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Central and Arctic Region

Canadian Science Advisory Secretariat
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RECOVERY POTENTIAL ASSESSMENT OF THREEHORN WARTYBACK (*Obliquaria reflexa*) IN CANADA

*Figure 1. Threehorn Wartyback (*Obliquaria reflexa*). Photograph by Environment Canada, reproduced with permission.*

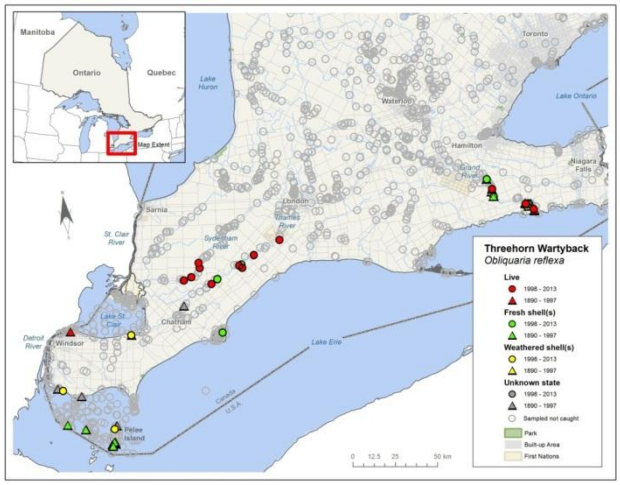

Figure 2. Distribution of Threehorn Wartyback in Canada

Context:

*In May 2013, the Committee on the Status of Endangered Wildlife in Canada (COSEWIC) assessed the status of Threehorn Wartyback (*Obliquaria reflexa*) and determined the designation to be Threatened. The reason provided for this designation is that, "This rare species historically occurred in the Great Lakes drainages including Lake St. Clair, western Lake Erie, and the Grand, Thames, and Detroit rivers. The species has not been found since 1992 in Lake St. Clair and the Detroit River and may be extirpated there due largely to the impacts of Zebra and Quagga mussels. It was last recorded from the Canadian side of Lake Erie in 1997. Pollution (sediment loading, nutrient loading, contaminants and toxic substances) related to both urban and agricultural activities represent a high and continuing threat at the three remaining riverine locations." Threehorn Wartyback is currently not listed under the* Species at Risk Act *(SARA).*

A species Recovery Potential Assessment process has been developed by Fisheries and Oceans Canada (DFO) Science to provide the information and scientific advice required to meet the various requirements of the SARA, including advice to the Minister of DFO regarding the listing of the species under the SARA. It is also used when analyzing the socio-economic impacts of adding the species to the list as well as during subsequent consultations, where applicable. If listed, this scientific advice will also be used in the development of a recovery strategy, and to support decision-making with regards to SARA agreements and permits. This assessment considers the scientific data available with which to assess the recovery potential of Threehorn Wartyback in Canada.

SUMMARY

- In Canada, the current and historic known distribution of Threehorn Wartyback (*Obliquaria reflexa*) is limited to five confirmed populations, one of which is currently considered to be extirpated. Extant populations include two tributaries of Lake St. Clair (Sydenham, and Thames rivers), and the Grand River (Lake Erie drainage; Figure 2).
- Threehorn Wartyback glochidia must encyst on the gills of an appropriate host fish to survive and metamorphose. The putative host fishes for Threehorn Wartyback in Canada include Common Shiner (*Luxilus cornutus*) and Longnose Dace (*Rhinichthys cataractae*). This is supported by laboratory infestations in the United States (as infestation experiments have not occurred with Canadian Threehorn Wartyback), and distributional overlap of known ranges in Canadian waters.
- Threehorn Wartyback is most commonly found in large rivers with stable gravel, sand and mud substrates with moderate current; however, it may also be found in shallow embayments and reservoirs with almost no current.
- Based on what is known of Threehorn Wartyback life history (low fecundity, short lifespan, relatively early maturity) previous modeling of Unionid mussels suggests that, compared to other Unionid species, Threehorn Wartyback is expected to be most sensitive to perturbation or uncertainty in juvenile survival, adult survival, and lifespan, and relatively insensitive to changes in glochidial survival, fecundity, or age at maturity.
- It appears that the greatest limiting factors to the stabilization and growth of Threehorn Wartyback populations in Canada are largely attributed to the presence of contaminants and toxic substances in their environment, habitat removal and alteration, and the introduction and establishment of various invasive species.
- A number of key sources of uncertainty exist for this species related to population distribution, population structure, habitat preferences and to the factors limiting their existence.
- Specifically, there is a need for a continuation of quantitative sampling to inform the population status assessment. There is a need for exploratory sampling in systems with habitat characteristics similar to those areas where Threehorn Wartyback is known to occur. To confirm host fishes in Canada, there is a need to complete laboratory, and if feasible field experiments. Many life history characteristics required to inform population modelling efforts are currently unknown for this species and should be investigated to inform modelling efforts.

BACKGROUND

In May 2013, the Committee on the Status of Endangered Wildlife in Canada (COSEWIC) assessed the status of Threehorn Wartyback and determined the designation to be Threatened. The reason provided for this designation is that, “This rare species historically occurred in the Great Lakes drainages including Lake St. Clair, western Lake Erie, and the Grand, Thames, and Detroit rivers. The species has not been found since 1992 in Lake St. Clair and the Detroit River and may be extirpated there due largely to the impacts of Zebra and Quagga mussels. It was last recorded from the Canadian side of Lake Erie in 1997. Pollution (sediment loading, nutrient loading, contaminants and toxic substances) related to both urban and agricultural activities represent a high and continuing threat at the three remaining riverine locations.” Threehorn Wartyback is currently not listed under the *Species at Risk Act* (SARA).

When COSEWIC designates an aquatic species as Threatened or Endangered and the Governor in Council decides to list it, the Minister of Fisheries and Oceans Canada (DFO) is

required by the SARA to undertake a number of actions. Many of these actions require scientific information such as the current status of the population, the threats to its survival and recovery, and the feasibility of its recovery. This scientific advice is developed through a Recovery Potential Assessment. This allows for the consideration of peer-reviewed scientific analyses in subsequent SARA processes, including permitting on harm and recovery planning. This Recovery Potential Assessment focuses on the Threehorn Wartyback populations in Canada and is a summary of the conclusions and advice from a Canadian Science Advisory Secretariat peer-review meeting that occurred on December 10, 2013 in Burlington, Ontario. A research document, providing background information on the species biology, habitat preferences, current status, sensitivity to perturbations, threats, and mitigations (Bouvier et al. 2014) provides an in-depth account of the information summarized below. Proceedings that document the activities and key discussions of the meeting are also available (DFO 2014). Please note that reference citations have been removed from the following document to minimize the length of the document. Complete reference citations are available at Bouvier et al. (2014).

