

*NAJAS FLEXILIS* (HYDROCHARITACEAE) IN ALASKA:  
A REASSESSMENT

DONALD H. LES<sup>1</sup>, ANGELA M. LES, AND URSULA M. KING

Department of Ecology & Evolutionary Biology,  
University of Connecticut, Storrs, CT 06269-3043

<sup>1</sup>e-mail: les@uconn.edu

E. L. PEREDO

The Ecosystems Center, Marine Biological Laboratory, 7 MBL Street,  
Woods Hole, MA 02543

**ABSTRACT.** Fifteen *Najas flexilis* collections were made in Alaska during the summer of 2012, with 13 of the stations representing either new or formerly undocumented localities for this imperiled Alaskan species. These field collections characterize the Alaskan habitats of *N. flexilis* as shallow water sites (<1.5 m) with sand-dominated substrates (71% of sites) and have documented an additional 28 species associates (a 300% increase). However, the additional collections have not extended the elevational, latitudinal, or longitudinal extent of *N. flexilis* from the limits indicated by previous Alaskan collections. *Najas flexilis* remains rare in Alaska as evidenced by a low specimen recovery rate (10%) from potentially suitable sites, and a total of only 12 geographically distinct localities known across the entire state. The new collections have furnished valuable study material for morphological and genetic analyses, which have confirmed the identity of Alaskan populations as *N. flexilis*, rather than *N. canadensis*, a recently identified, cryptic, allotetraploid derivative. A synthesis of information indicates that *N. flexilis* is indigenous to Alaska, where it originated via past (versus recent) migrations from other North American rather than Old World populations.

**Key Words:** biological collections, distribution, habitat, *Najas canadensis*, water-nymph

The aquatic genus *Najas* L. (Hydrocharitaceae) comprises approximately 40 species of submersed, hydrophilous (water-pollinated) angiosperms. The group is distributed worldwide, with nine species found in North America (Haynes 2000; Les et al. 2015). *Najas flexilis* (Willd.) Rostk. & W.L.E. Schmidt is an indigenous North American annual, which extends broadly across the north temperate zone from New England to Alaska (Haynes 2000; Les et al. 2015). *Najas flexilis* is intolerant of pollution and is regarded as an indicator of cool, clear, northern waters (Haynes 2000; Wentz and Stuckey 1971).

Fossils document the presence of *Najas flexilis* in central Alaska some 9300 years ago (Les et al. 2015; Robinson et al. 2007). Yet until fairly recently, the current North American range of *N. flexilis* was thought to extend only to northern Alberta, Canada (Haynes 2000; Porsild and Cody 1980). Shortly thereafter, Cook and Roland (2002) provided the first account of extant *N. flexilis* populations in Alaska, based on a collection made during their floristic survey of Wrangell-St. Elias National Park in 1994 (McCarthy Quad) and from earlier, unreported Alaskan herbarium material that had been collected in 1986 and 1994 from lakes in the Fort Yukon Quad and Anchorage Quad (Appendix). Subsequent collections from 1999–2011 have documented the presence of the species from additional Alaskan localities in the Big Delta Quad, Charley River Quad, Kantishna River Quad, Mt. McKinley Quad, and Talkeetna Quad (Appendix; Carlson et al. 2013; Roland 2004). The first report of *N. flexilis* from Canada's Yukon Territory was based on material collected in 2003 (Cody et al. 2005; Cody and Reading 2005). As a result of the few documented localities, *N. flexilis* currently is regarded as rare in Alaska (Morgan and Sytsma 2009) and is listed as critically imperiled (S1) in both Alaska and the Yukon Territory of Canada (NatureServe 2015).

Recently, Les et al. (2015) demonstrated that *Najas flexilis sensu lato* actually comprises two cryptic, sympatric species, which are divergent genetically and differ subtly by quantitative seed traits. Their sympatric occurrence across North America (they often grow together in the same water body) has long masked their identities as distinct species, which consist of a broader-seeded diploid (*N. flexilis*) and its thinner-seeded allotetraploid derivative (*N. canadensis* Michx.; Les et al. 2015). It is virtually impossible to distinguish from floristic accounts, whether one, the other, or both species occur(s) within an area, unless a photograph or an accurate illustration of the seed is provided for measurement. Thus, although "*N. flexilis*" had been reported from Alaska, it was essential to confirm the identity of that material by DNA analysis to determine whether one or both of the cryptic species occurred there (Les et al. 2015).

The resolution of which of the cryptic *Najas* species occur(s) in Alaska is phytogeographically important because Canada and the lower 48 United States contain both *Najas canadensis* and *N. flexilis*, whereas the Old World is inhabited only by *N. canadensis* (Les et al. 2015). Old World populations of *N. canadensis* extended (at least historically) from northwestern Europe to southern Siberia (Backman 1948; Casper and Krausch 1980), and it is conceivable that the species

could have migrated into North America via a trans-Beringian dispersal route. Thus, a definitive identification of the Alaskan *Najas* material could clarify whether it originated from New World or Old World sources.

It is important to document species distributions accurately in order to avoid spurious inferences. As one example, conclusions of a warmer Holocene climate that are based on the presumed extirpation of *Najas flexilis* in Alaska (Robinson et al. 2007) are arguable based on its actual presence in the state. Although some researchers have challenged this association given that the distribution of *N. flexilis* in northwestern North America remains inadequately understood (Mackay and Matthews 1983), it continues to be reiterated by others (Lowe and Walker 2014). Furthermore, the proper interpretation of such issues also would depend on whether the contemporary Alaskan records of *N. flexilis* represent persistent populations (perhaps indicated by unique genotypes) rather than recent migrants originating from southern North American localities in association with climate warming or other factors. More comprehensive distributional information for *N. flexilis* should help to clarify these uncertainties.

