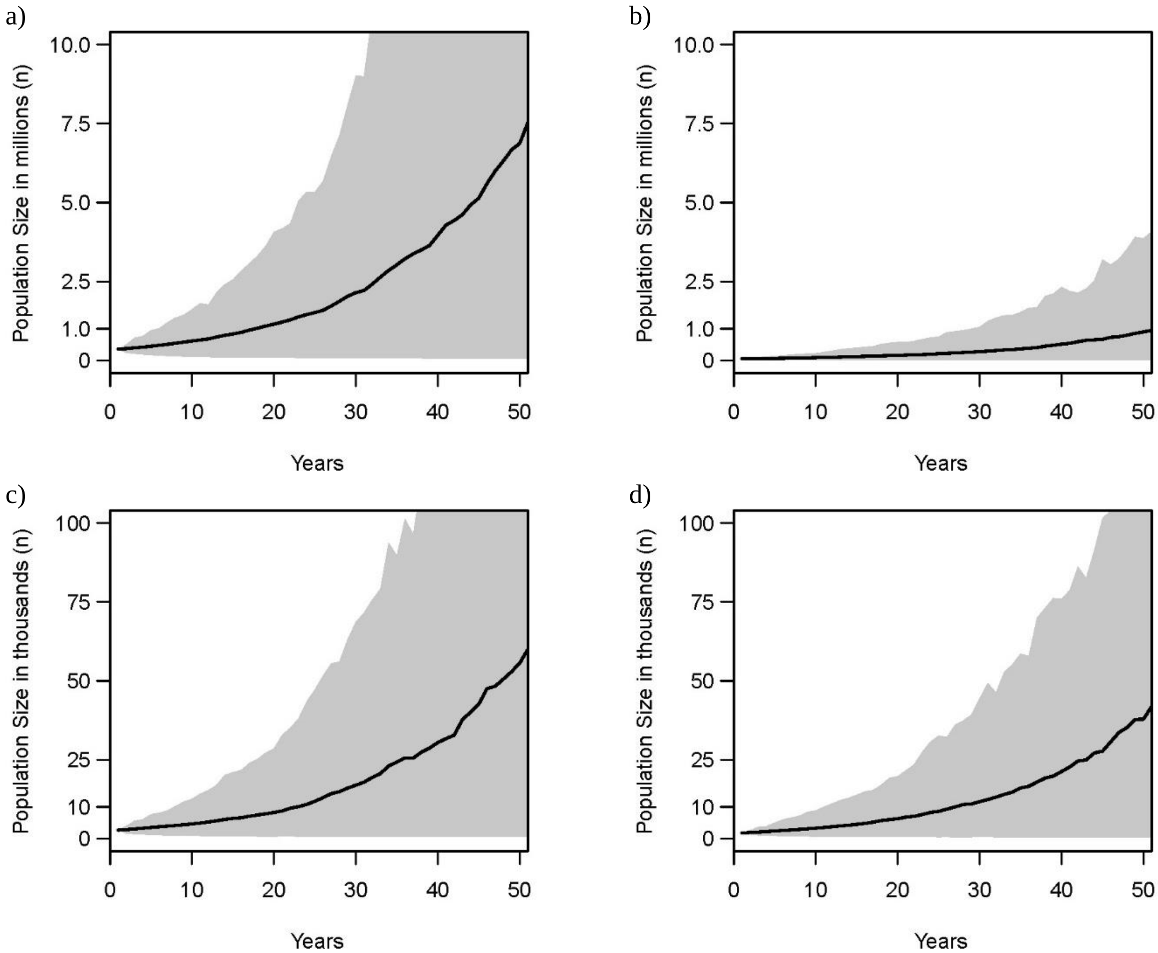

Figure 12. DLI population projections to 50 years using demographic life stage transitional data and different initial abundances based on rank with 1,000 iterations: a) initial abundance for populations of A rank, b) B rank, c) C rank, and d) D rank. Mean population (black line) and 95% confidence intervals (grey area). Note the change in magnitude of the units between a)/b) and c)/d) to improve visualization.

