



# SCIENCE ADVICE ON THE POTENTIAL HARM OF GRANULAR BAYLUSCID E APPLICATIONS TO FISH AND MUSSEL SPECIES AT RISK

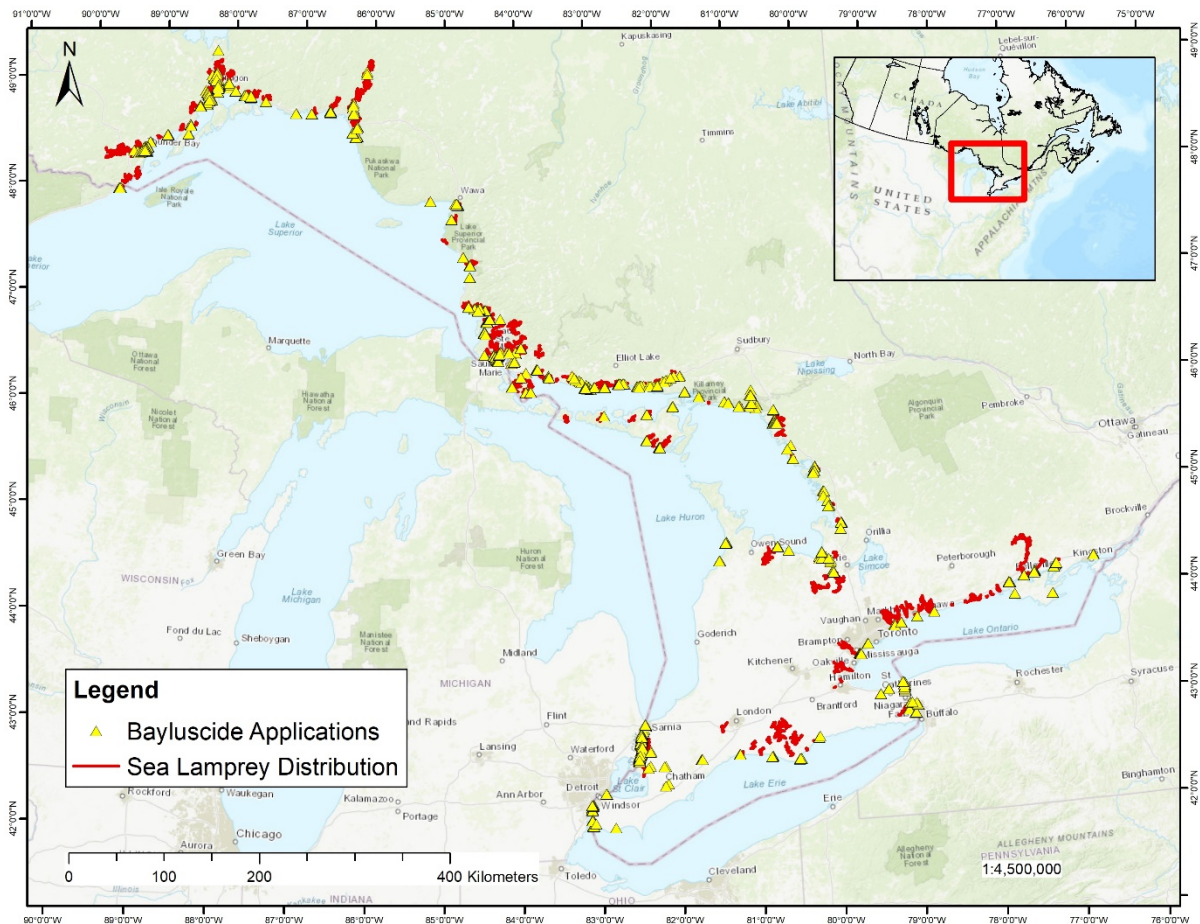


Figure 1. Granular Bayluscide applications (2011–2017) and the distribution of larval Sea Lamprey (based on 2011 to 2017 collections) in the Canadian waters of the Great Lakes basin.

### Context:

*Sea Lamprey (Petromyzon marinus), an invasive fish species in the Great Lakes basin, has caused widespread and significant mortality on fishes that support Indigenous, commercial, and recreational fisheries. To reduce Sea Lamprey abundance and ensure the ongoing productivity of fisheries, the Great Lakes Fishery Commission coordinates the bi-national Sea Lamprey Control Program, implemented by Fisheries and Oceans Canada (DFO) and the United States Fish and Wildlife Service (USFWS). Assessing the response of Sea Lamprey to control measures is a core component of the control program. Under certain conditions, control and assessment of Sea Lamprey is conducted via the application of granular Bayluscide, a chemical lampricide, within tributary streams or lake areas*

*within the basin. In some instances, Bayluscide has been applied in waterbodies that contain fish and mussel species currently listed under the Species at Risk Act (SARA) as well as other species of conservation concern. As a result of these applications, DFO's Species at Risk Program has requested science advice to understand the potential impacts of Bayluscide on fish and mussel species of conservation concern, and to identify best management practices and potential mitigation measures to minimize impacts. Therefore, the goal of this Science Advisory Meeting was to evaluate the potential lethal and sub-lethal impacts of Bayluscide applications to fish and mussel species of conservation concern in Canadian waters of the Great Lakes basin.*

*This Science Advisory Report is from the February 28–March 1, 2019 regional peer review meeting on Information on the Potential Harm to Fish and Mussel Species at Risk (SAR) from Bayluscide Applications. Additional publications from this meeting will be posted on the [Fisheries and Oceans Canada \(DFO\) Science Advisory Schedule](#) as they become available.*

## SUMMARY

- Granular Bayluscide (gB), a chemical lampricide, is applied in certain areas of the Great Lakes basin to assess and control Sea Lamprey (*Petromyzon marinus*) populations. Applications of gB have occurred since 1966 and are part of bi-national efforts to control the species, coordinated by the Great Lakes Fishery Commission and led by Fisheries and Oceans Canada and the U.S. Fish and Wildlife Service.
- The request for science advice resulted from gB applications in areas inhabited by fishes and mussels listed as Endangered, Threatened, or Special Concern under Canada's *Species at Risk Act* (SARA), raising questions about the ecological effects of the lampricide on fishes and mussels of conservation concern.
- Fishes and mussels exposed to gB in the aquatic environment may experience sub-lethal or lethal effects due to the toxicity of the chemical including changes in growth, movement, reproduction, or survival. Bayluscide may also lead to changes in ecosystem components (prey, competitors, predators, host species) resulting in indirect effects.
- A risk assessment was undertaken to determine the relative risk of direct mortality stemming from gB applications on 24 fishes (21 with SARA status) and 15 mussels (all with SARA status) in the Canadian waters of the Great Lakes basin. A modelling study was conducted to understand potential mortality from a typical gB application cycle and the population consequences of repeated applications, focusing on applications in the Huron-Erie corridor and nearby tributaries.
- Between 2011 and 2017, gB applications (Figure 1) occurred within the distribution of 21 fish and all 15 mussel species of conservation concern, including areas that contain or previously contained critical habitat for six SARA-listed fishes and 10 SARA-listed mussels. Granular Bayluscide exposure is relevant for up to 27% of a species' range and for less than 30% of gB application sites.
- For fishes, the relative risk of direct mortality was greatest for Silver Lamprey (*Ichthyomyzon unicuspis*), followed by Northern Brook Lamprey (*Ichthyomyzon fossor*), Lake Sturgeon (*Acipenser fulvescens*), and Northern Madtom (*Noturus stigmosus*). Relative risk for mussels was greatest for Salamander Mussel (*Simpsonaias ambigua*), Threehorn Wartyback (*Obliquaria reflexa*), and Hickorynut (*Obovaria olivaria*). Relative risk was based on historical patterns of gB applications. Deviations from historical application patterns will change the interpretation of relative risk.
- Modelling indicated that many fish and mussel species of conservation concern can experience direct mortality from gB applications due to their occurrence within application

sites and the assumed toxic effects of the compound. Although the likelihood of mortality is generally low, mortality on the order of one to tens of non-target fishes and potentially hundreds of native lampreys and mussels can occur. Repeated applications of gB can lead to population consequences for certain fish species.

- Changes to the size, number, and frequency of gB applications has model-based support to reduce mortality imposed by gB. Other mitigation measures exist, such as reducing target concentrations of gB, applying gB to areas outside of critical habitat, and seasonal application of gB outside of reproductive periods. Mitigation measures, if pursued, should be empirically tested to ensure intended benefits for species of conservation concern are realized.