The conservation status of *Najas flexilis* in Alaska and northwestern Canada also could be influenced by whether the extant occurrences represent long-established native populations rather than recently introduced plants. Furthermore, the limited availability of collection records for rare species like *N. flexilis* offers poor insight into geographical patterns and the full potential range of habitat characteristics, which can be helpful to researchers seeking to locate additional occurrences. Thus, clarification of the current distribution and habitat characteristics of *N. flexilis* in Alaska should facilitate conservation efforts as well.

Our interest in the Alaskan distribution of *Najas flexilis* developed while planning to collect specimens for an ongoing systematic study of this species, which required the procurement of plant material suitable for DNA analysis. The scarcity of reported Alaskan *N. flexilis* records was discouraging, but we wondered whether the few existing collections might be an artifact of insufficient field surveys rather than an accurate reflection of rarity. Roland (2004) remarked that *Najas flexilis*: "... is likely frequently overlooked [in Alaska] due to its aquatic habitat, which is under-collected in most plant inventories." However, actual rarity seemed entirely plausible given that nearly all specimens collected

before 2002 originated not from sites accessible via the Alaskan road system (where much collecting activity occurs), but from remote, wilderness areas.

We were reassured that at least some Alaskan field collections of *Najas flexilis* were possible. Specifically, two previously collected sites (Chokosna Lake, Wrangell-St. Elias National Park; Little Kiowa Lake, Anchorage) were relatively accessible by road and several additional localities had been discovered near the latter locality (N. Troyer, Joint Base Elmendorf-Richardson, pers. comm.). We also had learned of another recently discovered and readily accessible locality near Fairbanks (D. Klein, Institute of Arctic Biology, pers. comm.). Encouraged by these reports, we organized a field trip to collect material of *N. flexilis* from the three most promising localities: Little Kiowa Lake (Anchorage), Chokosna Lake (Wrangell-St. Elias National Park), and Little Lost Lake (Fairbanks; Appendix).

Because the reconnaissance of these stations required a complete circuit of the major Alaskan road system, we also planned to survey for *Najas flexilis* en route in as many sites as possible. The eventual procurement of Alaskan *N. flexilis* specimens enabled us to incorporate that material in a genetic survey of the species from other portions of North America (Les et al. 2015). Although the Alaskan specimens, surveyed by Les et al. (2015), included a number of those listed here, that study provided neither detailed collection information nor a discussion regarding the significance of the Alaskan material, which are among the main objectives of the present report.

#### MATERIALS AND METHODS

From August 4–14, 2012, we traversed 2960 km of Alaskan roads, surveying approximately 150 potential *Najas flexilis* habitats encountered en route. All of our Alaskan collections of *N. flexilis* were sanctioned by the Alaska Natural Heritage Program (M. L. Carlson, University of Alaska Anchorage, pers. comm.). Accessions originating from Wrangell-St. Elias National Park were allowed under collecting permit WRST-2012-SCI-0012 issued to D.H.L. Specimens collected at Joint Base Elmendorf-Richardson were approved by the Bureau of Land Management Anchorage Field Office (Letter of Agreement 1020 AKA 012) and were conducted under the supervision of N. Troyer, the base ecologist. Permission to collect plants on the property of the Alaskan Host B&B was granted by K. Huston.

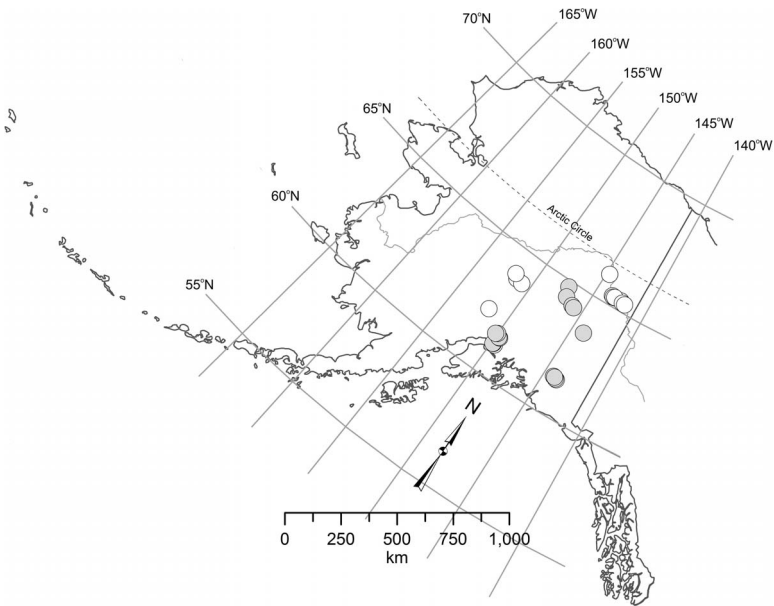


Figure 1. Map of Alaska showing the location of all extant occurrences documented for *Najas flexilis* (see Appendix). White circles = prior records; gray circles = new records.

Specimens were acquired by hand or were retrieved using a 3.7 m collapsible rake. All field-collected plants included material fixed in CTAB preservative (Rogstad 1992) for subsequent genetic analysis and as dried herbarium material for voucher specimens, which were deposited in the ALA and CONN herbaria (Appendix). Observations on associated species, depth, elevation, and substrate composition were recorded at the time of collection. Because collecting permits were not obtained for associated species, their identification was made *in situ*, as accurately as possible (following Crow and Hellquist 2000a, b). Field collections were georeferenced on site using a GPSmap 76CS portable GPS unit (Garmin International, Olathe, KS), which also provided site elevation data. Herbarium accessions without site coordinates were georeferenced manually using the locality information specified by the collectors. Georeferenced records were mapped using the ArcMap application as implemented in the ArcGIS 10 Desktop software package (ESRI: Environmental Systems Research Institute, Redlands, CA) with points displayed using a North America Lambert Conformal Conic