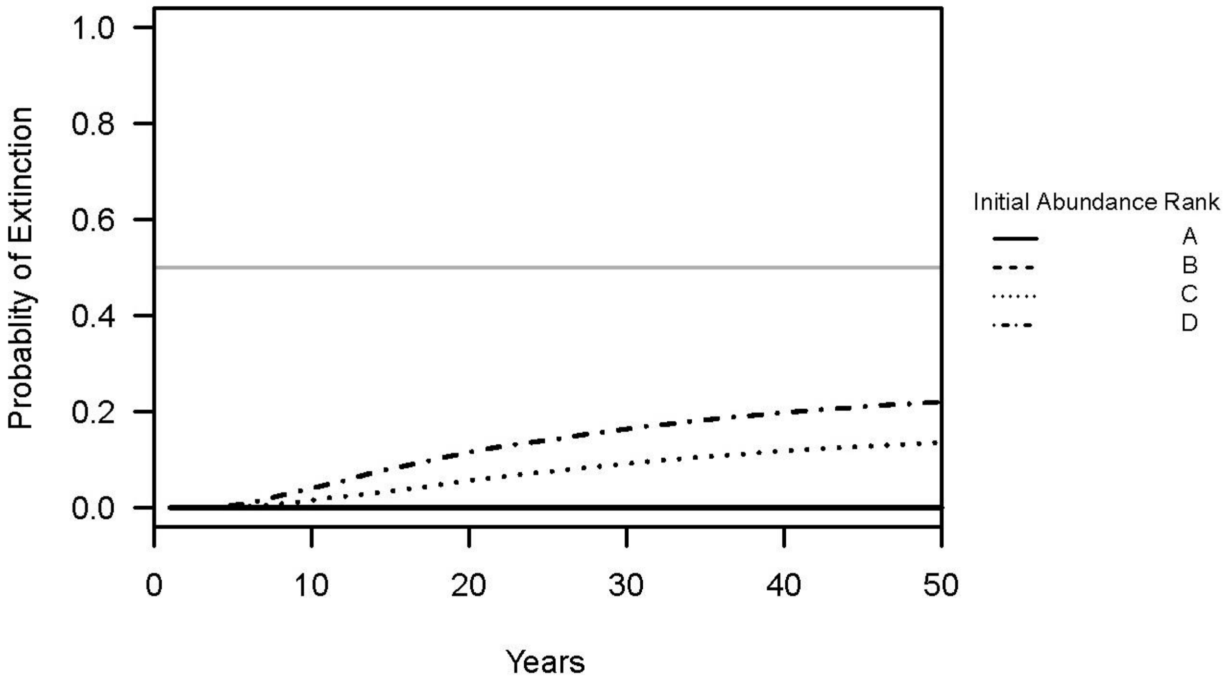

Figure 13. Extinction probabilities of populations with initial abundances based on EO Rank. Each style of black line represents the probability of extinction for a population of initial abundances listed in the legend (Table 6). The 0.50 probability is marked with a grey line.

Discussion

Population viability assessment outcomes and future needs
Among the count data intervals, the mean instantaneous stochastic growth rate (μ) for all sample sites was negative, but near zero with a large enough variance (σ^2) that could span positive values (Figure 7). This indicates that although the populations were likely decreased between counts, growth of the population was not outside of the realm of possibilities. This is further illustrated by the low extinction rate of 14.2% of the simulations. The growth rate determined by the demographic data (λ) fell within the range of variance determined by the count data in the positive growth direction. Large area and/or dense populations of dwarf lake iris are stable at their current and normal conditions, likely to persist several decades based on these analyses.