Species Description

Threehorn Wartyback is a medium-sized freshwater mussel with an average shell length of approximately 40 mm, while a maximum shell length of 80 mm has been reported from US waters. Lengths of Threehorn Wartyback recorded from the Sydenham River (n=37) ranged from 15 to 64 mm, while lengths recorded from the Thames River (n=24) ranged from 28 to 62 mm (Figure 3). Of the shells available from the Grand River (n=64), sizes ranged from 23.4 to 56 mm (Figure 3). In a study on the variations of reproductive traits, Haag and Staton (2003) noted that male Threehorn Wartyback collected from the Little Tallahatchie River (Mississippi, United States) were significantly larger than female Threehorn Wartyback; however, this information is not currently available from Canadian populations.

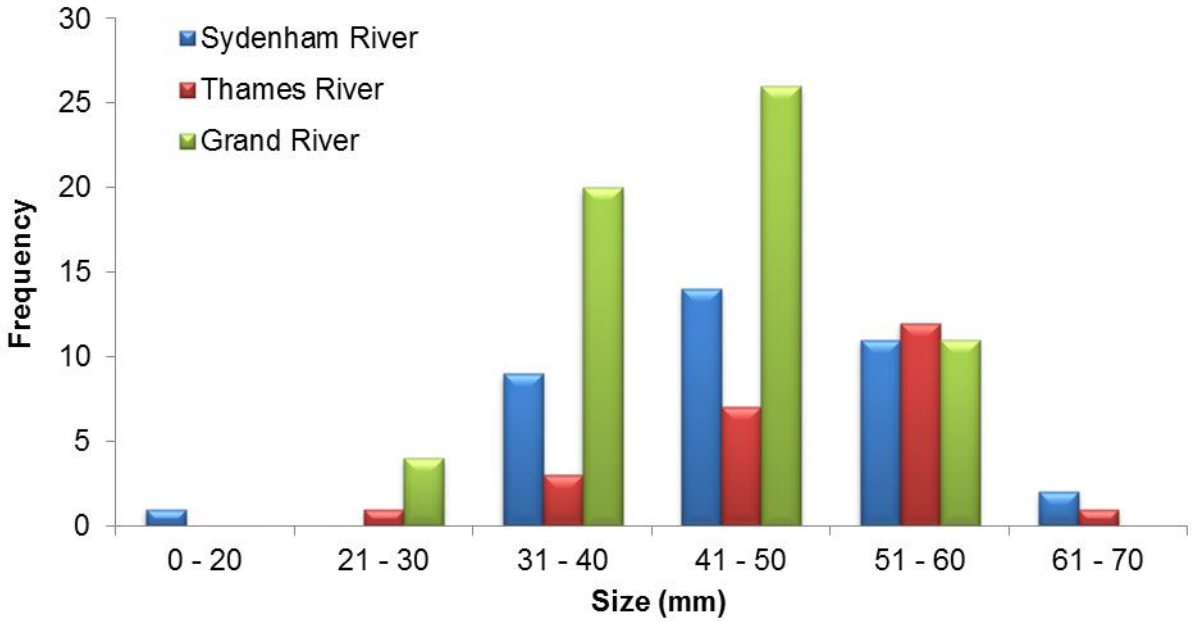

Figure 3. Size distribution of Threehorn Wartyback recorded from the Sydenham, Thames and Grand rivers [modified from COSEWIC (2013)].

The shell is described as thick, circular to triangular, and inflated (Figure 1). The anterior end is rounded, and the posterior end is bluntly pointed. While Threehorn Wartyback is dioecious, they lack pronounced sexual dimorphism. The most prominent shell feature is the single row of two to five knobs extending from the beak to the ventral margin, which alternate in position between

valves. The beaks are elevated and curved inward. The periostracum varies from green, tan or brown with rays, while the nacre is white and iridescent posteriorly. The hinge teeth are strong and fully developed.

Similar Species

Threehorn Wartyback is the only member of the genus *Obliquaria* known to occur in Canada. There are no morphologically similar species present in Canada, as Threehorn Wartyback can be easily distinguished by the presence of its characteristic alternating knobs.

Age and Growth

Threehorn Wartyback is considered to be a moderately short-lived species, with a maximum age estimate reported of 18 years. This age estimate is consistent for both Canadian (Morris, unpubl. data) and Ohio populations. Sixty valves sampled from the Grand River in 1997 were aged to determine the length at age relationship (Figure 4). Mussel ages ranged from two (29 to 41 mm) to 14 years of age (54 mm). No additional information on age and growth patterns is available, locally or globally for this species.

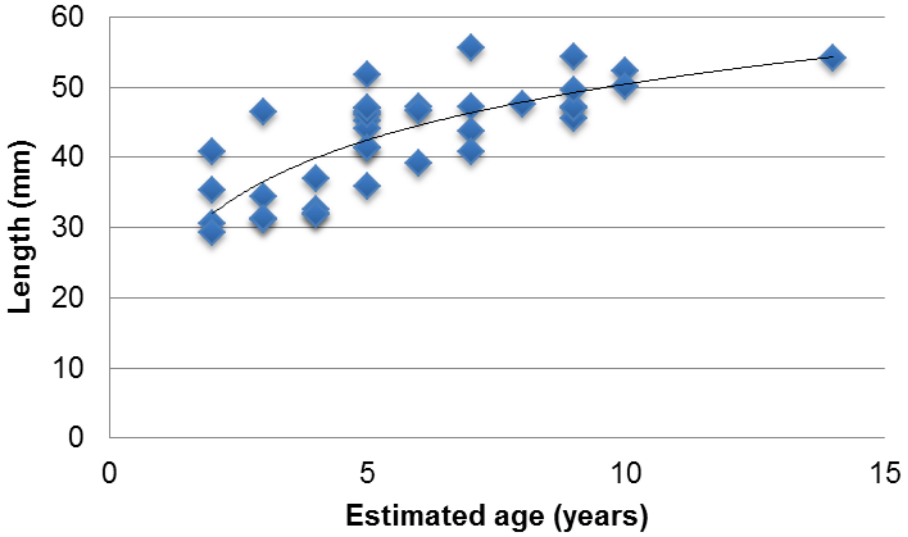

Figure 4. Length at age estimates for Threehorn Wartyback collected from the Grand River (DFO unpubl. data).