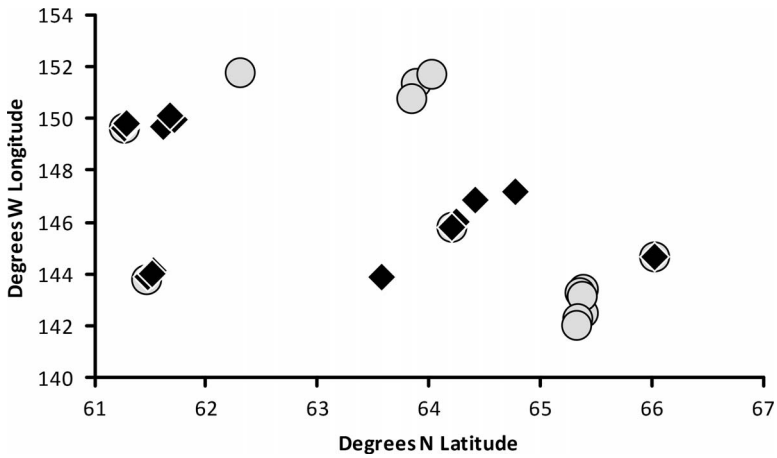


Figure 2. Scatter plot showing the geographical association of all known Alaskan *Najas flexilis* collections. Black diamonds = localities sampled for DNA analysis; gray circles = localities not sampled for DNA analysis.

(ESRI: 102009) projection. A more detailed comparison of record localities was provided by projecting the georeferenced coordinates on a simple scatter plot using MS Excel (Microsoft 2007, Computer Software, Redmond, WA).

A histogram of seed length:width (l:w) ratios specific to Alaskan *Najas flexilis* material was generated and the median calculated using MS Excel, after extracting the appropriate subset of values ( $n = 44$ ) from the full data set ( $n = 225$ ) analyzed by Les et al. (2015).

#### RESULTS

Field collections of *Najas flexilis* were made in 15 different Alaskan water bodies, which represented an approximate recovery rate of 10% from all potentially suitable localities searched (Figure 1). The ranges of latitude ( $61^{\circ}16'48''$ – $64^{\circ}46'12''$ N) and longitude ( $143^{\circ}55'12''$ – $150^{\circ}8'24''$ W) represented by the new collections fell completely within those of previous collections ( $61^{\circ}15'36''$ – $66^{\circ}1'12''$ N;  $142^{\circ}3'36''$ – $151^{\circ}48'0''$ W). A scatter plot of all collection records indicated that they represented at most 12 geographically distinct localities, with all but three of the main clusters including material confirmed previously by DNA analysis (Figure 2). When added to the seven taxa cited in earlier collection

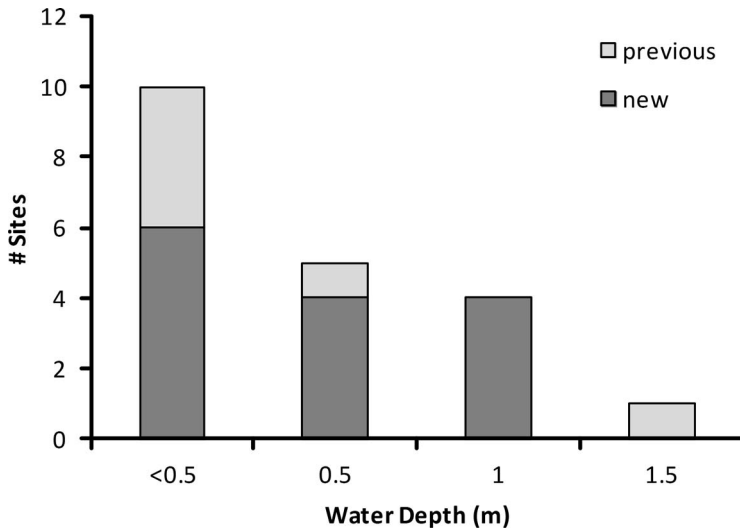


Figure 3. The distribution of Alaskan *Najas flexilis* across varying water depths, compiled from data on previous and new collection records. The bars show frequency of records at each water depth.

records, the 24 taxa found growing with *N. flexilis* resulted in a total compilation of 28 different associates (Table 1). The new collections were made at depths ranging from <0.5–1.0 m (Figure 3) and at elevations ranging 36–481 m ( $\bar{x}$  = 216 m). Out of the nine substrate types recorded at 14 of the collection localities, those dominated by sand (10; 71.4%) were most prevalent and included sand, sandy gravel, sandy gravelly silt, sandy muck, sandy or marly sandy silt, and sandy silty muck. Muck substrates occurred at two sites (14.3%), and silt-dominated types (silty muck, and silty sand) were found at two sites (14.3%).

The range of l:w ratios for *Najas flexilis* seeds sampled from nine Alaskan populations (2.29–3.30; Figure 4) fell within the range for all North American populations reported by Les et al. (2015) with a comparable median value of 2.9 vs. 2.8 for all North American populations.

#### DISCUSSION

New records for *Najas flexilis* collected in southeastern Alaska during the summer of 2012 have essentially doubled the number of

Table 1. Aquatic plant and algae species associated with *Najas flexilis* in Alaska.

New Records	Taxa Reported	Previous Records
<i>Callitriche</i> sp.		<i>Carex</i> sp.
<i>Chara</i> sp.		<i>Comarum palustre</i> L.
<i>Comarum palustre</i> L.		<i>Equisetum fluviatile</i> L.
<i>Calla palustris</i> L.		<i>Menyanthes trifoliata</i> L.
<i>Eleocharis</i> sp.		<i>Myriophyllum sibiricum</i> Kom.
<i>Elodea canadensis</i> Michx.		<i>Potamogeton alpinus</i> Balb.
<i>Equisetum</i> sp.		<i>P. pusillus</i> L.
<i>Isoetes</i> sp.		
<i>Myriophyllum heterophyllum</i> Michx.		
<i>Nuphar polysepala</i> Engelm.		
<i>Nymphaea tetragona</i> Georgi		
<i>Potamogeton ephydrus</i> Raf.		
<i>P. friesii</i> Rupr.		
<i>P. gramineus</i> L.		
<i>P. natans</i> L.		
<i>P. obtusifolius</i> Mert. & W.D.J. Koch		
<i>P. pusillus</i> L.		
<i>P. richardsonii</i> (A. Benn.) Rydb.		
<i>P. robbinsii</i> Oakes		
<i>P. zosteriformis</i> Fernald		
<i>Ranunculus</i> sp. ( <i>Batrachium</i> )		
<i>Sparganium</i> sp.		
<i>Stuckenia pectinata</i> (L.) Börner		
<i>Utricularia macrorhiza</i> Leconte		

known localities of this species in that region (Appendix). Although this result confirms earlier predictions that a more thorough search for this species would yield new records, we still hesitate to categorize *N. flexilis* as common in Alaska and recommend that it continue to be regarded as an imperiled species in the region for several reasons.