Unfortunately, the high variance also contributed to the large confidence intervals (Figure 8). This lowers our ability to rely on the population predictions the further out they are projected. To improve the predictions, more count data is needed. Fieberg and Ellner (2000) developed a general quantitative rule of thumb to determine the length of time a population analysis model can produce a reliable estimate:

$$t \leq \frac{n}{5} \; or \; t \leq \frac{n}{10}$$

where *t* is the time to project, *n* is the number of instantaneous stochastic growth rates that can be calculated. Given our n of 36, we could reliably project 4 to 7 years in the future. This rule of thumb is supported by the confidence intervals of the simulations which increase greatly at approximately year 10 (Figure 8). To reliably project 50 years in the future, we would need at least 250 to 500 N of count data intervals. If the number of [sub]populations remain constant from those we surveyed in 2019 to 2021, we would need 8 to 15 additional years of count survey data. Using the methods described here, years need not be consecutive. We could also increase the number of [sub]populations monitored to reduce the number of years needed.

The count-based PVA we used did not incorporate any catastrophic changes in the model (Elderd et al., 2003). The assumption of normally distributed population growth simplifies the model, but increases in weather extremes due to climate change could affect DLI populations. The exposed populations along the Great Lakes lakeshore are particularly vulnerable, which was observed by the missing demographic quadrats at the two lakeshore sites undergoing demographic surveys between 2019 and 2020. Conducting spatial surveys or measuring percent cover of ramets in occupied area at count sites each year could better account for such changes in count surveys. These actions would change the occupied area for each interval, but add considerable field and data analysis time.

From the demographic population analysis, disturbances or threats to the Sterile Adults life-stage would be most detrimental to the overall population. This stage had the highest growth probabilities both to and from this stage and accounted for the majority of the surviving ramets. Browsing was a common disturbance seen in this life stage, either by deer, rabbit, or slug, but our observations reported these threats affecting less than 50% of the population in most instances. Increases in these threats or the introduction of new threats at this life stage could drastically change DLI population survivorship.

Predicted extinction rates for DLI populations

The rank of eight EO records had moved to ‘failed to find’ in the last 10 to 55 years (Table 1). Two of the records were ranked B and BC, thus had a good probability of persisting beyond 20 years, according to experts at the time (EO 6713, EO 2472;Table 1; Table 2). Each of these populations were near either a roadside or in a developing residential area, except for the historical record in a State Park. These anthropogenic stressors may have contributed to their decline, but no data was collected or analyzed to support this. There are several seemingly stable populations of DLI on private, residential property that contradict the absoluteness of that observation (e.g., EO 3606, EO 8964). More research is needed into the possible positive, negative, or neutral effects of residential development on DLI populations. One EO had been ranked C in 1990 (EO 4640) and one H (EO 11844), so they were unlikely to persist in 2020. Two occurrences were recorded as E in the 1980’s and had underwent development since that survey (EO 743, EO 12375). One EO ranked B (EO 2235) was poorly georeferenced and likely a part of a large multi-polygon EO to the south (EO 5551/10080). We are unsure of what could have happened to the A ranked EO 2837 since 2002 to cause its decline.

The CDF predicted that DLI populations with 5,000 or less ramets had a 50% chance of extinction in 25 years (Figure 9). Of the 58 EO records visited, 9 populations and 12 subpopulations of multi-polygon populations had a minimum estimate of less than 5000 ramets (Michigan Natural Features Inventory, 2021). The 9 populations were ranked C or less, supporting the prediction of the CDF with a qualitative expert option (Table 2).

The extinction projections with the demographic analysis was less dire, with less than 25% chance of extinction of D ranked populations in 50 years at current growing conditions (Figure 13). The demographic analysis supports the idea that DLI populations are relatively stable in normal conditions barring a severe disturbance affecting the entirety or Sterile Adult portion of the population.

The level at which the transitional data was pooled (i.e., location-year) for analysis made a difference with extinction results. When the transitional matrix data was examined at a finer scale (i.e., quadrat-year), the variability of the yearly changes would result in approximately 50% probability of extinction in projections from all initial abundances by year 50 (not shown). This difference of extinction rates between scale supports the idea that changes among the population are heterogeneous: increasing in some areas and decreasing in others, not homogeneous throughout the population. Under normal conditions for large populations, DLI will likely persist in some capacity at the site, although finer-scale examinations may show more detrimental effects.