Diet

Like most other unionid mussels, Threehorn Wartyback is considered to be a filter feeder. Filter feeding (also called suspension feeding) is accomplished by using cilia to pump water through their incurrent siphon and over the gills. Particles are subsequently sorted by cilia on the gills and directed towards the mouth for consumption. In the early juvenile stage, when the mussel is most commonly buried in the substrate, food is obtained directly from the substrate in the form of algae and bacteria. Species-specific dietary information is not available for Threehorn Wartyback.

ASSESSMENT

Current Species Status

In Canada, the current and historic known distribution of Threehorn Wartyback is limited to five populations, one of which is currently considered to be extirpated and one that is represented by a single fresh shell. The Rondeau Bay location is represented by a single fresh shell recorded in 2011. Although Threehorn Wartyback is thought to be extirpated from the Canadian Great Lakes and connecting channels, 13 live individuals were recently detected in coastal wetlands and embayments in the western basin of Lake Erie and Sandusky Bay, Ohio (D. Zanatta, unpubl. data). Extant populations include the Sydenham and Thames rivers (tributaries of Lake St. Clair) and the Grand River (tributary of Lake Erie; Figure 2). Live individuals have been recorded from all extant sites, with the greatest number of Threehorn Wartyback being recorded from the Sydenham River (n=73 since 1998). It should be noted that the following maps represent all current and historic records of Threehorn Wartyback, and may not accurately represent the current distribution. Substantial mussel sampling has occurred throughout Ontario; however, there has been limited sampling of the Great Lakes proper and connecting channels for mussels, as it is believed that most freshwater mussels are now extirpated from these areas following the dreissenid mussel invasion. Therefore, the following maps may be an underrepresentation of the current distribution, if Threehorn Wartyback is persisting, undetected from Canadian waters.

Sydenham River

Threehorn Wartyback was first recorded from the Sydenham River in 1998 when one live individual was observed at Dawn Mills, and a second fresh shell was observed at Croton (Figure 5). The Dawn Mills site has been re-sampled yearly from 2002 to 2009 and has resulted in the observance of 72 live individuals (33 recaptures and 39 new individuals). Threehorn Wartyback are known to occupy the reach of the Sydenham River between Dawn Mills and a site slightly upstream from Florence. Quantitative mussel surveys have been conducted in the Sydenham River; although population size estimates are not available for Threehorn Wartyback as only a single live individual was collected during these surveys. It is believed that recruitment is occurring in the Sydenham River population based on the current size frequency distribution (Figure 3), and the observation of a 15 mm individual (K. McNichols-O'Rourke, DFO, pers. obs.).

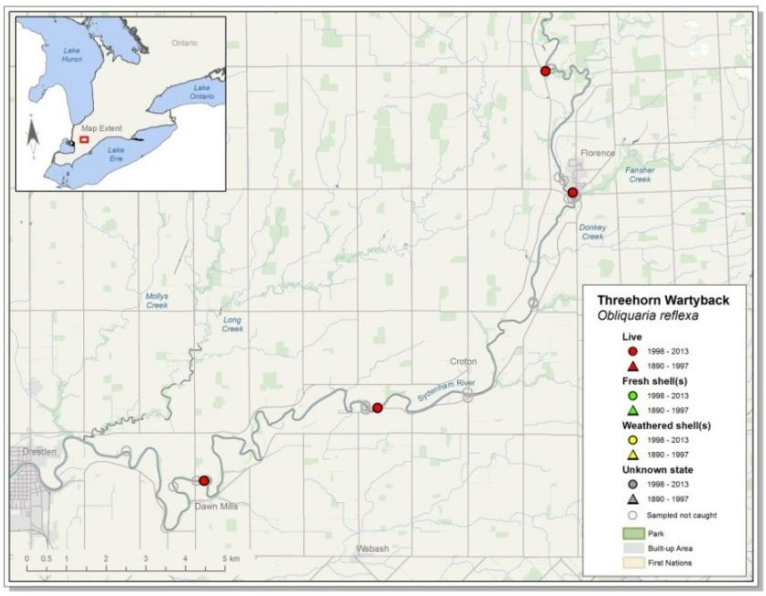

Figure 5. Distribution of all known current and historic Threehorn Wartyback records from the Sydenham River.

Thames River

There is a single historic record of Threehorn Wartyback in the Thames River, which was collected by J.P. Oughton in 1934. Threehorn Wartyback was not recorded from this system again until 1998 when one live individual and one fresh shell were recorded (Figure 6). Five additional live Threehorn Wartyback were recorded from four sites in the Thames River in 2004. DFO subsequently observed Threehorn Wartyback in the Thames River in 2005 and 2010. Threehorn Wartyback is currently known to occupy a 100 km reach of the Thames River. A total of 30 live individuals have been collected from this system since 1998. Morris and Edwards (2007) estimated that the relative abundance of Threehorn Wartyback in the Thames River is 0.22% with an overall relative abundance of 0.024 animals/m^2. Considering the known range of Threehorn Wartyback in this system, the population is estimated to be approximately 100 000 individuals.

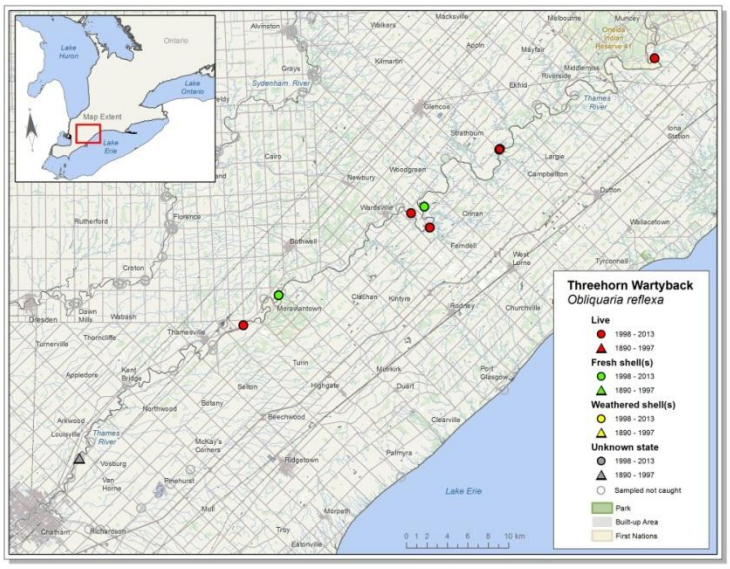

Figure 6. Distribution of all known current and historic Threehorn Wartyback records from the Thames River.