Two of the previous collections and one new collection represented duplicate localities, resulting in just 29 unique sites currently known for the entire state of Alaska (Figure 1). Because many of these sites were collected in close proximity (Figure 2), an even narrower distributional profile for *Najas flexilis* in Alaska is evident (i.e., the known localities of this species still represent only about a dozen geographically distinct sites). Furthermore, despite the senior author's familiarity with this species and its habitats, a significant number of potentially promising sites were carefully



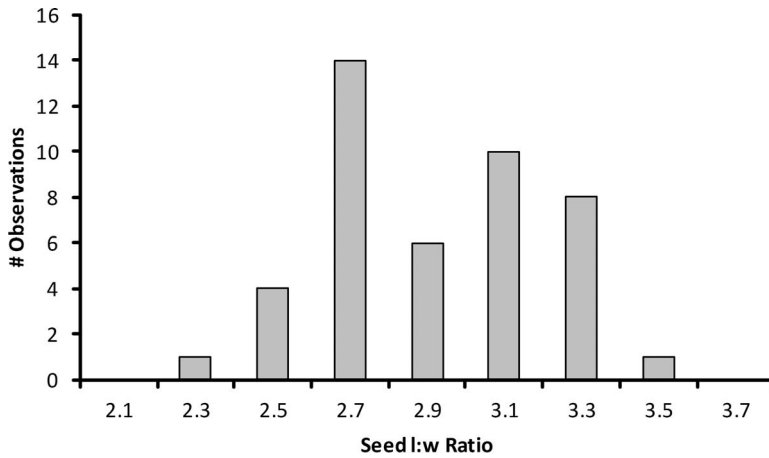


Figure 4. Histogram showing the distribution of length:width (l:w) ratios for *Najas flexilis* seeds sampled from nine Alaskan populations ( $n = 44$ ); the median l:w ratio is 2.9.

searched within the documented distributional range to no avail, at a success rate of only approximately 10%. In contrast, searches made by the senior author in regions where *N. flexilis* is common have routinely found the plants in excess of 75% of sites searched. There also are rather extensive areas (e.g., along the Denali Highway) where the species could not be found despite a fairly thorough search of numerous lakes in that vicinity. The new collections also did not extend the Alaskan range of *N. flexilis* beyond that established by previous collections, leaving substantial areas of the state devoid of any documented occurrences of the species. The Alaskan collections include the northernmost ( $66^{\circ}01'N$ ) and also the westernmost ( $151^{\circ}48'W$ ) sites among those known for extant North American *N. flexilis* populations, with only a slightly greater prior northern range (to  $68^{\circ}13'N$ ) being indicated by fossil records (Les et al. 2015).

Data provided by the additional *Najas flexilis* collections have helped to clarify its Alaskan habitat characteristics and are useful for emending literature reports. As one example, the statement that the Alaskan plants can occur “to depths of 4 m” (Morgan and Sytsma 2009) was based on the depth distribution of *N. flexilis* in other parts of North America but not Alaska, where none of the known collections has been made at a depth exceeding 1.5 m (Figure 3). Moreover, half of the Alaskan collections have been

made at depths of less than 0.5 m and 75% at depths <1 m, which provides a vastly different idea of where one might locate this species, especially if surveys are being conducted in deeper waters by boat. From our own field experience, *N. flexilis* simply did not extend into deeper water. Because these are annual plants, it is possible that they inhabit the warmer, shallower waters because their annual establishment occurs exclusively by seeds, which would not be prone to physical disruption by ice scour as might be the vegetative organs of perennials rooted in shallow sites.

The elevation range spanned by new collections (36–481 m) closely approximated that represented by previous collections (30–484 m) once the erroneous elevation of 619 m cited for Chokosna Lake (Cook and Roland 2002) was corrected to its actual elevation of 483 m (USGS, GNIS 2014). We visited Chokosna Lake during our 2012 survey but were unable to locate *Najas* plants there, despite the discovery of three new sites in similar (but smaller) ponds along McCarthy Road.

New field data have significantly expanded the list of species associated with *Najas flexilis* in Alaska (Table 1). In addition to the seven taxa reported by previous collectors, the recent collections have contributed 24 taxa for a total of 28 unique associates (Table 1). The genus *Potamogeton* L. is the most common associate with 10 species now documented to grow along with *N. flexilis* in Alaska (Table 1).

Prior collections have included little information on substrate composition. Although some substrates/sediments had been characterized as “mineral” and “silty,” other site descriptions such as “bogs” and “organic depressions” would imply more organic bottom compositions. Our collections represented various combinations of gravel, muck, sand, and silt, with sand-dominated types occurring in 71% of the sites. One site was noticeably of a marl composition. Although mucky sediments were common, no site was characterized by a highly organic substrate, and the species was not found in any of the organic-bottomed ponds that were sampled along the Denali Highway.

Thus, by integrating the information available from existing collection data, Alaskan *Najas flexilis* populations can best be characterized as occurring principally at elevations below 500 m, in shallow water depths (0.5–1.5 m), on sand-dominated mineral substrates, and in frequent association with *Potamogeton* species.