## BACKGROUND

Sea Lamprey (*Petromyzon marinus*), a fish species native to the Atlantic Ocean, was first observed in Lake Ontario in 1888 and invaded the remaining Great Lakes between 1921 and 1937 following modifications to the Welland Canal (Smith and Tibbles 1980, Eshenroder 2014). Sea Lamprey caused widespread and significant mortality on fishes that support Indigenous, commercial, and recreational fisheries, including Lake Trout (*Salvelinus namaycush*), Lake Whitefish (*Coregonus clupeaformis*), ciscoes (*Coregonus* spp.), and numerous other species. Early efforts to control Sea Lamprey led Canada and the United States to form the Great Lakes Fishery Commission (GLFC) in 1955, under the auspices of the *Great Lakes Fishery Convention Act*. Since then, the Commission has administered the integrated Sea Lamprey Control Program (SLCP) in cooperation with Fisheries and Oceans Canada (DFO), the U.S. Fish and Wildlife Service, and the U.S Army Corps of Engineers with the goal to reduce Sea Lamprey populations in the Great Lakes to levels that maintain or improve fisheries (Great Lakes Fishery Commission 1956).

Several tactics exist to control Sea Lamprey in natal streams, ranging from purpose-built barriers and traps to the application of chemical lampricides. Evaluating the effect of control requires that Sea Lamprey populations be assessed on a recurring basis to determine population responses to control, including whether additional control sites should be considered. Assessment of Sea Lamprey populations to inform the control program involves sampling the depositional zone of nursery streams and other areas supporting Sea Lamprey production (e.g., connecting channels, certain lake areas) to determine the incidence and abundance of larval Sea Lamprey. A standardized habitat classification exists to guide assessment activities, which focus on habitat preferred by larval Sea Lamprey (Type I – composed primarily of silt substrates) or habitat used by larvae but not preferred (Type II – composed primarily of sand substrates), while avoiding habitat that is unsuitable for burrowing due to larger substrates like cobble or bedrock (Type III; see Slade et al. 2003 and Smyth and Drake 2021 for detailed description of habitat classes).

The primary method to assess larval Sea Lamprey involves backpack electrofishing, which is used in wadeable streams. However, in some cases, deep (> 0.8 m) or turbid waters require alternate assessment methods to detect larvae such as the application of chemical lampricides. A chemical compound composed of 2', 5-dichloro-4'-nitrosalicylanilide or niclosamide ethanolamine salt (trade name Bayluscide; Dawson 2003) is regularly used for this purpose in granular formulation (hereafter, gB) containing 3.2% active ingredient. During application the granules are applied to plots  $\leq 500 \text{ m}^2$ , at a rate of 156 lbs/acre (175 kg/hectare) to achieve a Bayluscide concentration of 11 mg/L (9.3mg/L active ingredient niclosamide [Adair and Sullivan 2004, Larval Assessment Task Force 2012]). In some cases, application of gB may be used as a control tactic in deepwater habitats (e.g., St. Mary's River) where conventional applications of

the lampricide TFM (3-trifluoromethyl-4-nitrophenol; Hubert 2003) would be ineffective or overly costly.

The use of gB in the Great Lakes basin has been highly successful in detecting and suppressing larval Sea Lamprey populations and remains an important component of the bi-national control program. However, given the known toxicity of gB to non-target species (Dawson 2003, Boogaard et al. 2016, Newton et al. 2017), concern exists about the potential for direct and indirect effects on fish and mussel species of conservation concern within the Canadian waters of the Great Lakes basin. Based on the general effects of toxicant exposure identified in the literature, Bayluscide applications have the potential to influence fish and mussel species of conservation concern through various pathways, both directly and indirectly (Figure 2). Direct effects, defined as those acting principally on focal species, may include changes to vital rates such as mortality, growth, reproductive potential, and movement/migration, which can influence production (including trajectory, abundance, persistence) of the species in question. Indirect effects, defined as those acting on food-web components that interact with focal species, may impact the vital rates of prey, predators, and competitors, leading to additional Bayluscide-induced responses. A unique case of indirect effects may exist for freshwater mussels where changes to the vital rates of host fishes, which are required to complete an obligate parasitic life stage, may alter reproductive potential. Although previous studies have evaluated the toxicity of gB to a range of non-target organisms (Marking and Hogan 1967, Bills and Marking 1976, Gilderhus 1979, Scholefield and Seelye 1992), impacts to species of conservation concern in Canada have not been widely assessed.

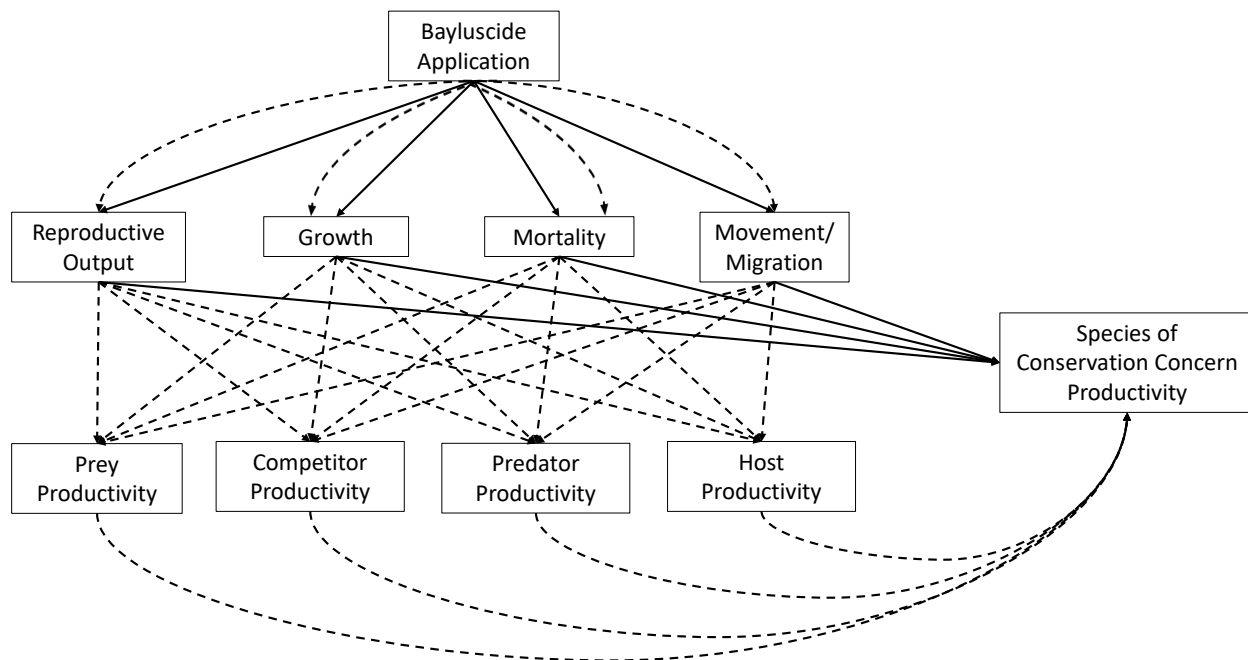


Figure 2. Bayluscide pathways of effects for species of conservation concern, including direct (solid lines) and indirect (dashed lines) pathways. Both direct and indirect pathways may influence the productivity of species of conservation concern through physiological (e.g., toxicity) and non-physiological (e.g., avoidance) mechanisms.

In 2011 and 2012, concerns were raised by managers within DFO’s Species at Risk Program regarding the application of gB in several areas of southwestern Ontario inhabited by fish and mussel species listed under Canada’s *Species at Risk Act* (SARA), including Lake St. Clair and the Detroit, St. Clair, Sydenham, and Thames rivers. As a result, staff from DFO’s Species at

Risk Program, DFO's Sea Lamprey Control Centre (SLCC), and the Great Lakes Laboratory for Fisheries and Aquatic Sciences (GLLFAS) identified the need to better understand the ecological risk of gB applications for species assessed by the Committee on the Status of Endangered Wildlife in Canada (COSEWIC) as Endangered, Threatened, or Special Concern as well as those listed as Endangered, Threatened, or Special Concern under SARA.

Scientific advice about the ecological risk of gB applications on SARA-listed species is needed for several reasons. First, DFO's Species at Risk Program is responsible for identifying threats to SARA-listed species, or species under consideration for listing, within the federal Species at Risk recovery planning framework which requires an understanding of how the risk of gB (if any) compares to other pertinent threats. Threat identification and evaluation (DFO 2014) is used to guide the development of federal recovery strategies, noting research and recovery actions needed to mitigate key threats. Second, Section 73(3)(c) of SARA stipulates that the competent minister may grant SARA permits for works/undertakings/activities (w/u/a) "...only if the competent minister is of the opinion that the activity will not jeopardize the survival or recovery of listed species", raising into question the ecological effects of gB from a regulatory perspective.