Grand River

Unlike the Sydenham and Thames rivers, historical records of fresh shells are available from museum collections for the Grand River. A total of 68 fresh shells were recorded from the Grand River between 1980 and 1988. The first live individuals were recorded in 1997 from Sulphur Creek, and the Grand River proper (Figure 7). During a 1997 survey three live individuals, 40 fresh shells and 14 weathered shells were recorded from seven sites. One fresh shell was recorded in 2005, and one weathered shell was recorded in 2007. Most recently, four live individuals, five fresh shells and seven weathered shells were recorded from seven sites sampled in 2011. Population size estimates are not available for Threehorn Wartyback in the Grand River.

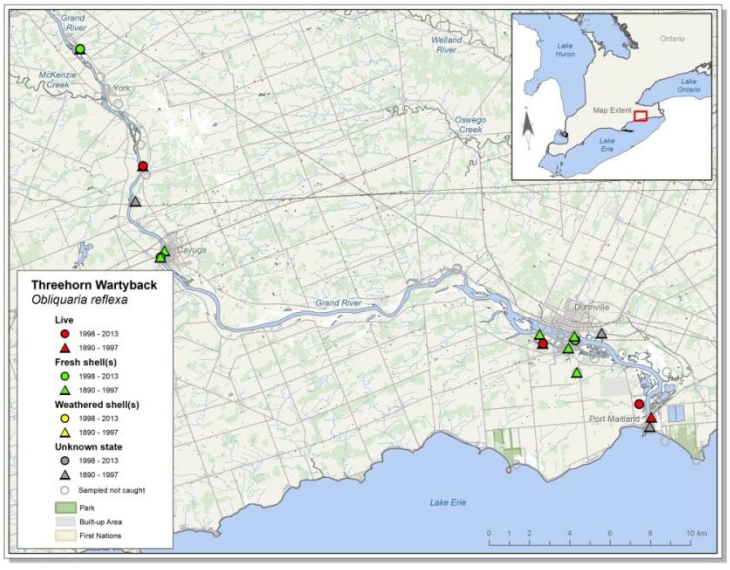

Figure 7. Distribution of all known current and historic Threehorn Wartyback records from the Grand River.

Rondeau Bay

One fresh Threehorn Wartyback shell was observed from Rondeau Bay in 2001 (collectors: D. Zanatta and D. Woolnough; Figure 8). This record represents the first, and only, record of Threehorn Wartyback in Rondeau Bay. Due to the scarcity of information related to Threehorn Wartyback in this system, Rondeau Bay will not be considered a population in the Population Status Assessment. Additional sampling in Rondeau Bay should be completed to determine whether a Threehorn Wartyback population persists in this system.

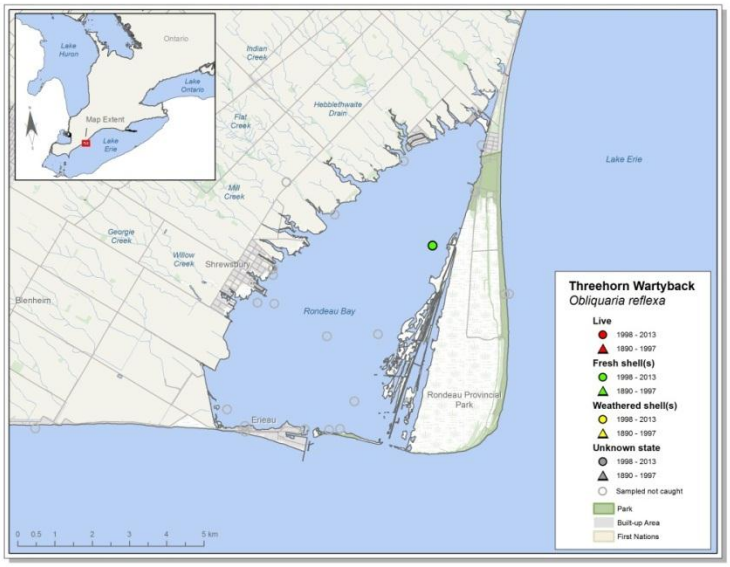

Figure 8. Distribution of all known current and historic Threehorn Wartyback records from Rondeau Bay.

Great Lakes and connecting channels

The first historic record of Threehorn Wartyback from the Great Lakes or their connecting channels was observed by M.E. Walker in 1925. The state or quantity of individuals observed is not available, but the location of this observation was near Oxley, on the northeastern shore of Lake Erie. Additional museum records provide evidence of shells recorded at various locations in Lake Erie including, Pelee Island (fresh shells collected from 1937 to 2005), East Sister Island (1967), Middle Sister Island (1952), The Meadows (2005) and the mouth of Big Creek (1982) (Figure 9). Despite these numerous shell collections, a live individual was not recorded until 1992 when Schloesser et al. (1998) recorded three live individuals from the Detroit River, near its confluence with Lake St. Clair. This record also represents the only live collection of Threehorn Wartyback from the Great Lakes and its connecting channels in Canadian waters. In 1998, additional sampling of previously visited sites yielded no observation of Threehorn Wartyback and it was concluded by investigators that unionids had been extirpated from the main river of the Detroit River as a result of the invasion of dreissenid mussels. Due to a lack of current observations of live Threehorn Wartyback in Canadian waters of the Great Lakes and connecting channels, this population will not be included in the Population Status Assessment.

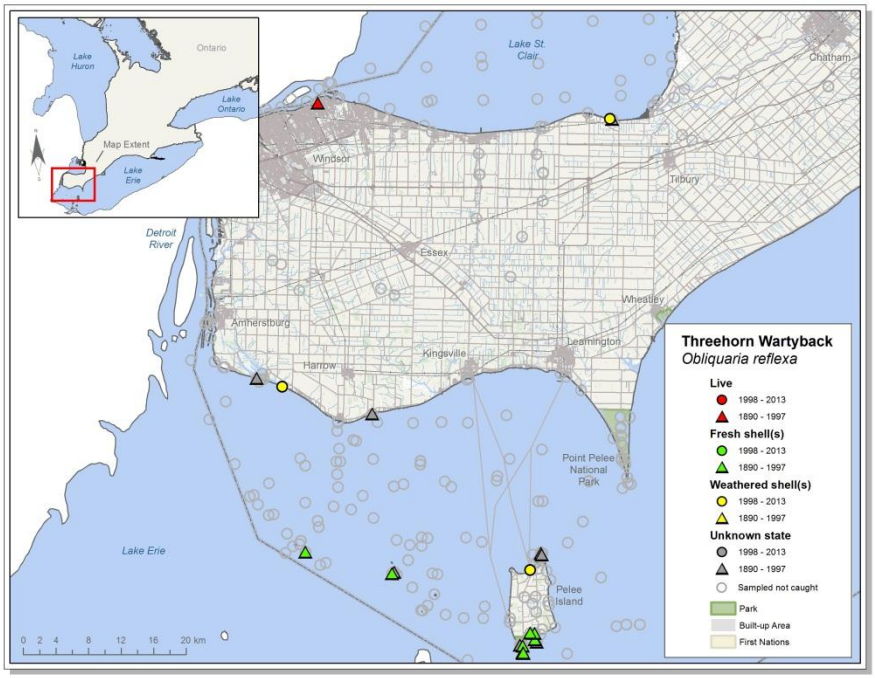

Figure 9. Distribution of all known current and historic Threehorn Wartyback records from the Canadian waters of the Great Lakes and connecting channels.