There are flaws with using DLI ramet estimates in the count-based analysis including 1) cognitive limitations for visualizing large numbers, 2) variation among surveyors, 3) hidden occupied areas of DLI not used when estimating total occupied area, and 4) overestimation without percent cover of occupied areas figured into estimates based on density. We attempted to preemptively counteract the first two items with standardized training including components to help with visualization. Seasonal technicians in 2020 were trained together. One of the exercises was to estimate the number of ramets in a set area (0.25 m^2), then compare it with an actual count. This allowed technicians to associate certain numbers with similar visual densities. Use of items as size reference (e.g., rulers, a standard piece of paper) were required for use when taking density photographs downward on a colony. This allowed technicians to maintain a relative density visual in their head regardless of whether they were conducting a qualitative or count survey.

Within a count survey, the DLI occupied area could be underestimated, especially if a spatial survey was not required for an EO record. Only the most dense or obvious areas may have been considered when making the population estimates. This issue reinforces the importance of spatial surveys and accurate mapping of DLI populations.

There is also the possibility of overestimating of ramets because of the way we used DLI density and occupied area to estimate the total number of ramets in a population. Although varying DLI densities are taken into consideration by using the mean density of quadrats within the same [sub]population, DLI rarely covered every meter of the mapped occupied area. One method of correction could be to measure or estimate a percent cover of occupied area with ramets, but this has its own faults with consistency, difficulty to visualize large populations, and time constraints.

Life-stage based demographic analysis is not without flaws including 1) not identifying the life stage the new growth likely originated from, and 2) the time range of the survey not adequately representing the long-life of the species. The assumption was made that the New Growth of a year could be attributed proportionally to having originated from any of the three life-stages, which it could have favored one over the other. More detailed examinations including unearthing of rhizomes and more frequent visits would be needed to determine the true proportion for the new vegetative growth. An earlier spring visit would be required to identify seedlings from other growth, which would be attributed only to the Reproductive Adults of the previous year. Our visits were too late to identify the seedlings with the 'hooked' leaf, which had wilted since emerging (Brotske, 2018). Since DLI is a perennial species, they can live several years. Three years is unlikely long enough to observe a ramet for its entire life, which can pose complications to the projections of the information far into the future (Brigham and Thomson, 2003).

Conservation of DLI

To protect, conserve, and manage DLI populations, examination of the species relative to its redundancy, resiliency, and representation is needed (US Fish and Wildlife Service, 2016). The best action for protecting DLI is protecting lands of existing populations. Shoreline development is suspected to be a leading cause in DLI loss, but there has not been definitive research in this area for this species.

High priority DLI population should have representation across the following characteristics: inland and shoreline populations, flower color (e.g., rare white flowers), natural community/habitat, large area, and range-wide geography. The few populations associated with inland populations, white flowers, and rare community types (e.g., alvar) were prioritized regardless of rank, area, or geography (see bolded records in Table 1).

The variety of identified natural communities within a DLI population and the types of the natural communities should both be taken into consideration for prioritization to improve not only representation but resiliency of the species. Each of these communities has different natural processes that affect rates of succession and each face different levels of threats and disturbance. The demographic analysis among two populations with lakeshore and wooded dune and swale complex (EO 7130; EO 12376) and one inland population in boreal and mesic forest (EO 15125) supported the different threat level: The quadrat-year transitions that had the most negative effects were those most affected by flooding in 2020 in lakeshore or wooded dune and swale natural communities: both lost and inundated quadrats had high mortality. Quadrats from the inland site in boreal and mesic northern forest showed more stability and growth on the whole with greater transitional values at the pooled location-year level (Appendix A: Table 12). These events further support the vulnerability of DLI populations to the increase of extreme weather and climate changes like changes in Great Lakes water levels.