The first research component of this science advisory process focused on evaluating the relative risk of gB applications to fishes and mussels of conservation concern throughout the Canadian waters of the Great Lakes basin. Evaluating relative risk focused on the direct physiological pathway based on four lines of evidence: 1) distribution and 2) intensity of past gB applications in relation to the distribution of fishes and mussels of conservation concern; 3) habitat associations that predispose fish and mussel species to direct exposure within application sites; and, 4) the toxicity of gB to fish and mussel species, including surrogate species where appropriate.

The second research component identified the absolute risk of direct, gB-induced mortality to fishes and mussels of conservation concern associated with gB applications in the Huron-Erie corridor and nearby tributaries. Specifically, the potential for fishes and mussels of conservation concern to be exposed to and experience individual and population-level mortality from gB was quantified for the Detroit, St. Clair, Sydenham, and Thames rivers. Although this analysis did not incorporate mussels in the St. Clair and Detroit rivers, it has since been confirmed that mussel species at risk do inhabit the Detroit River (e.g., Three-horned Wartyback [*Obliquaria reflexa*], Mapleleaf [*Quadrula quadrula*], and Round Pigtoe [*Pleurobema sintoxia*]; see Allred et al. 2020). This research component evaluated the potential for altered population dynamics for select species of conservation concern. A quantitative assessment of the effect of potential mitigation measures (changes to plot size, number, and the frequency of application) was also conducted.

Mitigation measures have been identified that may reduce the scope for direct and indirect effects during gB applications should risks be deemed non-negligible for fishes and mussels of conservation concern.

## **ASSESSMENT**

### **Relative Risk Assessment for the Canadian Waters of the Great Lakes basin**

A risk assessment was developed to evaluate the relative ecological risk of direct mortality to fishes and mussels of conservation concern stemming from physiological effects. The risk assessment was based on four metrics including: 1) the proportion of the species range that has experienced gB applications; 2) the intensity of applications within the species range; 3) habitat associations that predispose species to exposure (i.e., preferential occupancy within Type I or Type II habitat); and, 4) standardized toxicity of Bayluscide to fishes and mussels based on

focal or surrogate species. Individual metrics, which were derived between 0 and 1 (described below), were selected as the most reasonable proxy variables to describe relative differences in the likelihood of exposure (distribution, intensity, habitat associations) and toxicity among species of conservation concern in the Great Lakes basin. The relative risk of direct mortality,  $RR_M$ , was calculated as  $RR_M = R \times I \times H \times T$ , where  $R$  represents the species range variable,  $I$  represents application intensity,  $H$  represents habitat associations, and  $T$  represents toxicity. Multiplication was used to estimate  $RR_M$  due to the conditional nature of each contributing process. Untransformed values of  $RR_M$  range between 0 (low relative risk) and 1 (high relative risk) with values close to 1 reflecting a species with high distributional overlap, high relative application intensity, high preference for Type I or Type II habitat, and high relative toxicity to Bayluscide. Because of uncertainty about how these factors influence direct mortality, equal weighting among variables  $R$ ,  $I$ ,  $H$ , and  $T$  was assumed to be the most reasonable method to estimate relative risk. However, alternative weightings are possible. Individual variables ( $R$ ,  $I$ ,  $H$ , and  $T$ ) provide stand-alone components that may be used to evaluate specific factors about the relative risk of Bayluscide applications. For example, omitting  $R$  and  $I$  from the relative risk equation would provide the relative risk of direct mortality of species of conservation concern at a known application site.

## Estimating Direct Mortality in the Huron-Erie Corridor and Nearby Tributaries

### Species Exposure to gB

Estimating species exposure considered the potential for each species to exist in the habitats selected for gB application and the corresponding density of each species. Likelihood of occurrence and density were estimated for each species within a focal river (e.g., Thames, Sydenham, Detroit, St. Clair), with the exception of Lake Sturgeon and native lampreys, which were calculated using different methods given available data (Smyth and Drake 2021). The likelihood of occurrence and density of mussels was not estimated for the Detroit and St. Clair rivers as current distribution data were not available for those systems.

### Calculating gB Concentration and Species-Specific gB Toxicity

The target concentration of gB and its duration in the aquatic environment is not known with certainty as the protocol to apply gB is based on weight of compound per unit area (175 kg of gB per hectare) (USFWS and DFO 2016). Therefore, concentration benchmarks were derived based on the concentration of Bayluscide that would lead to 50% and 99.9% mortality of Sea Lamprey over the course of a 9 hour exposure. Concentration benchmarks were combined with two dose-response curves generated from surrogate LC50 values to estimate mortality for each fish species of conservation concern at each concentration. Mussel mortality was estimated previously by Newton et al. (2017) where target concentration of gB was assumed to be 11 mg/L. This concentration was substantially higher than LC50 values estimated for numerous fishes (e.g., Marking and Hogan 1967, Dawson 2003). Given this discrepancy, the concentration and corresponding mortality for mussels was taken directly from Newton et al. (2017), so results for fishes and mussels are not directly comparable (Smyth and Drake 2021).

### Estimating Mortality from gB Applications

The number of individuals experiencing mortality from a single Bayluscide application cycle (i.e., six 500 m<sup>2</sup> application sites) was estimated based on the likelihood of a species of conservation concern occurring within a single application site, species density at a site, and estimated Bayluscide-induced mortality. These components were combined within a decision tree framework where the output of a single path through the tree represented the potential mortality of a species of conservation concern in a focal river following the application of Bayluscide at a single 500 m<sup>2</sup> site (Figure 3). The total mortality of a species within a focal river following a

single Bayluscide application cycle was based on the sum of the results of six paths through the decision tree, representing applications at six sites.

### **Sensitivity Analyses**

To understand how changes to the gB application cycle would lead to different mortality estimates, the number and size of gB application sites relative to the standard application cycle (i.e., six 500 m<sup>2</sup> application sites) were modified during sensitivity analysis. In addition, an alternative approach to estimate the density of species of conservation concern was examined to determine if underestimates of species density, as may be expected with imperfectly-detected field collection records, influenced the results. Descriptions for these assessments are provided in Smyth and Drake (2021).

### **Calculating Population-Level Effects from gB**

Population-level effects were assessed for select species including Eastern Sand Darter (*Ammocrypta pellucida*) in the Thames River, Northern Madtom (*Noturus stigmosus*) in the Thames and Detroit rivers, Channel Darter (*Percina copelandi*) in the Detroit River, and *Ichthyomyzon* spp. in the Thames and St. Clair rivers. Population-level effects were only assessed for fishes as population models for mussels are limited in scope.

To convert the species- and tributary-specific mortality estimates from a gB application cycle (i.e., a site specific mortality rate given  $n$  fishes present and  $n$  fishes killed) to population-level mortality rates, the population abundance of each focal species (i.e., total population size in a given focal river) was calculated. The calculation incorporated the species' system-specific density and either: the area of recognized or proposed critical habitat, if available for a SARA Threatened or Endangered species; or, the area bounding the species' recorded distribution within a study system if critical habitat had not been defined. Since the suitability of the critical habitat polygon or bound area is unknown for most species (i.e., not all areas within the habitat polygon may support individuals of the species), a habitat correction factor was incorporated that estimated the resulting population size and concordant effect of gB applications if 0.01% (very little suitable habitat) to 100% (maximum suitable habitat) of the habitat polygon supported the species. The population models represented a worst-case application scenario in which species of conservation concern did not possess the ability to recover following gB applications.