Population Status Assessment

For the purposes of this Recovery Potential Assessment, characteristics to be considered when delineating populations include movement of the individual mussel (including movement of the host fishes), availability of suitable habitat between two locations, state of the Threehorn Wartyback recorded, and date of the record. The putative host fishes for Threehorn Wartyback in Canada include Common Shiner and Longnose Dace. Distribution of these putative host fishes directly overlaps that of Threehorn Wartyback. These characteristics were used when determining the population structure used for the Population Status Assessment. Refer to Bouvier et al. (2014) for a thorough review of population categorization.

To assess the population status of Threehorn Wartyback in Canada, each population was ranked in terms of its abundance and trajectory. The level of certainty was associated with each assignment (1=quantitative analysis; 2=CPUE or standardized sampling; 3=expert opinion). The Abundance Index and Population Trajectory values were combined in the Population Status matrix to determine the Population Status for each population. Each Population Status was subsequently ranked as Poor, Fair, Good, Unknown or Extirpated (Table 1). The Certainty assigned to each Population Status is reflective of the lowest level of certainty associated with either initial parameter. Refer to Bouvier et al. (2014) for details on the methods used in the assessment of Population Status.

Table 1. Population Status of all Threehorn Wartyback populations in Canada, resulting from an analysis of both the Relative Abundance Index and Population Trajectory. Certainty assigned to each Population Status is reflective of the lowest level of certainty associated with either initial parameter (Relative Abundance Index, or Population Trajectory).

Population	Population Status	Certainty
Sydenham River	Poor	3
Thames River	Poor	3
Grand River	Poor	3

Habitat Requirements

Glochidium

To fully understand the habitat requirements of freshwater mussels, we must first understand their unique life cycle. Although Threehorn Wartyback are dioecious, they are believed to express very little sexual dimorphism. During the spawning period, males located upstream release sperm into the water column via the excurrent siphon. Females subsequently utilize their gills to filter the sperm from the water column, and the sperm is deposited in the posterior portion of the female gill in a specialized region where the ova are fertilized. The fertilized ova are held until they reach a larval stage. Haag and Staton (2003) noted that Threehorn Wartyback only brood their young in the outer pair of gills, in what appear to be modified water tubes located approximately in the middle of each of the outer gills. It was also observed that the individual eggs of Threehorn Wartyback were bound tightly within the gills. Threehorn Wartyback release their glochidia in a sausage-shaped conglutinate. For Threehorn Wartyback, the mean number of conglutinates per individual has been recorded to be 8.1 conglutinates/individual (± 0.6), while the mean fecundity was observed from one site in Alabama as 25,767 (3250-82,500=range of observations) and one site in Mississippi as 40,975 (447 – 135,750=range of observations).

Freshwater mussels are often categorized in terms of their brooding and glochidial release patterns. Two brooding strategies are long-term brooders (bradytictic) and short-term brooders (tachytictic). Threehorn Wartyback is classified as a short-term brooder, with glochidia being formed and released in May until the end of July. Collections of Threehorn Wartyback in May in June in Mississippi resulted in a high percentage of gravid females (97%). Gravid females have also been observed in the Sydenham River in June when water temperatures were approximately 20°C.

Regardless of brooding strategy, once females release their glochidia they must encyst on the gills of an appropriate host fish. Although it has been suggested that Threehorn Wartyback may not require a host fish to complete its life cycle, there has been no additional support for this suggestion in the literature. Glochidial mortality is currently unknown but it is estimated that as little as 0.001% of glochidia successfully attach to an appropriate host fish. Metamorphosis from glochidia to juvenile cannot occur without a period of encystement, which has been recorded to last 17-19 days post attachment.

Host fishes

Infestation experiments to determine host fish for Threehorn Wartyback in Canada have not occurred, but Common Shiner, Longnose Dace and Silverjaw Minnow (*Notropis buccata*) have been identified to be appropriate host fish in the United States. In addition, Barnhart and Baird (2000) recorded a natural infestation of Threehorn Wartyback on Goldeye (*Hiodon alosoides*). They concluded that this host association was highly probable as the glochidia were numerous and had grown while encysted. Distributional overlap with the extant range of Threehorn Wartyback in Canada does exist for Common Shiner and Longnose Dace, providing

circumstantial evidence of host fish interaction. A detailed account of host fish interactions for Threehorn Wartyback can be found at Bouvier et al. (2014).

Juvenile

Subsequent to metamorphoses, juvenile freshwater mussels are released from the gills of the host fish and burrow into the substrate until maturity. Time to maturity can vary from one mussel species to another and accurate estimates are not known for most species. It is difficult to classify required habitat for juvenile mussels because they are difficult to detect, as they have a tendency to burrow. Once sexually mature they emerge from the substrate to participate in gamete exchange. Threehorn Wartyback age at maturity is currently unknown.

Adult

Threehorn Wartyback is most commonly found in large rivers with stable gravel, sand and mud substrates with moderate current; however, it may also be found in shallow embayments and reservoirs with almost no current. It has been suggested that Threehorn Wartyback can tolerate a wide range of water temperatures, depths, substrates and flows. Additional details on adult habitat preferences are discussed in Table 2.

Residence

Residence is defined in SARA as “dwelling-place, such as a den, nest or other similar area or place, that is occupied or habitually occupied by one or more individuals during all or part of their life cycles, including breeding, rearing, staging, wintering, feeding or hibernating”. Residence is interpreted by DFO as being constructed by the organism (DFO 2010). In the context of the above narrative description of habitat requirements during glochidial, juvenile and adult life stages, Threehorn Wartyback does not construct a residence during its life cycle.