The hydrology and substrate of communities such as fens and glades maintain a slower rate of canopy closure than that of swamps and forests. Since several studies have concluded that sexual reproduction is lower in shaded populations than in unshaded sites (Van Kley, 1989; Brotske, 2018), the open nature of fen and glade communities likely facilitates the stability and persistence of DLI populations. DLI population in these communities may also be the easiest to maintain by allowing for and/or imitating the natural processes of the communities themselves. Prioritizing DLI in these communities would minimize the needs for human management and maximize the long-term persistence of the species.

Communities that rely more on natural disturbance to maintain open canopy like sand and gravel beaches, boreal forests, and Great Lakes barrens may match fens and glades in the ability to maintain open canopy or gaps in the canopy for DLI, but the disturbance may directly affect the stability of the populations. For example, populations on sand and gravel beaches benefit from a partially open canopy due to the Lake processes, but those processes can bury or wash away DLI populations in those areas. This project documented this instability with the loss of 'permanent' demographic quadrats from 2019 to 2021.

Although DLI reproduction has been shown to be effected by light and litter levels (Van Kley, 1989; Brotske, 2018), the benefits of management by simulating canopy gaps and increasing light levels have not been examined outright. There is a balance between light, moisture, and conifers that may not be able to be artificially replicated for the benefit of DLI populations. More experimental management research is needed to track possible effects, preferably five to ten years in duration given the response lag of most plant species.

DLI populations also face growing threats of invasive species, mostly plants, although herbivory from slugs was noticed affecting up to 50% of the plants in many populations. In most natural communities DLI preferentially occurs in habitat where few other plants thrive: rocky areas with a shallow organic layer. Shading by encroaching invasive shrubs and taller grasses is likely the greatest threats posed by invasive species.

Invasive plants in DLI occupied areas seem to follow regional and natural community type patterns. Spotted knapweed (*Centaurea stoebe*) and invasive hawkweed (e.g., *Hieracium caespitosum, Hieracium piloselloides*) in sand and gravel beaches, glades, Great Lakes barrens, and wooded dune and swale complexes; reed canary grass (*Phalaris arundinacea*) in fens and some shorelines. In drier habitats, invasive shrubs such as invasive honeysuckle (e.g., *Lonicera morrowii, Lonicera x bella*), multiflora rose (*Rosa multiflora*), and autumn olive (*Elaeagnus umbellata*) are encroaching. While current direct encroachment by these species was minimal, these species spread quickly as evidenced statewide and as documented in our surveys of Pitcher's thistle (*Cirsium pitcheri*) occurrence sites (Slaughter and Cuthrell, 2015). Early detection and removal is likely to be the most cost-effective way to minimize this threat.

A few regions had very specific invasive species not seen elsewhere. In Emmet County, wall lettuce (*Mycelis muralis*) was persistent in forested habitats and increased in abundance from 2019 to 2021 (EO 7130, EO 3606). This species has been recently documented growing on limestone boulders that support the state threatened walking fern (*Asplenium rhyzophyllum*) in the Upper Peninsula. At a limestone glade in Presque Isle, common barberry (*Berberis vulgaris*) was becoming an increasing problem (EO 8162). In Delta and Schoolcraft Counties, two herbaceous invasive species were observed increasing in abundance from 2019 to 2021, but were not yet growing within the same area and habitat as DLI: helleborine (*Epipactuis helleborine*) and hound's-tongue (*Cynoglossum officinale*). These species should be monitored for possible invasion into DLI populations.

DLI is an endemic species to the Great Lakes with its greatest stronghold in Michigan. Efforts should be made to continue documenting the status of remaining DLI populations to better predict its persistence. The PVA conducted for this project indicated a general small decline in population growth for the species, but the variance of the growth rate throws uncertainty onto any sweeping statements about its growth, stability, or decline (Figure 7, Figure 8). As more reliable population data is collected, efforts should be made to prioritize, conserve, and protect known DLI populations. Populations categorized as high-priority or fitting into the characteristics described as such, should be targeted for focused attention. Efforts should be made to protect more of the surrounding land from development and other anthropogenic disturbances and to manage encroaching woody and herbaceous invasive species. Efforts to better understand possible management effects, specifically canopy manipulation, on DLI populations is needed to better advise landowners on how to promote the species fitness and persistence. In addition, genetic studies should be undertaken to help further refine the selection of priority sites and shed more light on sexual and vegetation reproduction of DLI.