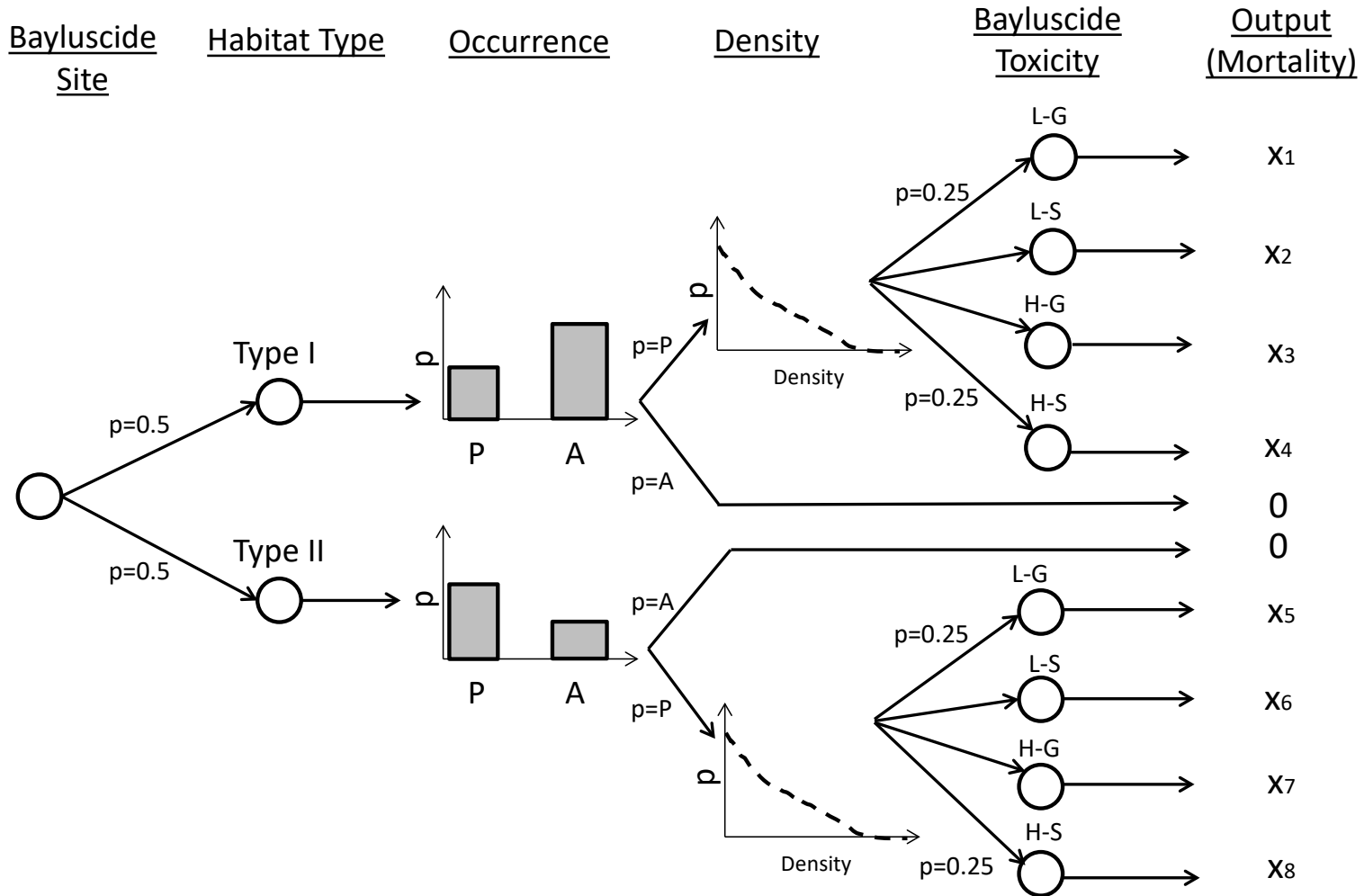


Figure 3. Decision tree used to calculate the mortality of fish species of conservation concern during gB applications. The diagram displays the outputs ( $X_n$ ) of all potential pathways through the uncertainties surrounding habitat type (Type I or Type II), occurrence (P – present or A – absent), density, and Bayluscide toxicity (L-G – low concentration, gentle dose-response slope; L-S – low concentration, steep dose-response slope; H-G – high concentration, gentle dose-response slope; H-S – high concentration, steep dose-response slope). The corresponding probabilities for each uncertainty state ( $p$ ) provides information regarding the probability of each output occurring.



## Results

The evaluation of the potential harm of gB to fishes and mussels of conservation concern in the Canadian waters of the Great Lakes basin revealed several insights, including knowledge that: 1) a gradient of relative risk exists across species based on gB application locations, habitat features, and toxicity; 2) gB applications occurred within the range of 36 fish and mussel species of conservation concern (including within areas currently or previously identified as critical habitat for 6 fishes and 10 mussels) in the Great Lakes basin from 2011 to 2017; 3) gB exposure is relevant for up to 27% of a species' range and for nearly 30% of gB application sites; 4) gB applications are not expected to pose direct mortality in most cases, but high mortality is possible under certain conditions, especially for native lampreys and SARA-listed freshwater mussels, and to a lesser extent for SARA-listed fishes; 5) native lampreys and some SARA-listed fishes could experience long-term population declines in certain systems; and, 6) mitigation measures, such as changes to application site size and application frequency, have model-based support to reduce the likelihood of extreme mortality events. These results are expanded upon below.

### Relative Risk Assessment

The highest values of  $R$  (proportion of range overlap) for fishes were Lake Sturgeon (0.261), Silver Lamprey and unidentified *Ichthyomyzon* sp. (0.251; Figure 4), Silver Lamprey (excluding unidentified *Ichthyomyzon* sp.; 0.246), and River Darter (*Percina shumardi*; 0.222), and for mussels were Salamander Mussel (*Simpsonaias ambigua*) and Hickorynut (*Obovaria olivaria*) (0.167; Figure 5), Fawnsfoot (*Truncilla donaciformis*; 0.133), and Threehorn Wartyback (0.13). These results indicated that the maximum proportion of the range overlapped by gB applications was as high as 27% but in many cases was much less. Further spatial analyses identified the proportion of gB sites within the range of species of conservation concern. These values were greatest for Silver Lamprey and unidentified *Ichthyomyzon* sp. (0.284), and Northern Brook Lamprey and unidentified *Ichthyomyzon* sp. (0.220), and for Rainbow (*Villosa iris*; 0.023) and Eastern Pondmussel (*Ligumia nasuta*; 0.018), indicating that up to 30% of gB applications can be in proximity to a species of conservation concern.

Intensity values ( $I$ ; the intensity of gB applications within a species range) for fishes were highest for Channel Darter (1.0; Figure 4), Northern Brook Lamprey (0.843), Northern Madtom (0.538), Black Redhorse (*Moxostoma duquesnei*; 0.517), and for mussels were highest for Rainbow (0.733; Figure 5), and Eastern Pondmussel (0.425). Differences in intensity indicated that certain species will experience a greater frequency of gB exposure.

Habitat association values,  $H$ , representing the likelihood that each species occupies Type I or Type II habitat relative to Type III habitat, were highest for the four native lamprey species groupings and Silver Chub (*Macrhybopsis storeriana*) (1.0; Figure 4). Habitat values were also very high for Bridle Shiner (*Notropis bifrenatus*; 0.970), Lake Chubsucker (*Erimyzon sucetta*; 0.944), and Pugnose Shiner (*Notropis anogenus*; 0.937). For mussels, habitat values were the highest for Eastern Pondmussel (1.0; Figure 5), Lilliput (*Toxolasma parvum*; 1.0), Rayed Bean (*Villosa fabalis*; 0.884), and Hickorynut (0.840) indicating that for some species the association with habitats potentially classified as Type I or Type II can be up to 100%.

Toxicity values,  $T$ , were highest for native lampreys (0.972; Figure 4), followed by American Eel (*Anguilla rostrata*; 0.632), Lake Sturgeon (0.532), and Northern Madtom (0.532), though each rating was based on surrogate assignments. Toxicity values can be interpreted as the mortality that would occur when exposed to Bayluscide concentrations of 0.057 mg/L over nine hours (e.g., 97.2% mortality for native lampreys). For mussel species, the highest toxicity values were approximately 54% at a Bayluscide concentration of 11 mg/L (active ingredient 9.3 mg/L) over eight hours. Note results for fishes and mussels are not directly comparable due to different

concentration benchmarks and assessment methods. Many mussel species shared the highest toxicity score (Rayed Bean, Northern Riffleshell [*Epioblasma rangiana*], Lilliput, Kidneyshell [*Ptychobranthus fasciolaris*]) of  $T = 0.54$  (Figure 5) due to surrogate assignments.