Functions, Features and Attributes

A description of the functions, features, and attributes associated with Threehorn Wartyback habitat can be found in Table 2. The habitat required for each life stage has been assigned a function that corresponds to a biological requirement of Threehorn Wartyback. In addition to the habitat function, a feature has been assigned to each life stage. A feature is considered to be the structural component of the habitat necessary for the survival or recovery of the species. Habitat attributes have also been provided, which describe how the features support the function for each life stage. Optimal habitat attributes from the literature for each life stage have been combined with habitat attributes from current records (recorded from 1997 to present) to show the maximum range in habitat attributes within which Threehorn Wartyback may be found (see Table 2 and references therein). This information is provided to guide any future identification of critical habitat for this species. It should be noted that habitat attributes associated with current records may differ from those presented in the scientific literature as Threehorn Wartyback may be currently occupying areas where optimal habitat is no longer available.

Table 2. Summary of the essential functions, features and attributes for each life stage of Threehorn Wartyback. Habitat attributes from published literature, and habitat attributes recorded during recent Threehorn Wartyback surveys (recorded since 1997) have been combined to derive the habitat attributes required for the delineation of critical habitat (see text for a detailed description of categories).

			Habitat Attributes		
Life Stage	**Function**	**Feature(s)**	**Scientific Literature**	**Current Records**	**For Identification of Critical Habitat**
Spawning and fertilization (short-term brooder: glochidia being formed and released in May until the end of July)	Reproduction	Large rivers with moderate flow		• Gravid female Threehorn Wartyback have been observed in the Sydenham River in June at temperatures of ~20°C (Castanon, pers. comm. 2011 in COSEWIC 2013)	• Same habitat as adult
Encysted glochidial stage on host fish until drop off	Development	Appropriate host fish(es)	• Infestation experiments have determined that Common Shiner, Longnose Dace, and Silverjaw Minnow are appropriate host fishes in United States [see Watters et al. 2009 for species-specific references] • There is a single record of a natural infestation of Threehorn Wartyback on the gills of Goldeye in Missouri, USA (Barnhart and Baird 2000)	• There are no records of natural or laboratory infestations of Threehorn Wartyback glochidia on gills of putative host fishes	• Presence of sufficient host fish (putative host fishes in Canadian waters are Common Shiner and Longnose Dace)
Adult/juvenile	Feeding Cover Nursery	Large rivers with moderate flow	**General** • Categorized as occupying large rivers with moderate current, and shallow embayments and reservoirs with almost no current (Metcalfe-Smith et al. 2005; Watters et al. 2009)	• General characteristics taken from the literature supported by recent reports of live individuals	
			Substrate • Threehorn Wartyback occupies areas with stable gravel, sand or mud substrates (Metcalfe-Smith et al. 2005) and areas with muddy sand, or cobble (Watters et al. 2009)	• The majority of sites where live Threehorn Wartyback were recorded were composed of a combination of boulder, rubble, gravel and sand (DFO, unpubl. data)	• Most often found in areas where the substrate is composed of boulder, rubble, gravel and sand, or a combination thereof

			Habitat Attributes		
Life Stage	**Function**	**Feature(s)**	**Scientific Literature**	**Current Records**	**For Identification of Critical Habitat**
			Depth • Threehorn Wartyback has been found in water as deep as 6-7 m in other parts of its range (Parmalee and Bogan 1998; Univeristy of Georgia Museum of Natural History 2013)	• A scuba diving survey of the lower Grand River in 1997 recorded one live Threehorn Wartyback in water 4 m deep, and another in 5 m (Metcalfe-Smith et al. 1998) • Sites where live Threehorn Wartyback have been recorded in Ontario had an average site water depth of 0.64 ± 0.22 m (DFO, unpubl. data)	
			Presence of dreissenid mussels • Introduction and establishment of dreissenid mussels has negatively affected freshwater mussels in the Great Lakes	• Zebra Mussel (*Dreissena polymorpha*) present at Thames River site (TR-50) in 2010 where live Threehorn Wartyback were recorded (DFO, unpubl. data) • Zebra Mussel present from the Fanshawe Reservoir in London, downstream to near Thamesville, and are likely present all the way to the river mouth (Morris and Edwards 2007) • Zebra Mussel present in the Grand River up to the Dunnville Dam (G. Mackie, pers. comm.)	

Population sensitivity to perturbation

There was insufficient information on the life history of Threehorn Wartyback in Canada to complete a population model of the species. For use in such data-poor scenarios, Young and Koops (2011) used a population matrix model framework to explore the sensitivity of Unionid mussel populations to perturbations.

Sensitivity was quantified using elasticities, which can be used to describe the expected percent change in the long-term population growth rate as a result of a percent change in a vital rate (Caswell 2001). A range of possible Unionid life histories were classified into groups with similar elasticities. It was found that sensitivity groups could be predicted if certain vital rates were known to be on either the high or the low end of the parameter range. Life histories were classified into the following groups:

- Reproduction dominant: population growth was most sensitive to perturbation or uncertainty in age at maturity; glochidial survival and fecundity were more influential in this group than in others.
- Adult survival dominant: adult survival influenced population growth much more than juvenile survival. Remaining vital rates were relatively less important.
- Juvenile survival dominant: population growth was most influenced by juvenile survival.

The maximum observed lifespan in Ontario is 18 years (COSEWIC 2013), which classifies it as a “short lived” species (values used in previous modelling were 10 years and 50 years for “low” and “high” values, respectively). It therefore must also have “early” age at maturity due to how maturity was defined in previous modelling. Mean fecundity of Threehorn Wartyback was found to be 40,975 in Massachusetts and 25,767 in Alabama, and did not exceed 136,000 in either state (Haag and Staton 2003). We therefore classify Threehorn Wartyback as having “low” fecundity. Using the classification system from Young and Koops (2011), Threehorn Wartyback falls into the adult survival dominant group. An updated version of this classification system (DFO, unpubl. data) also suggests that the species may fall into a fourth “low sensitivity” group. This group is similar to the adult survival dominant group but with lower sensitivity to adult survival (i.e., population growth is less sensitive to all vital rates compared to other groups). In this group, population growth is equally sensitive to changes in adult survival, juvenile survival, and lifespan.

Note that sensitivity analyses are meant to compare expected responses in population growth to changes in vital rate. Pertinent threats to the species may affect life stages not identified as being most sensitive to perturbation.

Threats to Survival and Recovery

A wide variety of threats negatively affect Threehorn Wartyback across its range. Our knowledge of threat impacts on Threehorn Wartyback populations is limited to general documentation, as there is a paucity of threat-specific cause and effect information in the literature.