In addition to providing more accurate habitat information for Alaskan *Najas flexilis* populations, the simultaneous collection of material for DNA analysis eventually yielded critical insight into the Alaskan distribution of this species. Using the collections referenced here (Figure 2; Appendix), Les et al. (2015) found that all of the combined genetic profiles (based on nrITS, *matK*, *trnK*, and *rbcL* sequence data) of Alaskan populations matched those of *N. flexilis*, with none matching the profiles of *N. canadensis*. Consequently, the genetic data evaluated in that study confirmed that all 17 Alaskan populations that were sampled belonged to *N. flexilis*, and indicated no known occurrences of *N. canadensis* anywhere in Alaska. The median and range of l:w ratios for seeds sampled from nine Alaskan populations corroborated those results. They corresponded closely with ratios obtained for specimens of *N. flexilis* sampled throughout its range, and lacked correspondence with values determined for *N. canadensis* (Les et al. 2015). Consequently, the identity of all studied Alaskan *Najas* material indicates the sole occurrence of the diploid *N. flexilis* by both genetic and morphological evidence (Les et al. 2015). Confirmation that Alaska is inhabited only by *N. flexilis* has clarified that the populations must have originated from North America rather than from Eurasia, where only *N. canadensis* is known to occur.

Moreover, Les et al. (2015) found the same nrITS genotype ('nrF1') in all North American accessions of *Najas flexilis* surveyed except for several Alaskan populations, which possessed a unique genotype ('nrF2'). Alaskan material was found to include not only the nrF1 and nrF2 genotypes, but also a hybrid genotype (nrF1 × nrF2), which was detected in Canada's Yukon Territory as well (Les et al. 2015). The unique Alaskan nrF2 genotype indicates that *N. flexilis* likely has occupied the region for a considerable length of time, and possibly represents an endemic genotype that persisted by surviving in Pleistocene Alaskan refugia. However, the co-occurrence of the common nrF1 genotype (and the hybrid genotypes) would indicate that Alaska also has received migrants from other portions of North America and that hybridization is occurring as the genetically different populations experience secondary contact in Alaska and the Yukon Territory.

Together with contemporary collection records (Appendix), the unique genetic composition of several Alaskan *Najas flexilis* populations establishes that the presence of the species in the state cannot be entirely attributed to recent migration from existing

southern North American localities. Consequently, this species should not be used as an indicator of past climatic history, as often has been proposed (Lowe and Walker 2014; Robinson et al. 2007).

Although recent collections have helped to clarify the status of *Najas flexilis* in northwestern North America, additional searches for the species in Alaska and northwestern Canada are encouraged and should help to further the understanding of the post glacial distribution of this interesting aquatic plant species.

ACKNOWLEDGMENTS. Invaluable assistance was provided by G. T. Bleakley, K. W. Boggs, M. L. Carlson, M. Cook, M. Duffy, D. Etcheverry, S. Ickert-Bond, D. Klein, A. Larsen, J. Metzgar, C. Parker, P. Ray, C. Roland, C. Ross, L. Thorpe, and N. Troyer. We also thank two anonymous reviewers. We are especially grateful to Desi (Fox Run Lodge) for her cordial hospitality. Portions of this work were funded by National Science Foundation grant DEB-0841658 to D.H.L.

#### LITERATURE CITED

- BACKMAN, A. L. 1948. *Najas flexilis* in Europa während der Quartärzeit. Acta Bot. Fenn. 43: 1–43.
- CARLSON, M. L., R. LIPKIN, C. ROLAND, AND A. E. MILLER. 2013. New and important vascular plant collections from south-central and southwestern Alaska: A region of floristic convergence. *Rhodora* 115: 61–95.
- CASPER, S. J. AND H.-D. KRAUSCH. 1980. Süßwasserflora von Mitteleuropa. Band 23: Pteridophyta und Anthophyta. 1. Teil: Lycopodiaceae bis Orchidaceae. Gustav Fischer Verlag, Stuttgart, Germany.
- CODY, W. J., B. A. BENNETT, AND P. CASWELL. 2005. New records of vascular plants in the Yukon Territory VII. *Canad. Field-Naturalist* 119: 417–436.
- AND K. L. READING 2005. Additions and range extensions to the vascular plant flora of the continental Northwest Territories and Nunavut, Canada III. *Canad. Field-Naturalist* 119: 276–290.
- COOK, M. B. AND C. A. ROLAND. 2002. Notable vascular plants from Alaska in Wrangell-St. Elias National Park and Preserve with comments on the floristics. *Canad. Field-Naturalist* 116: 192–304.
- CROW, G. E. AND C. B. HELLQUIST. 2000a. Aquatic and wetland plants of northeastern North America. Vol. 1. Pteridophytes, Gymnosperms, and Angiosperms: Dicotyledons. The University of Wisconsin Press, Madison, WI.
- AND ———. 2000b. Aquatic and wetland plants of northeastern North America. Vol. 2. Angiosperms: Monocotyledons. The University of Wisconsin Press, Madison, WI.
- HAYNES, R. R. 2000. Najadaceae Jussieu. Naiad or water-nymph family, pp. 77–83. *In*: Flora of North America Editorial Committee, eds., Flora of