The overall relative risk of mortality,  $RR_M$ , was greatest in the four native lamprey species groupings (Silver Lamprey [0.105], Northern Brook Lamprey [0.080], Northern Brook Lamprey and unidentified *Ichthyomyzon* sp. [0.078]; Silver Lamprey and unidentified *Ichthyomyzon* sp. [0.075]) followed by Lake Sturgeon (0.034; noting methodological differences in the calculation of  $R$  for the species; see Andrews et al. 2021), and Northern Madtom (0.030; Figure 6). However, most SARA-listed fishes and those assessed by COSEWIC as Endangered, Threatened, or Special Concern exhibited non-zero relative risk values. Relative risk estimates were sensitive to the surrogate values assumed (Figure 6). The native lamprey species ranked highly given their high habitat and toxicity values (1.000 and 0.972, respectively). Lake Sturgeon ranked highly due to high spatial, intensity, and habitat values. However, because lake records of Lake Sturgeon were not incorporated, spatial and intensity scores for Lake Sturgeon are likely inflated thereby increasing the proportion of the range deemed susceptible to gB (similar issues exist for American Eel). The relative risk assessment for mussels indicated that Salamander Mussel had the greatest relative risk of mortality (0.0128) followed by Threehorn Wartyback (0.0072) and Hickorynut (0.0065; Figure 7). Salamander Mussel and Threehorn Wartyback exhibited high relative risk due to their very high spatial values, reflecting high overlap between past gB applications and the species' range as well as high toxicity. A high relative risk ranking for Hickorynut was driven by a high habitat association value and high potential spatial overlap with gB applications in comparison to other mussel species.

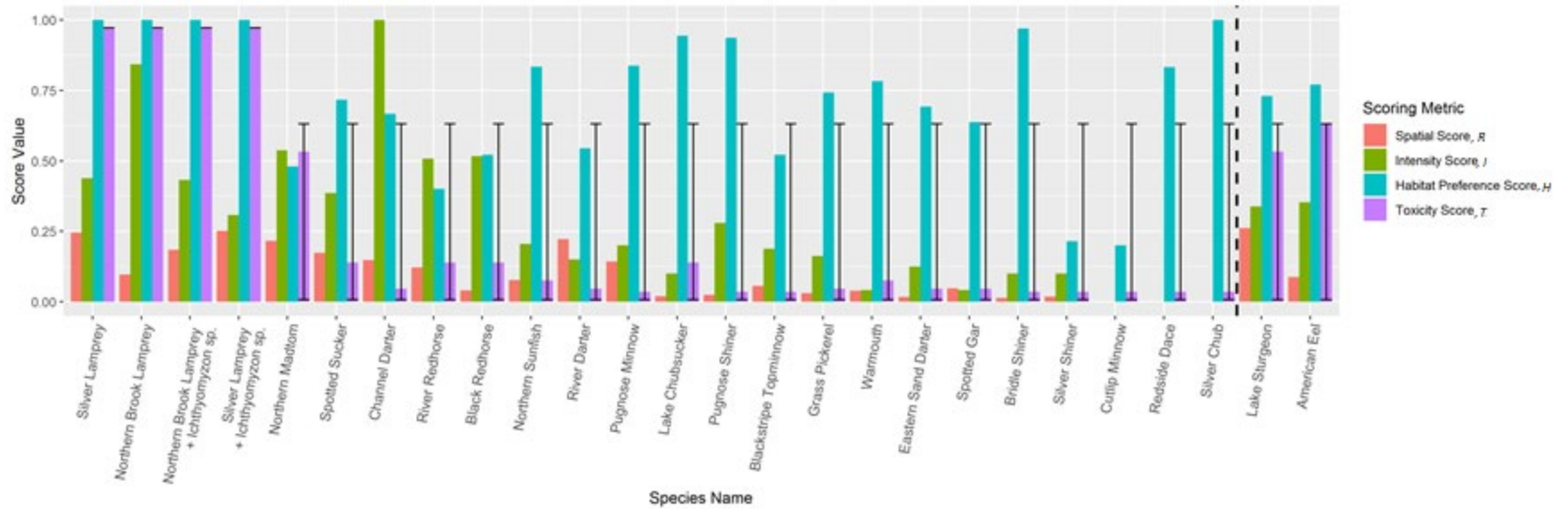


Figure 4. Relative risk assessment metrics ( $R$ ,  $I$ ,  $H$ ,  $T$ ) for fish species of conservation concern. The species order reflects overall relative risk,  $RR_M$ , from highest relative risk on the left to lowest on the right. Error bars on non-lamprey species represent the highest and lowest toxicity value based on all known non-lamprey fish surrogates. Lake Sturgeon and American Eel are separated by a dotted line and are not presented in rank order as their spatial and intensity values are not directly comparable with other fishes.

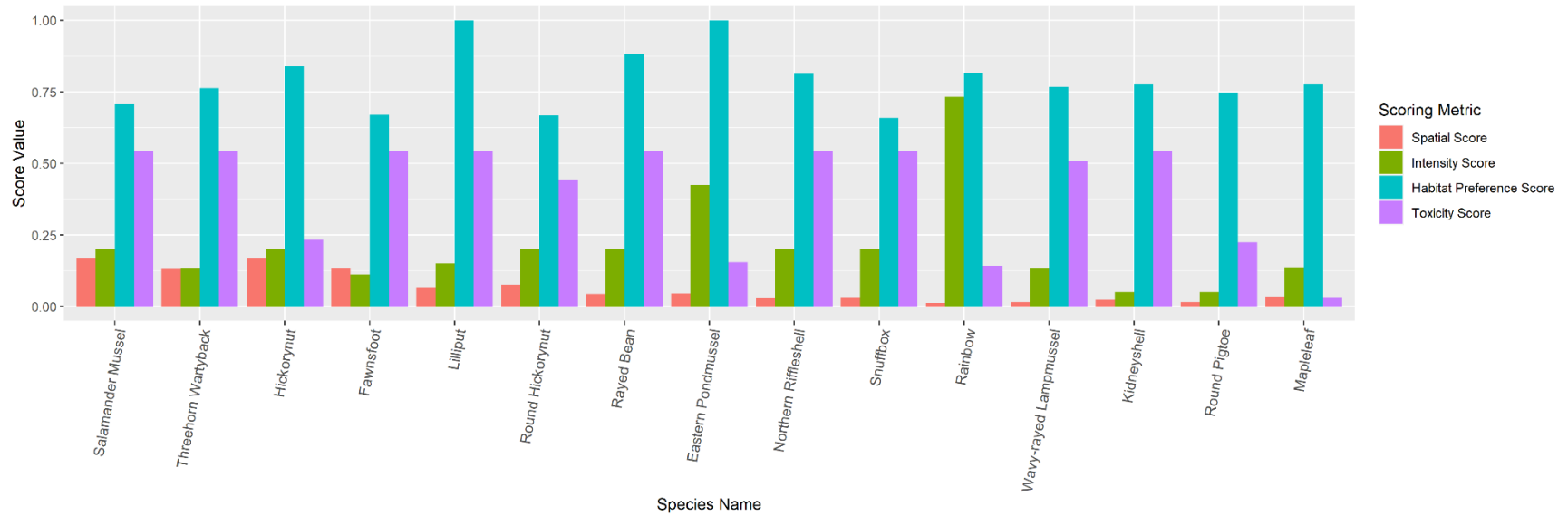


Figure 5. Relative risk assessment metrics (R, I, H, T) for mussel species of conservation concern. The species order reflects the rankings of the relative risk assessment values,  $RR_M$ , from highest relative risk on the left and lowest relative risk on the right.