The threats thought to have the largest effect on the survival and recovery of Threehorn Wartyback in Canada are largely attributed to the presence of contaminants and toxic substances in the environment, habitat removal and alteration, and the introduction and establishment of various invasive species. Decreases in the quality of freshwater mussel habitat resulting from increases in nutrient loading, and turbidity and sediment loading are currently affecting Threehorn Wartyback populations. In addition, due to the obligate glochidial

encystement stage, Threehorn Wartyback is directly affected by host fish abundance and indirectly affected by the threats affecting the host fish. It is important to note the threats discussed may not always act independently on Threehorn Wartyback populations; rather, one threat may directly affect another, or the interaction between two threats may introduce an interaction effect on Threehorn Wartyback populations. It is difficult to quantify these interactions and cumulative effects; therefore, each threat is discussed independently. Refer to Bouvier et al. (2014) for a thorough review of threats thought to be negatively affecting Threehorn Wartyback populations.

Threat Level Assessment

Each threat was ranked in terms of the Threat Likelihood and Threat Impact for all river systems where it is believed that a population of Threehorn Wartyback may exist (see Bouvier et al. 2014 for complete details on threat assessment approach). Threat Impact categorization was assigned on a location-by-location basis. If no information was available on the Threat Impact at a specific location, a precautionary approach was used - the highest level of impact from all sites was applied. The Threat Likelihood and Threat Impact for each population were subsequently combined in the Threat Status Matrix resulting in the final Threat Status for each location (Table 3). Certainty has been classified for Threat Impact and is based on: 1= causative studies; 2=correlative studies; and, 3=expert opinion [level of certainty listed from highest (1) to lowest (3)].

Table 3. Threat Level for Threehorn Wartyback populations, resulting from an analysis of both the Threat Likelihood and Threat Impact. The number in brackets refers to the level of certainty assigned to each Threat Level, which relates to the level of certainty associated with Threat Impact. Certainty has been classified as: 1= causative studies; 2=correlative studies; and 3=expert opinion.

Threat	Sydenham River	Thames River	Grand River
Contaminants and toxic substances	High (3)	High (3)	High (3)
Nutrient loading	Medium (3)	Medium (3)	High (3)
Turbidity	Medium (3)	Unknown (3)	Unknown (3)
Sediment loading	Medium (3)	Medium (3)	Medium (3)
Invasive species	Low (2)	High (2)	High (2)
Habitat removal and alteration	High (3)	High (3)	High (3)
Altered flow regimes	Low (3)	Low (3)	Medium (3)
Host fish (invasive species)	Unknown (3)	Unknown (3)	Unknown (3)

Mitigations and Alternatives

Threats to species survival and recovery can be reduced by implementing mitigation measures to reduce or eliminate potential harmful effects that could result from works or undertakings associated with projects, or activities in Threehorn Wartyback habitat. Threehorn Wartyback has been assessed as Threatened by COSEWIC and is not currently listed nor protected under the *Endangered Species Act,* 2007*.*

Within Threehorn Wartyback habitat, a variety of works, undertakings, and activities have occurred in the past few years with project types including: water crossings (e.g., bridge maintenance), shoreline and streambank works (e.g., stabilization), instream works (e.g., channel maintenance), and the placement or removal of structures in water. A review has been completed summarizing the types of work, activity, or projects that have been undertaken in habitat known to be occupied by Threehorn Wartyback (Table 4). The DFO Program Activity Tracking for Habitat (PATH) database, as well as summary reports of fish habitat projects reviewed by partner agencies (e.g., conservation authorities), have been reviewed to estimate the number of projects that have occurred during the three-year period, 2010-2012. Only 18 projects were identified in Threehorn Wartyback habitat, but likely do not represent a comprehensive list of activities that have occurred in these areas (Table 4). Some projects occurring in proximity but not in the area of habitat may also have impacts, but were not included. Some projects may not have been reported to partner agencies or DFO if they occurred under conditions of an Operational Statement. It was noted that seven were completed under conditions of Operational Statements primarily for bridge maintenance.

Only one project to replace the Cayuga bridge was authorized under the *Fisheries Act* and permitted under the SARA since a mussel relocation for other SARA species was required. No Threehorn Wartyback were found during the relocation. The remaining projects were also deemed low risk to fish and fish habitat and were addressed through letters of advice with standard mitigation. Without appropriate mitigation, projects or activities occurring adjacent or close to these areas could have impacted Threehorn Wartyback (e.g., increased turbidity or sedimentation from upstream channel works).

The most frequent project type (nine of 18) was for water crossings, which includes directional drilling for piping. Based on the assumption that historic and anticipated development pressures are likely to be similar, it is expected that similar types of projects will likely occur in or near Threehorn Wartyback habitat in the future. The primary project proponents were local municipalities.

As indicated in the Threat Analysis, numerous threats affecting Threehorn Wartyback populations are habitat-related threats that have been linked to the Pathways of Effects developed by DFO Fish Habitat Management (FHM) (Table 4). DFO FHM has developed guidance on mitigation measures for 19 Pathways of Effects for the protection of aquatic species at risk in the Central and Arctic Region (Coker et al. 2010). This guidance should be referred to when considering mitigation and alternative strategies for habitat-related threats. At the present time, we are unaware of mitigation that would apply beyond what is included in the Pathways of Effects.

Non-habitat related activities require additional discussion as these activities are not considered in the guidance on mitigation measures (Coker et al. 2010). Mitigation and alternative measures to invasive species and host fishes, as it relates to Threehorn Wartyback, are proposed.

Invasive Species

As discussed in the **THREATS** section, aquatic invasive species (e.g., dreissenid mussels, Round Goby and Common Carp) introduction and establishment may have a negative effect on Threehorn Wartyback populations. Mitigation and alternatives should not only be considered for current established invasive species but species that may invade in the future.

Mitigation

- Evaluate the likelihood that a waterbody will be invaded by an invasive species.
- Monitor watersheds for invasive species that may negatively affect Threehorn Wartyback populations directly, or negatively affect Threehorn Wartyback preferred habitat.
- Develop a plan to address potential risks, impacts, and proposed actions if monitoring detects the arrival or establishment of an invasive species.
- Introduce a public awareness campaign on proper boat cleaning methods when transferring boats from an infested waterway, and on the proper identification of native and invasive freshwater mussels. The public awareness campaign could include an educational fact sheet to better educate the public on native and invasive species.
- Encourage the use of existing invasive species reporting systems.
- Restrict the use of boats in areas particularly susceptible to Zebra Mussel introduction and infestation.

Alternatives

- Unauthorized
    - None.
- Authorized
    - Use only native species.
    - Follow the National Code on Introductions and Transfers of Aquatic Organisms for all aquatic organism introductions (DFO 2003).