- North America North of Mexico. Vol. 22. Magnoliophyta: Alismatidae, Arceidae, Commelinidae (in part), and Zingiberidae. Oxford University Press, New York, NY.
- LES, D. H., E. PEREDO, U. M. KING, L. K. BENOIT, N. P. TIPPERY, C. J. BALL, AND R. K. SHANNON. 2015. Through thick and thin: Cryptic sympatric speciation in the submersed genus *Najas* (Hydrocharitaceae). *Molec. Phylogen. Evol.* 82: 15–30.
- LOWE, J. J. AND J. C. WALKER. 2014. Reconstructing Quaternary environments, 3rd ed. Routledge, New York, NY.
- MACKAY, J. R. AND J. V. MATTHEWS JR. 1983. Pleistocene ice and sand wedges, Hooper Island, Northwest Territories. *Canad. J. Earth Sci.* 20: 1087–1097.
- MORGAN, V. H. AND M. SYTSMA. 2009. Introduction to common native & potential invasive freshwater plants in Alaska. The Center for Lakes and Reservoirs at Portland State University, Portland, OR.
- NATURESERVE. 2015. NatureServe Explorer: An online encyclopedia of life [web application]. Version 7.0. NatureServe, Arlington, VA. Website (<http://www.natureserve.org/explorer>). Accessed: January 2015.
- PORSILD, A. E. AND W. J. CODY. 1980. Vascular plants of continental Northwest Territories, Canada. National Museum of Natural Sciences, Ottawa, ON, Canada.
- ROBINSON, S., A. B. BEAUDOIN, D. G. FROESE, J. DOUBT, AND J. G. CLAGUE. 2007. Plant macrofossils associated with an early Holocene beaver dam in interior Alaska. *Arctic* 60: 430–438.
- ROGSTAD, S. H. 1992. Saturated NaCl-CTAB solution as a means of field preservation of leaves for DNA analyses. *Taxon* 41: 405–458.
- ROLAND, C. A. 2004. The vascular plant floristics of Denali National Park and Preserve: A summary, including the results of plant inventory fieldwork 1998–2001. N.P.S. Final Technical Report CAKN-04-01, National Park Service, Denali Park, AK.
- USGS, GNIS. 2014. Geographic Names Information System. U.S. Geological Survey, Reston, VA. Website (<http://geonames.usgs.gov/>). Accessed October 2014.
- WENTZ, W. A. AND R. L. STUCKEY. 1971. The changing distribution of the genus *Najas* (Najadaceae) in Ohio. *Ohio J. Sci.* 71: 292–302.

#### APPENDIX

Previous and new Alaskan collection records of *Najas flexilis* arranged chronologically. Specimens marked by an asterisk (\*) indicate material sampled by Les et al. (2015) in a genetic survey of the species.

#### PREVIOUS REPORTS

Alaska: Fort Yukon Quad. Yukon Flats, Heglund Plot L, Lake 522, near Preacher Cr.; 66°01' N, 144°42' W; fresh standing water <50 cm deep, class 2 bog dominated by *Calla palustris* in old river oxbow, water low in nutrients

(TP=22 ug/L., TN=1502 ug/L) and low in color (32 PtU), with circumneutral pH (6.7). *P. Heglund 86-351*, 16 Jul 1986 (ALA).

Alaska: Fort Yukon Quad. Yukon Flats, Heglund Plot L, Lake 23; 66°01'N, 144°42'W. *P. Heglund 86-363*, 17 Jul, 1986 (ALA).\* [in close proximity to previous record]

Alaska: Anchorage Quad. Anchorage Borough, Cook Inlet lowlands, Little Kiowa Lake, lake margin at parking area; 61°15'29"N, 149°39'23"W; 30 m; wetland, shallow water 0.25 m. *Michael Duffy & Jerry Tande 1030*, 04 Aug 1994 (ALA).\*

Alaska: Anchorage Quad. Fort Richardson Military Reservation, Cook Inlet lowlands, Little Kiowa Lake, lakeshore at the parking area; 61°15'29"N, 149°39'23"W; 30 m; lake/pond about 40 cm deep. *Michael Duffy & Jerry Tande 1117*, 29 Aug 1994 (ALA). [same locality as previous]

Alaska: McCarthy Quad. Wrangell Mountains, Chokosna Lake; 61°27'26"N, 143°49'0"W [61°27.45'N, 143°49'W], 619 m; shallow water. *C. Roland 94-325*, 10 Aug 1994 (UAF).

Alaska: Talkeetna Quad, Denali National Preserve, Yentna R. lowlands, 1.4 km W of E Fork Yentna R., 0.8 km N of Park border, vic. pond W of river; 62°18'0"N, 151°48'0"W [62.3/-151.8]; growing in small shallow pond in extensive wetland and bog complex, growing with *Potamogeton alpinus*, *Potamogeton pusillus* and *Equisetum fluviatile*. *Carl Roland & Carolyn L. Parker 4272*, 17 Aug 1999 (ALA).

Alaska: Mt. McKinley Quad. Denali National Park and Preserve, Alaska Range, Lower Kantishna River east of Chilchukabena Lake, UTMzone 5; E578588, N7085207; 63°52'48"N, 151°23'24"W [63.88/-151.39]; boreal-riparian wet meadow organic depressions with water horsetail-beaked sedge wet meadow pond margin, margin of old slough, in 20 cm of water. *Michael Duffy 00-269*, 14 Aug 2000 (ALA).

Alaska: Mt. McKinley Quad. Denali National Park and Preserve, lowlands W of Kantishna Hills, vic. pond betw. Bearpaw R. and Otter Cr., 3.3 km S of VABM Diamond; 63°50'24"N, 150°48'0"W [63.84/-150.8]; occasional in shallow water in beaver-impounded pond on silty substrate. *Carl Roland & Alan R. Batten 5202A*, 06 Aug 2001 (ALA).

Alaska: Charley River Quad. Yukon-Charley Rivers National Park, Yukon River valley, Yukon R. corridor, 5.2 km SW of VABM Wedge, large bog complex; 65°22'48"N, 143°26'60"W [65.38/-143.45]; well developed bog with small areas of open water interspersed throughout, margin dominated by black spruce upland. *Amy Larsen & Alan R. Batten 02-2333*, 22 Jul 2002 (ALA).

Alaska: Charley River Quad. Yukon-Charley Rivers National Preserve, Yukon River at mouth of Woodchopper Cr., vic. of small pond along Yukon R. 1.2 km downstream from Woodchopper Roadhouse; small depressional wetland with gently sloping shoreline near mouth of Woodchopper Cr., with predominately mineral substrate; 65°21'0"N, 143°19'12"W [65.35/-143.32]. *Amy Larsen & Alan R. Batten 02-2369*, 23 Jul 2002 (ALA).