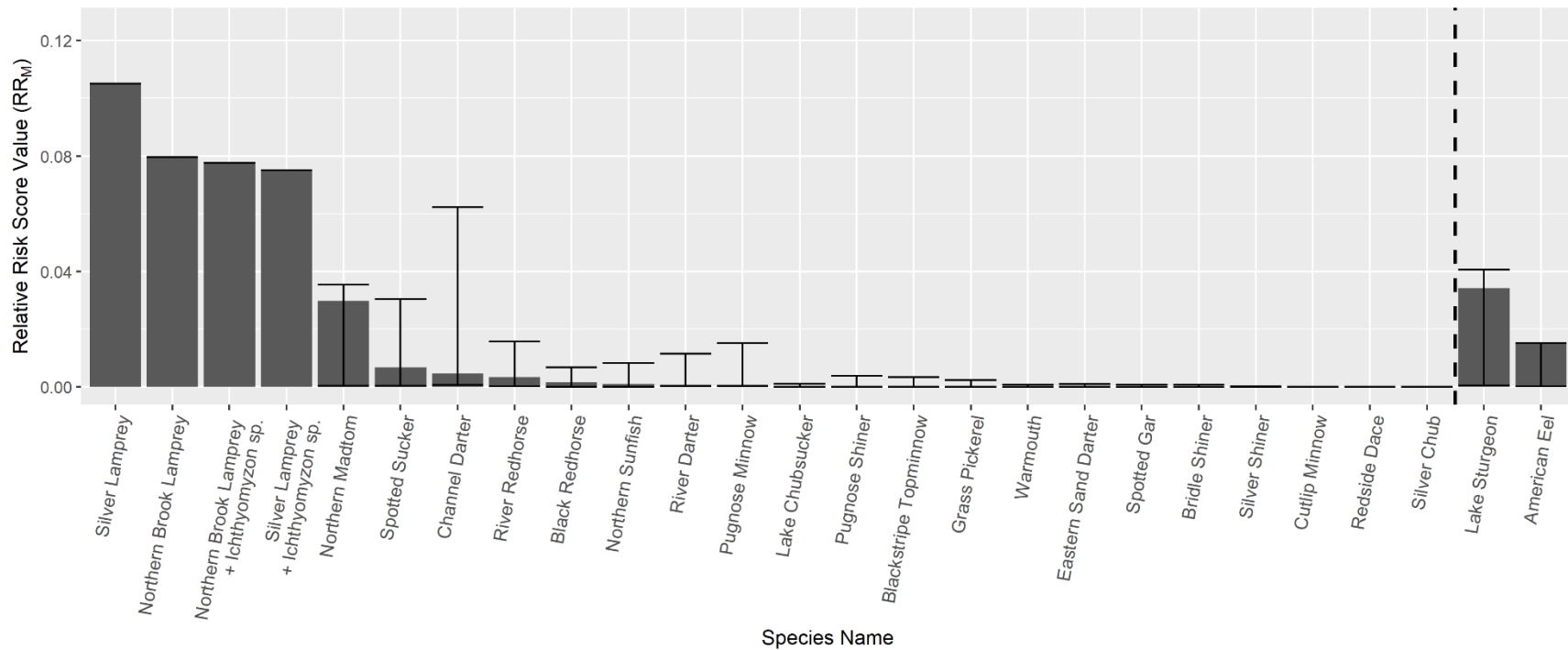


Figure 6. Relative risk, RR<sub>M</sub>, for fish species of conservation concern. Error bars (solid line) on non-lamprey species represent the highest and lowest possible relative risk using toxicity values from the most sensitive and least sensitive non-lamprey fish surrogates, respectively. Lake Sturgeon and American Eel are separated by a dashed line and not presented in rank order as their relative risk is not directly comparable with other fishes due to assessment methods. Because Silver Lamprey and Northern Brook Lamprey cannot be distinguished as larvae, the risk assessment was conducted using only records identified to the species level or with species level plus Ichthyomyzon sp. records included. A value of 1 in the y-axis indicates that the entire species range is susceptible to granular Bayluscide (gB) applications, that applications within the range occur with high intensity, that the species occurs only in Type I or Type II habitat, and that the species would experience complete mortality given the exposure benchmark. A value of 0 represents no range overlap or intensity, species occurrence in Type III habitat, and no mortality expected given the exposure benchmark.

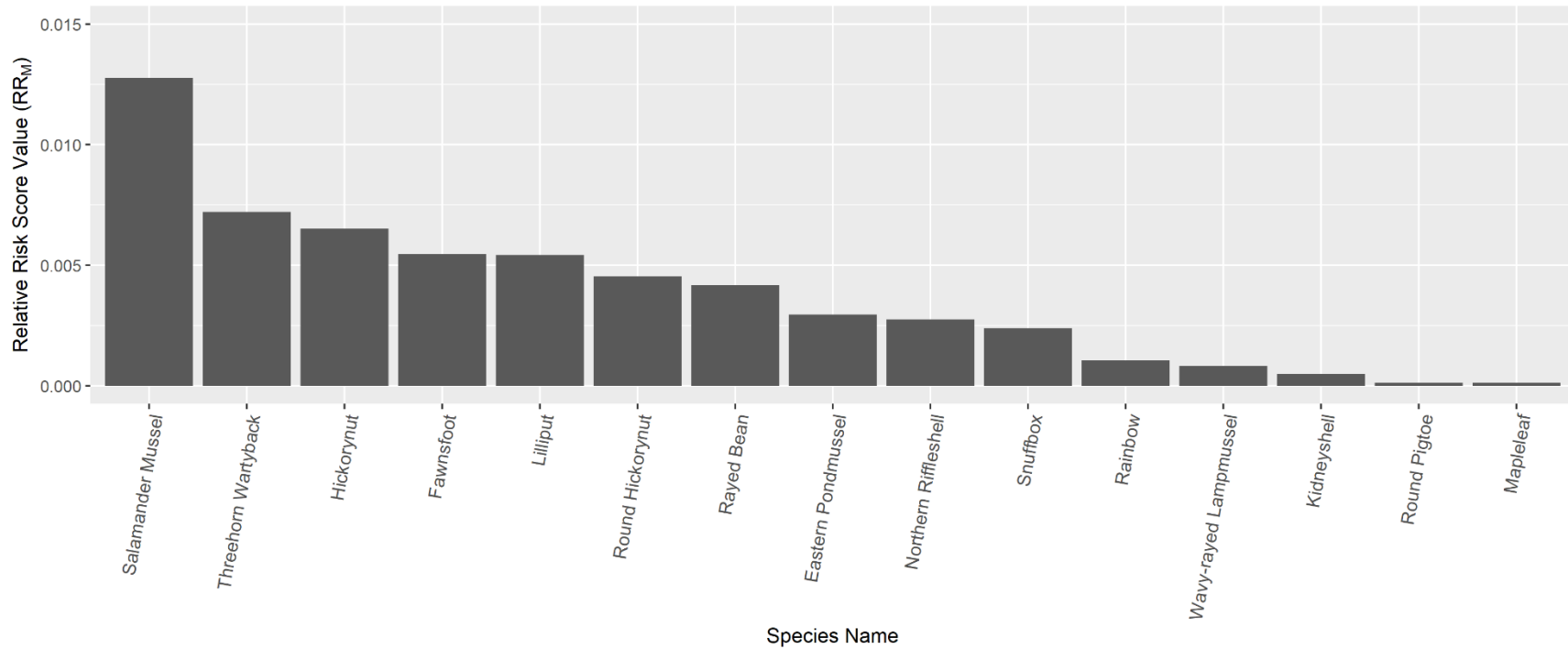


Figure 7. Relative risk, RR<sub>M</sub>, for mussel species of conservation concern. A value of 1 in the y-axis indicates that the entire species range is susceptible to gB applications, that applications within the range occur with high intensity, that the species occurs only in Type 1 or Type 2 habitat, and that the species would experience complete mortality given the exposure benchmark. A value of 0 represents no range overlap or intensity, species occurrence in Type 3 habitat, and no mortality expected given the exposure benchmark.

## Direct Mortality in the Huron-Erie Corridor and Nearby Tributaries

### *Species Exposure to gB in the Huron-Erie Corridor*

The likelihood of occurrence of fishes in Sea Lamprey Type I or Type II habitat classes varied across classes. Species with the greatest likelihood of occurrence in Type I or Type II habitat were Spotted Sucker (*Minytrema melanops*;  $p = 0.402$ ; Type I habitat), Eastern Sand Darter ( $p = 0.308$ ; Type II habitat), and Blackstripe Topminnow (*Fundulus notatus*;  $p = 0.288$ ; Type II habitat). However, almost all fish species of conservation concern had non-zero probabilities of occurring within Type I or Type II habitat with the exception of Black Redhorse and River Redhorse (*Moxostoma carinatum*). Results indicated that the majority of species of conservation concern considered in the analysis may be susceptible to Bayluscide exposure based on habitat factors alone. The estimated density of fishes ranged from a mean value of 0.08 fish/100 m<sup>2</sup> (River Redhorse) to 76.7 fish/100 m<sup>2</sup> (Northern Brook Lamprey and unidentified *Ichthyomyzon*).

The likelihood of occurrence for mussel species was generally lower than for fishes. However, the estimated density of mussel species was substantially greater than for fishes. All mussel species of conservation concern had non-zero values of occurring within Type I or Type II habitat. Likelihood of occurrence was generally higher in Type II than Type I habitat. In some cases, moderately high values were found for certain species such as Kidneyshell ( $p = 0.189$ , Type II habitat), Northern Riffleshell ( $p = 0.165$ , Type II habitat), and Mapleleaf ( $p = 0.141$ , Type II habitat). Density values varied greatly and ranged from 100 mussels/100 m<sup>2</sup> (e.g., Round Hickorynut in the Sydenham River) to 283.6 mussels/100 m<sup>2</sup> (e.g., Mapleleaf in the Thames River). Similar to the results for fishes, there was no discernable pattern between the likelihood of occurrence and density for mussels.