Table 4. Summary of works, projects and activities that have occurred during the period of January 2010 to December 2012 in areas known to be occupied by Threehorn Wartyback. Threats known to be associated with these types of works, projects, and activities have been indicated by a checkmark. The number of works, projects, and activities associated with each Threehorn Wartyback population, as determined from the project assessment analysis, has been provided. Applicable Pathways of Effects have been indicated for each threat associated with a work, project or activity (1 - Vegetation clearing; 2 – Grading; 3 –Excavation; 4 – Use of explosives; 5 – Use of industrial equipment; 6 – Cleaning or maintenance of bridges or other structures; 7 – Riparian planting; 8 – Streamside livestock grazing; 9 – Marine seismic surveys; 10 – Placement of material or structures in water; 11 – Dredging; 12 – Water extraction; 13 – Organic debris management; 14 – Wastewater management; 15 – Addition or removal of aquatic vegetation; 16 – Change in timing, duration and frequency of flow; 17 – Fish passage issues; 18 – Structure removal; 19 – Placement of marine finfish aquaculture site).

Work/Project/Activity	**Threats** (associated with work/project/activity)						**Watercourse / Waterbody** (number of works/projects/activities between 2010-2012)		
	Contaminants and toxic substances	**Nutrient loading**	**Turbidity and sediment loading**	**Habitat removal and alteration**	**Altered flow regimes**	**Host fish (barriers to movement)**	**Sydenham River**	**Thames River**	**Grand River**
Applicable pathways of effects for threat mitigation and project alternatives	1,4,5,6, 7,11,12,13, 14,15,16,18	1,4,7,8, 11,12, 13,14,15,16	1,2,3 4,5, 6,7,8,10, 11,12,13, 15,16,18	1,2,3,4,5,7,8, 10,11,13,14, 15,16,18	10,16, 17	10,16, 17			
Water crossings (bridges, culverts, open cut crossings)	✓		✓	✓	✓	✓	4	2	3
Shoreline, streambank work (stabilization, infilling, retaining walls, riparian vegetation management)	✓		✓	✓	✓			1	1
Dams, barriers, structures in water (maintenance, modification, hydro retrofits)			✓	✓	✓	✓			
Instream works (channel maintenance, restoration, modifications, realignments, dredging, aquatic vegetation removal)	✓	✓	✓	✓	✓		2		1
Water management (stormwater management, water withdrawal)	✓	✓	✓		✓				
Structures in water (boat launches, docks, effluent outfalls, water intakes)	✓	✓	✓	✓	✓			3	1

Host Fishes

As discussed in the **THREATS** section, decreases in the number of individual host fish or decreases in the area of overlap between host fish and freshwater mussel may decrease the likelihood that a fish-mussel encounter will occur.

Mitigation

- Once the functional host fishes are confirmed for Threehorn Wartyback in Canadian waters, and if populations appear to be decreasing, a management plan for the appropriate host fish should be implemented. This would increase the host's survival, increasing the number of hosts available, creating a healthy host population and subsequently increasing the likelihood that the host fish would encounter a gravid freshwater mussel.
- The removal of barriers to host fish movement should be considered to allow increased host fish access to areas known to be inhabited by Threehorn Wartyback, if barriers to movement is deemed a limiting factor in the survival and recovery of Threehorn Wartyback.

Alternatives

- Enhance fish passage where barriers may be impeding the movement of host fishes.
- Artificially propagate host fish species where the abundance of host fish species is determined to be a limiting factor in the recovery or survival of Threehorn Wartyback.
- In areas where host fish species is abundant, artificial propagation of Threehorn Wartyback to enhance current populations should be explored.

Sources of Uncertainty

Despite concerted efforts to increase our knowledge of Threehorn Wartyback in Canada, there are still a number of key sources of uncertainty for this species related to population distribution, structure, habitat preferences and to the factors limiting their existence.

There is a need for a continuation of quantitative sampling of Threehorn Wartyback in areas where it is known to occur to determine population size, current trajectory, and trends over time. There is also a need for additional targeted sampling in the Grand River, as very few current records of live individuals exist for this system. Exploratory sampling should be completed in systems with habitat characteristics similar to those areas where Threehorn Wartyback is known to occur to determine the extent of their distribution. Sampling of rarely sampled deep water habitat should also occur to determine if Threehorn Wartyback are occupying these areas. In addition, supplementary sampling is necessary for all populations that were assigned a low certainty in completing the population status assessment. As is now common practice, shell length of all live individuals should be recorded to gain information on population structure and to understand recruitment within each population. These baseline data are required to monitor Threehorn Wartyback distribution and population trends as well as the success of any recovery measures implemented.

Additional studies on habitat requirements are imperative to determine critical habitat for all Threehorn Wartyback life stages. Laboratory experiments, and if feasible field experiments, should be completed to determine the functional host fish of Threehorn Wartyback in Canada. Currently, putative host fish species are inferred from infestation experiments in the United States. Infestation experiments, using samples from Canadian populations, should be completed to verify the usage of Common Shiner and Longnose Dace as host fishes for Threehorn Wartyback. Sampling of putative host fish should be completed in areas known to be inhabited by Threehorn Wartyback, during which the gills should be inspected and sampled for

Threehorn Wartyback glochidia. This may aid in determining the host fish from a natural infestation. Once host fish species have been confirmed, additional investigations to determine the glochidial carrying capacity, as well as the relationship between mussel attachment probability and host-mussel density should be completed.

The largest barrier preventing accurate population modelling of Threehorn Wartyback is a lack of sufficient length- or age-frequency data. Small sample size is often a challenge when studying rare species and existing sample sizes are insufficient to perform catch curve analyses for estimation of adult survival. In addition, very little is known about glochidial attachment and survival of Unionids in general.

Numerous threats have been identified for Threehorn Wartyback populations in Canada, although the direct impact that these threats may have is currently unknown. There is a need for more quantitative studies to evaluate the direct impact of each threat on Threehorn Wartyback populations with greater certainty. In the literature, the threat impacts are generally discussed at a broad level (i.e., mussel assemblage level). It is important to further our knowledge on threat likelihood and impact at the species level. Research is needed to determine the effect of contaminants and toxic substances on Threehorn Wartyback, as these pollutants are known to occur in areas where Threehorn Wartyback is currently found. This type of research would provide insight on the factors currently limiting Threehorn Wartyback populations. Thresholds for other water quality parameters (e.g., nutrients, turbidity) should also be investigated.

SOURCES OF INFORMATION

This Science Advisory Report is from the December 10, 2013 Recovery Potential Assessment of Threehorn Wartyback (*Obliquaria reflexa*). Additional publications from this meeting will be posted on the Fisheries and Oceans Canada (DFO) Science Advisory Schedule as they become available.

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