Alaska: Charley River Quad. Yukon-Charley Rivers National Park, Yukon River valley, north bank of Yukon R., 1.8 km downstream of Kandik R.; 65°22'48"N, 142°31'48"W [65.38/-142.53]; large open water pond with narrow littoral band dominated by *Menyanthes trifoliata* and *Carex*, uplands predominately birch. *Amy Larsen & Alan R. Batten 02-2397*, 24 Jul 2002 (ALA).

Alaska: Charley River Quad. Yukon-Charley Rivers National Preserve, Yukon River valley, Yukon River, westernmost edge of Andrew Creek Flats, vic. small pond adjacent to river 1.5 km N of McGregor's Cabin; 65°21'36"N, 143°10'12"W [65.36/-143.17]; large shallow pond with, with mineral sediments and a well-developed band of *Equisetum fluviatile*. *Amy Larsen & Mary Beth Cook 02-2456*, 03 Aug 2002 (ALA).

Alaska: Charley River Quad. Yukon-Charley Rivers National Preserve, Yukon River valley, vic. of Andrew Creek, small pond adjacent to Yukon R in westernmost edge of Andrew Cr. Flats, 1.5 km N of McGregor's Cabin, T6N R21E SE4 SE4 Sec. 3; 65°22'11.4"N, 143°10'32.4"W [65.36983333/-143.175667], elev = 198 m; *Equisetum* meadow surrounding pond in white spruce forest. *Mary Beth Cook & Amy Larsen 02-519*, 03 Aug 2002 (ALA). [same site as previous]

Alaska: Charley River Quad. Yukon-Charley Rivers National Park, Yukon River valley, wetlands along N bank of Yukon R., at base of Kathul Mt.; 65°19'48"N, 142°19'12"W [65.33/-142.32]; small steep sided pond with well developed bog. *Amy Larsen & Mary Beth Cook 02-2520*, 04 Aug 2002 (ALA).

Alaska: Charley River Quad. Yukon-Charley Rivers National Park, Yukon River valley, small lake along N bank of Yukon R., 6.7 km W of VABM Wedge; 65°19'12"N, 142°3'36"W [65.32/-142.06]; small lake near Seymour Lake, highly productive submersed vegetation; littoral zone was burned during the 1999 Nation fire. *Amy Larsen & Mary Beth Cook 02-2549*, 05 Aug 2002 (ALA).

Alaska: Kantishna River Quad. Denali National Park and Preserve, Minchumina Basin, Beaverlog Lakes, Transect 02MC130 Stop 02, UTM zone 5, E561186, N7100277; 64°1'12"N, 151°44'24"W [64.02/-151.74]; in buckbean-marsh five-finger wet meadow on boreal-riparian sedge wet meadow organic depressions, at pond margin in 1.5 m of water. *Michael Duffly MD02-332*, 02 Sep 2002 (ALA).

Alaska: Big Delta Quad. Southeast Fairbanks Borough, Shaw Cr Flats: NE corner of lake N of W end of Quartz Lake [Little Lost Lake]; 64°12'02.26"N, 145°50'25.02"W; specimens coll. inadvertently, along w/#2604-2606, thus fragmentary; shoots apparently loose on the bottom of ~0.5 m deep water, along w/*Myriophyllum sibiricum* #2604. This family & sp. are not listed by either Hulten or Cody! *David R. Klein 2608*, 30 Jul 2011 (ALA).

#### NEW REPORTS

Alaska: Anchorage Quad. Anchorage Borough, Anchorage, Elmendorf-Richardson Joint Base, Lower Six Mile Lake, NW side of lake, access road off of Loop Road; 61°17'30.5"N, 149°48'15.8"W  $\pm 7$  m, elev = 36 m; locally common but widely spaced along edge of lake in <0.5 m water, on sandy muck; with *Myriophyllum*, *Potamogeton*, *Chara*, *Nymphaea*, *Comarum palustre*, *Calla palustris*. *Donald H. Les 1126 & Angela M. Les s.n.*, 04 Aug 2012 (ALA; CONN).\*

Alaska: Anchorage Quad. Anchorage Borough, Anchorage, Elmendorf-Richardson Joint Base, Green Lake, north side of lake by picnic area; 61°16'49.7"N, 149°50'09.5"W  $\pm 30$  m, elev = 41 m; <0.5 m depth, on sandy muck; with *Sparganium*, *Potamogeton richardsonii*, *Chara*. *Donald H. Les 1127 & Angela M. Les s.n.*, 04 Aug 2012 (ALA; CONN).\*



Alaska: Anchorage Quad. Matanuska-Susitna Borough, Cloudy Lake, NW end, from road access off N Phillips Drive, just N of Meadow Lakes Loop; 61°36'43.6"N, 149°38'22.1"W ±7 m, elev = 95 m; occasional on mucky bottom of small lake, in 1 m water; with *Potamogeton gramineus*, *Utricularia*, *Nuphar polysepala*. Donald H. Les 1128 & Angela M. Les s.n., 05 Aug 2012 (ALA; CONN).\*

Alaska: Anchorage Quad. Matanuska-Susitna Borough, Frog Lake, at end of public access road at E side of lake; 61°36'50.0"N, 149°43'23.9"W ±6 m, elev = 89 m; common in shallow (<0.5 m) water along shore of lake on sandy gravel bottom; with *Potamogeton richardsonii*, *P. gramineus*, *Isoetes*. Donald H. Les 1129 & Angela M. Les s.n., 05 Aug 2012 (ALA; CONN).\*