### *Species-Specific gB Toxicity*

Surrogate values of Bayluscide toxicity varied across fishes and mussels. However, toxicity to fish species exhibited greater variability than toxicity to mussel species, likely due to the greater variety of surrogate species used to estimate toxicity. Overall, Bayluscide toxicity was greatest for *Ichthyomyzon* spp., followed by species with Channel Catfish assigned as a surrogate. Across all species, Bayluscide was the least toxic to *Percidae* and *Lepomis* species. Toxicity was greatest for the mussel species Kidneyshell and Wavy-rayed Lampmussel (*Lampsilis fasciola*) and species that used those species as a surrogate for toxicity estimates.

### *Mortality from gB Applications*

Overall, the estimated mortality from Bayluscide applications displayed a strongly right-skewed probability distribution of the number of fishes and mussels killed per application cycle. Zero Bayluscide-induced mortality was the most likely outcome across the range of species for a single application cycle. However, higher mortality, while not the norm, was possible for both fishes and mussels, with severity based on whether the median or 95<sup>th</sup> percentile values of the mortality distribution were of interest. For non-lamprey fishes, most species were predicted to experience no bayluscide-related mortality based on median values (only Blackstripe Topminnow had a median mortality value of one fish or greater). However, a greater number of individuals were predicted to be killed based on the 95<sup>th</sup> percentile values (i.e., mortality that occurs 5% of the time from a single application cycle). Generally, *Ichthyomyzon* spp. yielded the greatest mortality based on the 95<sup>th</sup> percentile, with > 300 individuals killed for some scenarios (i.e., Silver Lamprey in the Sydenham River as well as Silver Lamprey and unidentified *Ichthyomyzon* sp.; Northern Brook Lamprey and unidentified *Ichthyomyzon* sp.; and unidentified *Ichthyomyzon* sp. in the Thames River). Substantial mortality was observed for other fish species based on the 95<sup>th</sup> percentile, including 22 individuals killed (Lake Chubsucker in the St.



Clair River), 19 individuals killed (Blackstripe Topminnow in the Sydenham River), and 3 individuals killed (Eastern Sand Darter in the Detroit, Sydenham, or Thames rivers).

The mortality results for mussel species were similar to fishes in that median results were characterized by a lack of Bayluscide-induced mortality for all study species except Kidneyshell (43 individuals killed; Sydenham River). Some species that may experience high mortality 5% of the time or less, including Rayed Bean (1,442 individuals killed; Sydenham River), Northern Riffleshell (1,304 individuals; Sydenham River), and Kidneyshell (1,131 individuals killed; Sydenham River). In contrast, some species yielded no mortality based on 95<sup>th</sup> percentiles for both the Thames and Sydenham rivers including Threehorn Wartyback, Round Hickorynut, and Salamander Mussel. This contrasts the high estimated risk ratings from the relative risk assessment for two of these species, which can be largely explained by differences in geographic scope between both research documents (Andrews et al. 2021, Smyth and Drake 2021) and because density was not considered in the relative assessment. Similar to the fish results, the mortality results for mussels demonstrated that most Bayluscide application cycles will result in no or relatively low mortality, but substantial mortality (> 1,000 individuals killed) has the potential to occur 5% of the time or less.

#### *Adjustments to gB Application Methods*

Overall, increasing the number or size of application sites for a single Bayluscide application cycle increased the range of mortality for a species. Decreasing the number or size of application sites decreased the range of mortality provided that non-zero mortality occurred under baseline conditions. For most species, increasing the number or size of application sites did not affect median mortality (e.g., Lake Chubsucker and Northern Madtom) but when it did (e.g., Northern Brook Lamprey and Silver Lamprey), the distribution remained heavily right skewed and dominated by low mortality. The most substantial change in median results was observed when increasing the number of application sites for species with high density (e.g., Mapleleaf and Kidneyshell). Although these distributions remained heavily right skewed, the increase in median mortality, specifically when changing the number of application sites, is due to the highly dense and patchy distribution of these species. As the number of application sites increases, the likelihood of encountering a patch of many individuals increases, thus increasing median mortality. Therefore, the greatest effect of adjustments to the gB application cycle (in terms of number or size of sites) will be a reduction in extreme mortality outcomes, rather than changes to the average condition.

#### *Population-level Effects of gB*

Population-level effects of gB were evaluated for a subset of fishes. The population-level effect of gB was strongly affected by application frequency as well as the amount of habitat occupied by the species. The benefit of decreasing application frequency from once every year to once every 10 years was dependent on population size where smaller populations (0.01% of bounded range) benefited more than larger populations (100% of bounded range). The relationship between the percentage of initial population abundance remaining after 100 years and application frequency was non-linear (Figure 8).

Both Northern Brook Lamprey and Silver Lamprey, when evaluated with unidentified *Ichthyomyzon* sp. records, experienced severe population decline (> 90%) after 100 years if the minimum amount of habitat is occupied, regardless of the frequency of gB application cycles. Eastern Sand Darter also experienced significant population declines after 100 years, but decline was more heavily dependent on gB application frequency and habitat occupancy. In a scenario where the minimum amount of habitat was occupied and an annual gB application cycle was assumed, a ~ 90% population decline for Eastern Sand Darter occurred after 100 years (Smyth and Drake 2021). Similarly, Northern Madtom populations experienced near

collapse after 100 years when the minimum amount of habitat was occupied and the gB application cycle was five years or less (Figure 8). For Channel Darter, > 20% population decline occurred after 100 years when the minimum amount of habitat was occupied and the gB application cycle was two years or less. In all cases, the estimate of population-level effects was based on a series of worst-case assumptions regarding the ability of focal species to rebound following mortality events, indicating that the population declines presented here are worst-case scenarios. For specific details on the population-level effects of gB on these fish species, see Smyth and Drake (2021).