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Appendix A: Demographic survey data

Demographic survey data at the quadrat-transition year level.

Table 10. Life stage counts to develop transitional matrices at the quadrat-transitional year level. Columns are life stage in the first year; rows are the life stage in the second year.

EO 7130 - quadrat 1, 2019-2020	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	4	1	0	0
Sterile Adult	18	6	31	0
Reproductive Adult	0	0	0	0
Dead	0	1	4	0

EO 7130 - quadrat 1, 2020-2021	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0	0	1	0
Sterile Adult	10	2	38	0
Reproductive Adult	0	0	0	0
Dead	0	3	14	0

EO 7130 - quadrat 3, 2019-2020	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	2	0	0	0
Sterile Adult	8	4	17	1
Reproductive Adult	0	0	1	0
Dead	0	0	3	0

EO 7130 - quadrat 3, 2020-2021	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	2	0	3	0
Sterile Adult	7	1	7	0
Reproductive Adult	0	0	0	0
Dead	0	1	20	1

EO 7130 - quadrat 4, 2019-2020	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	3	0	0	0
Sterile Adult	10	1	15	0
Reproductive Adult	0	0	1	0
Dead	0	0	2	0

EO 7130 - quadrat 4, 2020-2021	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	2	0	2	0
Sterile Adult	6	1	12	1
Reproductive Adult	0	0	1	0
Dead	0	2	11	0

EO 7130 - quadrat 5, 2019-2020	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	5	0	0	0
Sterile Adult	25	2	8	1
Reproductive Adult	2	0	4	0
Dead	0	0	2	0

EO 7130 - quadrat 5, 2020-2021	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	3	0	0	0
Sterile Adult	24	4	21	1
Reproductive Adult	5	0	1	1
Dead	0	0	6	1

EO 7130 - quadrat 6, 2019-2020	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0	0	0	0
Sterile Adult	0	0	0	0
Reproductive Adult	0	0	0	0
Dead	0	6	9	2

EO 7130 - quadrat 8, 2019-2020	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0	0	0	0
Sterile Adult	0	0	0	0
Reproductive Adult	0	0	0	0
Dead	0	10	49	9

EO 7130 - quadrat 10, 2019-2020	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0	0	0	0
Sterile Adult	0	0	0	0
Reproductive Adult	0	0	0	0
Dead	0	15	42	14

EO 7130 - quadrat 13, 2020-2021	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	3	0	2	0
Sterile Adult	2	7	23	1
Reproductive Adult	0	0	0	0
Dead	0	1	18	6

EO 12376 - quadrat 1, 2019-2020	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0	0	0	0
Sterile Adult	0	0	6	0
Reproductive Adult	0	0	0	0
Dead	0	0	1	0

EO 12376 - quadrat 1, 2020-2021	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	5	0	4	0
Sterile Adult	0	0	0	0
Reproductive Adult	0	0	0	0
Dead	0	0	0	0

EO 12376 - quadrat 2, 2019-2020	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0	0	0	0
Sterile Adult	0	0	0	0
Reproductive Adult	0	0	0	0
Dead	0	0	38	0

EO 12376 - quadrat 3, 2019-2020	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	3	0	1	1
Sterile Adult	8	0	27	6
Reproductive Adult	0	0	3	0
Dead	0	0	4	0

EO 12376 - quadrat 3, 2020-2021	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0	0	5	1
Sterile Adult	13	3	29	1
Reproductive Adult	0	0	0	0
Dead	0	2	7	1

EO 12376 - quadrat 5, 2019-2020	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	5	0	0	0
Sterile Adult	26	0	41	13
Reproductive Adult	1	0	16	9
Dead	0	0	4	0