Alaska: Tyonek Quad. Matanuska-Susitna Borough, Willow, small unnamed lake adjacent to Alaskan Host Bed & Breakfast; 61°42'39.7"N, 149°59'41.7"W ±9 m, elev = 83 m; common in clear water <1 m deep on silty muck bottom; with *Nuphar polysepala*, *Myriophyllum heterophyllum*, *Ranunculus (Batrachium)*, *Sparganium*, *Potamogeton* spp., and *Callitriche*. Donald H. Les 1130 & Angela M. Les s.n., 05 Aug 2012 (ALA; CONN).\*

Alaska: Tyonek Quad. Matanuska-Susitna Borough, Willow, Nancy Lake State Recreation Area, South Rolly Lake, SE edge, near campground entrance, access from campsite A40; 61°40'02.2"N, 150°08'18.3"W ±10 m, elev = 54 m; only occasional on silty sand in <1 m of clear water; with *Potamogeton epihydrus*, *Isoetes*, *Nuphar polysepala*. Donald H. Les 1131 & Angela M. Les s.n., 06 Aug 2012 (ALA; CONN).\*

Alaska: Fairbanks Quad. North Star Borough, Fairbanks, Chena Lakes Recreation Area, Chena Lake, E end of lake on Lake Park Road; 64°46'06.9"N, 147°12'50.7"W ±6 m, elev = 119 m; common but flat on bottom of sandy substrate in 0.5 m water; in fruit; plants brittle and falling apart easily; with *Potamogeton richardsonii*, *P. robbinsii*, *Stuckenia pectinata*, and *Elodea canadensis*. Donald H. Les 1132 & Angela M. Les s.n., 08 Aug 2012 (ALA; CONN).\*

Alaska: Big Delta Quad. North Star Borough, Fairbanks, Little Harding Lake, public access site on E end of small bog lake, W of Harding Lake; 64°24'40.5"N, 146°53'38.7"W ±5 m, elev = 228 m; occasional in clear water <0.5 m deep on sandy gravel silt bottom; with *Nymphaea tetragona*, *Myriophyllum*, *Potamogeton pusillus*, *Utricularia macrorhiza*, *Sparganium*. Donald H. Les 1134 & Angela M. Les s.n., 09 Aug 2012 (ALA; CONN).\*

Alaska: Big Delta Quad. Southeast Fairbanks Borough, small unnamed pond ["milepost 284 pond"] in Shaw Creek Flats area, approx. 285 Richardson Highway (just N of milepost 284), W side of road, ~ 5 mi N of Quartz Lake; 64°14'29.5"N, 146°03'01.8"W ±5 m, elev = 283 m; occasional in 0.5 m water on marly sandy silt; in fruit; with *Chara*, *Potamogeton friesii*, *P. gramineus*. Donald H. Les 1135 & Angela M. Les s.n., 09 Aug 2012 (ALA; CONN).\*

Alaska: Big Delta Quad. Southeast Fairbanks Borough, Quartz Lake, western shore between boat rental docks and public boat launch; 64°11'55.4"N, 145°49'35.3"W ±5 m, elev = 291 m; occasional in clumps on sandy silt bottom in <0.5 m water; very brittle; many fragments also; with *Potamogeton gramineus*, *P. friesii*, *Stuckenia pectinata*, *Myriophyllum*, and emergent *Equisetum*. Donald H. Les 1136 & Angela M. Les s.n., 09 Aug 2012 (ALA; CONN).\*



Alaska: Big Delta Quad. Southeast Fairbanks Borough, Little Lost Lake, dock at public access site, SE end of lake, S side of dock; 64°11'54.0"N, 145°50'25.4"W ±5 m, elev = 294 m; abundant in water <1 m deep on sandy silty muck bottom; with *Potamogeton richardsonii*, *P. robbinsii*, *Nymphaea tetragona*, *Potamogeton obtusifolius*. Donald H. Les 1137 & Angela M. Les s.n., 09 Aug 2012 (ALA, CONN).\* [same general site as Klein 2608 (ALA)]

Alaska: Mount Hayes Quad. Southeast Fairbanks Borough, Jan Lake, public access on N side of lake, off highway 2; 63°33'58.4"N, 143°55'07.8"W ± 5 m, elev = 481 m; common on sandy bottom in 30 cm clear water; with *Myriophyllum*, *Potamogeton zosteriformis*, *P. richardsonii*, emergent *Eleocharis*. Donald H. Les 1138 & Angela M. Les s.n., 09 Aug 2012 (ALA, CONN).\*

Alaska: Valdez Quad. Valdez-Cordova Borough, Wrangell-St. Elias National Park, McCarthy Road, small unnamed pond [pond #1] on N side of road; 61°31'20.9"N, 144°10'09.6"W ±5 m, elev = 367 m; abundant in pond in ~1 m water; with *Nuphar polysepala*, *Nymphaea tetragona*, *Sparganium*, *Chara*, *Myriophyllum*, *Potamogeton* spp. Donald H. Les 1139 & Angela M. Les s.n., 11 Aug 2012 (ALA, CONN).\* [collecting permit: No. WRST-2012-SCI-0012]

Alaska: Valdez Quad. Valdez-Cordova Borough, Wrangell-St. Elias National Park, McCarthy Road, small unnamed pond [pond #2] on N side of road; 61°28'06.3"N, 143°56'01.8"W ±4 m, elev = 395 m; common in 0.5 m water on muck; with *Potamogeton natans*, *Chara*, *Sparganium*. Donald H. Les 1140 & Angela M. Les s.n., 11 Aug 2012 (ALA, CONN). [collecting permit: No. WRST-2012-SCI-0012]

Alaska: Valdez Quad. Valdez-Cordova Borough, Wrangell-St. Elias National Park, McCarthy Road, small unnamed pond [pond #3] on S side of road; 61°30'18.0"N, 144°03'15.7"W ±5 m, elev = 385 m; common in 0.5 m water on sandy muck; with *Nuphar polysepala*, *Potamogeton gramineus*, *Chara*. Donald H. Les 1141 & Angela M. Les s.n., 11 Aug 2012 (ALA, CONN).\* [collecting permit: No. WRST-2012-SCI-0012]