EO 12376 - quadrat 5, 2019-2020	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0	0	0	1
Sterile Adult	0	3	34	6
Reproductive Adult	8	0	4	2
Dead	0	2	10	3

EO 12376 - quadrat 6, 2019-2020	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0	0	0	1
Sterile Adult	12	0	23	0
Reproductive Adult	0	0	6	0
Dead	0	0	2	0

EO 12376 -quadrat 6, 2020-2021	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	3	0	1	0
Sterile Adult	7	1	24	4
Reproductive Adult	0	0	5	0
Dead	0	0	5	2

EO 15125 - quadrat 1, 2019-2020	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0	0	1	0
Sterile Adult	11	0	75	2
Reproductive Adult	0	0	2	0
Dead	0	0	15	0

EO 15125 - quadrat 1, 2020-2021	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	4	0	1	0
Sterile Adult	43	0	62	1
Reproductive Adult	3	0	6	0
Dead	0	1	19	1

EO 15125 - quadrat 4, 2019-2020	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	4	0	0	0
Sterile Adult	3	0	10	4
Reproductive Adult	0	0	0	0
Dead	0	0	0	1

EO 15125 - quadrat 4, 2020-2021	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	2	2	0	0
Sterile Adult	15	2	12	0
Reproductive Adult	1	0	1	0
Dead	0	0	0	0

EO 15125 - quadrat 5, 2019-2020	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	1	0	0	0
Sterile Adult	14	0	38	1
Reproductive Adult	0	0	1	0
Dead	0	0	6	0

EO 15125 - quadrat 5, 2020-2021	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	5	0	2	0
Sterile Adult	18	1	32	1
Reproductive Adult	0	0	5	0
Dead	0	0	7	0

EO 15125 - quadrat 7, 2019-2020	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	15	0	0	0
Sterile Adult	31	3	15	6
Reproductive Adult	0	0	5	0
Dead	0	0	1	0

EO 15125 - quadrat 7, 2020-2021	New Growth	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	9	3	6	0
Sterile Adult	30	9	29	1
Reproductive Adult	0	1	4	0
Dead	0	1	4	1

Table 11. Transitional matrices at the quadrat-transitional year level. Columns are life stage in the first year; rows are the life stage in the second year.

EO 7130 - quadrat 1, 2019-2020	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.22	0.09	0.00
Sterile Adult	1.17	1.31	0.00
Reproductive Adult	0.00	0.00	0.00

EO 7130 - quadrat 1, 2020-2021	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.00	0.02	0.00
Sterile Adult	0.50	0.90	0.00
Reproductive Adult	0.00	0.00	0.00

EO 7130 - quadrat 3, 2019-2020	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.09	0.07	0.09
Sterile Adult	1.35	1.11	1.35
Reproductive Adult	0.00	0.05	0.00

EO 7130 - quadrat 3, 2020-2021	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.09	0.16	0.00
Sterile Adult	0.82	0.45	0.00
Reproductive Adult	0.00	0.00	0.00

EO 7130 - quadrat 4, 2019-2020	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.18	0.16	0.00
Sterile Adult	1.59	1.36	0.00
Reproductive Adult	0.00	0.06	0.00

EO 7130 - quadrat 4, 2020-2021	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.04	0.14	0.12
Sterile Adult	0.45	0.66	1.35
Reproductive Adult	0.00	0.04	0.00

EO 7130 - quadrat 5, 2019-2020	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.34	0.29	0.33
Sterile Adult	2.66	2.00	2.67
Reproductive Adult	0.14	0.40	0.13

EO 7130 - quadrat 5, 2020-2021	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.11	0.09	0.07
Sterile Adult	1.89	1.45	0.93
Reproductive Adult	0.18	0.18	0.46

EO 7130 - quadrat 13, 2020-2021	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.09	0.11	0.02
Sterile Adult	0.94	0.58	0.15
Reproductive Adult	0.00	0.00	0.00

EO 12376 - quadrat 1, 2019-2020	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.00	0.00	0.00
Sterile Adult	0.00	0.86	0.00
Reproductive Adult	0.00	0.00	0.00

EO 12376 - quadrat 1, 2020-2021	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.00	2.25	0.00
Sterile Adult	0.00	0.00	0.00
Reproductive Adult	0.00	0.00	0.00

EO 12376 - quadrat 3, 2019-2020	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.00	0.10	0.22
Sterile Adult	0.00	0.96	1.07
Reproductive Adult	0.00	0.09	0.00

EO 12376 - quadrat 3, 2020-2021	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.00	0.12	0.33
Sterile Adult	0.80	0.98	0.55
Reproductive Adult	0.00	0.00	0.00

EO 12376 - quadrat 5, 2019-2020	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.00	0.06	0.06
Sterile Adult	0.00	0.98	0.92
Reproductive Adult	0.00	0.27	0.42

EO 12376 - quadrat 5, 2020-2021	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.00	0.00	0.08
Sterile Adult	0.60	0.71	0.50
Reproductive Adult	0.12	0.25	0.32

EO 12376 - quadrat 6, 2019-2020	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.00	0.00	1.00
Sterile Adult	0.00	1.12	0.40
Reproductive Adult	0.00	0.19	0.00

EO 12376 -quadrat 6, 2020-2021	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.09	0.10	0.06
Sterile Adult	1.20	0.86	0.80
Reproductive Adult	0.00	0.14	0.00

EO 15125 - quadrat 1, 2019-2020	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.00	0.01	0.00
Sterile Adult	0.00	0.92	1.14
Reproductive Adult	0.00	0.02	0.00

EO 15125 - quadrat 1, 2020-2021	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.00	0.06	0.03
Sterile Adult	0.00	1.18	0.80
Reproductive Adult	0.00	0.10	0.02

EO 15125 - quadrat 4, 2019-2020	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.00	0.29	0.23
Sterile Adult	0.00	1.21	0.97
Reproductive Adult	0.00	0.00	0.00

EO 15125 - quadrat 4, 2020-2021	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.62	0.12	0.00
Sterile Adult	1.44	1.86	0.00
Reproductive Adult	0.06	0.14	0.00

EO 15125 - quadrat 5, 2019-2020	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.00	0.02	0.03
Sterile Adult	0.00	1.15	1.35
Reproductive Adult	0.00	0.02	0.00

EO 15125 - quadrat 5, 2020-2021	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.12	0.15	0.12
Sterile Adult	1.45	1.08	1.45
Reproductive Adult	0.00	0.11	0.00

EO 15125 - quadrat 7, 2019-2020	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.52	0.49	0.52
Sterile Adult	2.07	1.73	2.07
Reproductive Adult	0.00	0.24	0.00

EO 15125 - quadrat 7, 2020-2021	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.38	0.30	0.09
Sterile Adult	1.18	1.20	0.79
Reproductive Adult	0.07	0.09	0.00

Table 12. Transitional matrices at the location-transitional year level. Columns are life stage in the first year; rows are the life stage in the second year.

EO 7130, 2019-2020	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.06	0.06	0.02
Sterile Adult	0.47	0.65	0.15
Reproductive Adult	0.01	0.04	0.00

EO 7130, 2020-2021	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.07	0.09	0.04
Sterile Adult	0.93	0.80	0.43
Reproductive Adult	0.03	0.03	0.11

EO 12376, 2019-2020	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.00	0.04	0.13
Sterile Adult	0.00	0.78	0.94
Reproductive Adult	0.00	0.15	0.31

EO 12376, 2020-2021	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.01	0.14	0.11
Sterile Adult	0.74	0.81	0.59
Reproductive Adult	0.06	0.13	0.18

EO 15125, 2019-2020	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.52	0.09	0.30
Sterile Adult	2.07	1.10	1.49
Reproductive Adult	0.00	0.05	0.00

EO 15125, 2020-2021	Young Ramet	Sterile Adult	Reproductive Adult
Young Ramet	0.39	0.14	0.07
Sterile Adult	1.19	1.21	0.93
Reproductive Adult	0.06	0.10	0.01