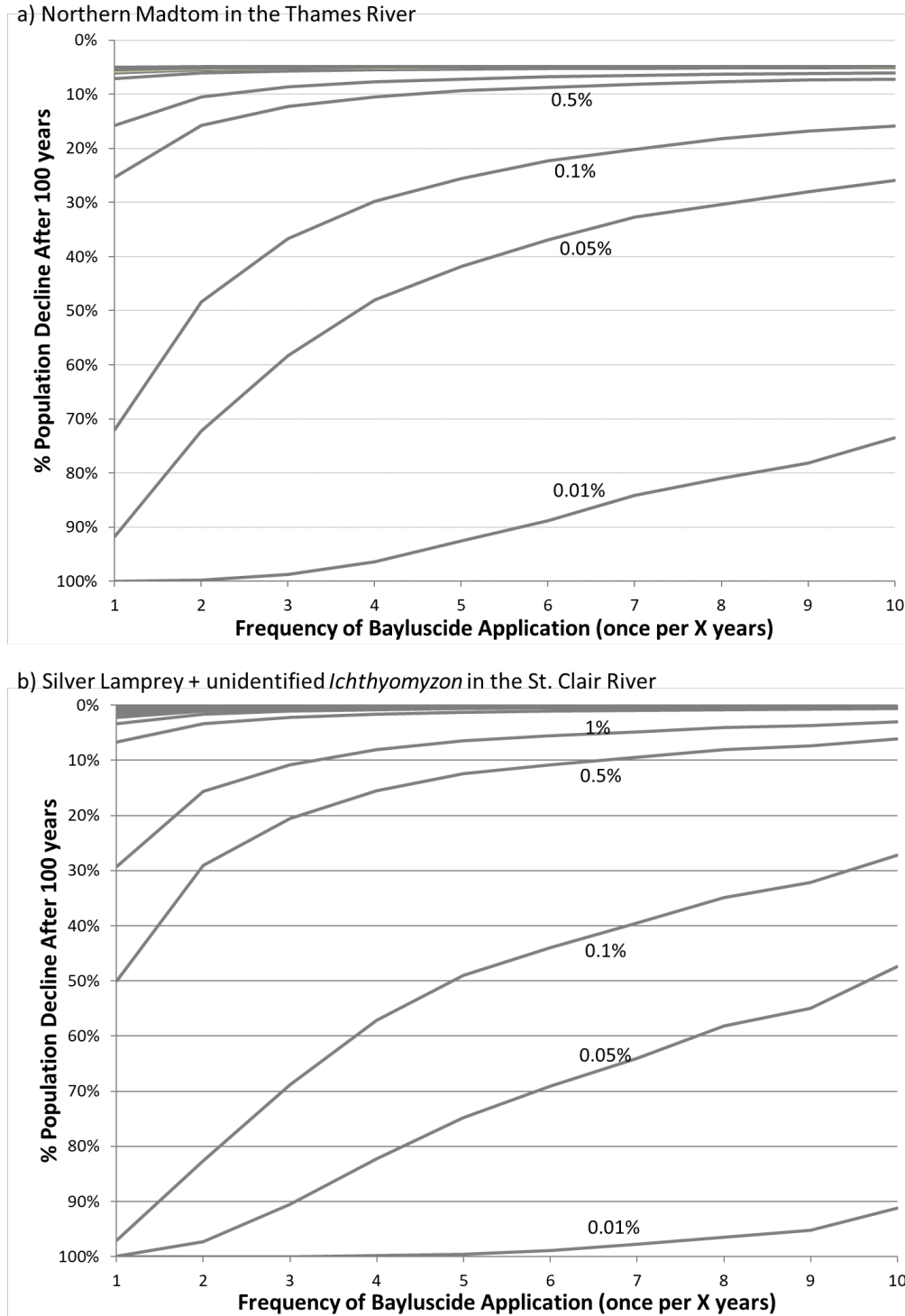


Figure 8. Percentage decline in population abundance of a) Northern Madtom in the Thames River and b) Silver Lamprey + unidentified *Ichthyomyzon* sp. in the St. Clair River following simulated granular Bayluscide application cycles across different frequencies (one to ten years) for 100 years. The percentage values beside each grey line represent the percentage of critical habitat area or bounded range occupied by the species.

*Comparing Results from the Relative Risk Assessment with Mortality Estimates from the Huron-Erie Corridor*

In most cases, the results from Smyth and Drake (2021) align with results from Andrews et al. (2021). For example, the relative risk assessment demonstrated that native lampreys (*Ichthyomyzon* spp.) exhibited the greatest relative risk among fishes. This finding was consistent with Smyth and Drake (2021), who indicated that native lampreys can experience very high mortality events (among the highest of all species considered) as a result of gB applications under certain conditions. Excluding native lampreys, relative risk for SARA-listed fishes was highest for Northern Madtom. This was also consistent with findings from Smyth and Drake (2021) where analyses demonstrated that gB applications can significantly affect Northern Madtom populations depending on the application frequency and the number of fish occupying critical habitat.

Similarly for freshwater mussel species, consistent results between both research documents (Andrews et al. 2021, Smyth and Drake 2021) were found in terms of the overall risk rankings for Fawnsfoot, Mapleleaf and Rainbow, and the estimated absolute mortalities for these species in the Thames and Sydenham rivers. Low absolute mortality estimates for Rainbow and Mapleleaf in the Sydenham River found in Smyth and Drake 2021 were consistent with their lower risk rankings compared to other mussel species in Andrews et al. 2021. Likewise, absolute mortality (95<sup>th</sup> percentile) for Fawnsfoot in the Thames River ranked third highest among mussels (Smyth and Drake 2021), which again was consistent with Fawnsfoot ranking fourth highest among mussels in Andrews et al. 2021.

Some inconsistencies occurred between relative risk rankings from Andrews et al. (2021) and mortality estimates from Smyth and Drake (2021), the majority of which can be explained by geographic scope, species abundances and distribution, and habitat methods. In Smyth and Drake (2021), mortality was estimated in four focal rivers with the likelihood of occurrence based on the probability that a species would occur in Type I or Type II habitat. Whereas, habitat use in the risk assessment (Andrews et al. 2021) was identified as the percentage of occurrences in Types I and II habitats across the Great Lakes region. Furthermore, absolute mortality estimates considered species densities, whereas abundance was not included in the relative risk calculation. Inconsistent results include Lake Sturgeon's high relative risk (Andrews et al. 2021), yet zero mortality was estimated in models from Smyth and Drake (2021) under all scenarios for the species in the Detroit and St. Clair rivers. Multiple factors likely played a role in this discrepancy, including its low population density and removal of offshore records from the risk assessment. Inconsistencies also occurred for mussel results. For example, Smyth and Drake (2021) estimated zero mortality (95<sup>th</sup> percentile) for Salamander Mussel and Threehorn Wartyback, yet these two species ranked highest in the relative risk assessment. The discrepancy can similarly be explained by the way in which risk was evaluated in both documents. Species density was only incorporated within Smyth and Drake (2021) and the zero mortality estimated in that document was driven by extremely low species densities (i.e., very few individuals found during surveys where density could be calculated). Results suggest that encountering Salamander Mussel and Threehorn Wartyback during gB application is very unlikely, but mortality may be substantial if it an occupied habitat patch is encountered.

**Sources of Uncertainty**

- The concentration of gB in the aquatic environment is poorly known beyond what can be inferred from a handful of laboratory studies. It is uncertain whether or for how long gB remains within (or extends beyond) the application site, including the consequence of species exposure outside of these areas.

- The toxicity of gB, including the concentration and duration of exposure leading to lethal or sub-lethal responses, is unknown for most fishes and mussels of conservation concern. The suitability of surrogate species is also poorly understood.
- Estimates of relative risk and direct mortality are contingent on patterns of past gB applications, which may not reflect future application patterns. Future shifts in application location and intensity will change overall risk for a particular species, but the direction of this change is unknown.
- In addition to factors outlined above, the population consequences of gB are contingent on assumed population sizes, current population trajectories, and the ability of populations to recover following a mortality event. These factors are poorly known for most fishes and mussels of conservation concern.
- An understanding of how the control of Sea Lamprey has improved the persistence of species susceptible to Sea Lamprey predation (e.g., Lake Sturgeon), relative to the scope for gB-induced mortality, remains poorly understood.
- Avoidance responses of fishes and mussels to gB exposure, including the ecological consequences of avoidance, have only been investigated in a few laboratory studies.
- Changes to growth, reproduction, or movement of species of conservation concern and related components of the ecosystem (prey, competitors, predators) have not been evaluated and remain poorly understood.

Despite increased knowledge about the potential effects of gB, numerous uncertainties remain including underlying uncertainty about the ecology and population abundance of fishes and mussels of conservation concern, the concentration of gB in the aquatic environment, the response and avoidance of fishes and mussels to gB, and the importance of indirect pathways (e.g., food-web effects).

Sampling to understand the distribution and abundance of species at risk has been limited, with sampling programs often employing different assessment methods or sampling intervals. This is particularly relevant for mussels in the St. Clair and Detroit rivers where lack of sampling in recent decades prevents knowledge of the current status of several species. Field sampling in 2019 revealed that mussel species at risk inhabit the Detroit River (Allred et al. 2020) but the related occurrence data were not available for the analysis (Andrews et al. 2021). Gaps in distribution records for Lake Sturgeon in areas where they are known to occur resulted in different risk assessment methods for that species.

Imperfect detection and sparse sampling also led to uncertainty about range boundaries while species densities were often based on a low number of field samples. Models from Smyth and Drake (2021) indicated that the long-term population consequences of gB-induced mortality depend heavily on population sizes of fishes and mussels. However, population abundances for most species of conservation concern were unknown. To illustrate a range of population sizes and the corresponding consequences of gB-induced mortality across 50 or 100 years, Smyth and Drake (2021) extrapolated patch-specific species densities and made assumptions about the proportion of the bounded range with non-zero density. Gaining knowledge of true population abundance would significantly improve the ability to estimate long-term population consequences.

In addition to detection and density estimates, habitat attributes were measured differently across field programs. Inconsistencies between the identification of substrate, or whether Types I and II habitat is homogenous throughout a gB application site, would influence the results presented here. Collectively, these factors highlight uncertainty pertaining to the range,

intensity, and habitat variables of the relative risk assessment (Andrews et al. 2021) and the likelihood of occurrence estimates (Smyth and Drake 2021). Factors related to species detection, density, and habitat classification have ultimately led to imperfect knowledge of gB exposure in both research documents.

Estimating the potential for gB-induced mortality requires that the concentration of the compound can be estimated in the aquatic environment. Although gB is applied at a constant rate to achieve a peak concentration of 11 mg/L (9.3 mg/L active ingredient niclosamide) in the bottom 5 cm of the water column (Adair and Sullivan 2004), it is likely that variability in environmental conditions (river discharge, habitat complexity) can lead to variability in environmental concentrations of the compound. Refined in-water estimates of gB concentrations including the effect of flow, depth, distance from application site, and other water quality variables (e.g., temperature, pH, conductivity) are needed to refine the likelihood of mortality. Both documents incorporated the simplifying assumption that gB applications occur only within Types I and II habitat. The extent and severity of gB diffusion to Type III habitat is largely unknown and will influence species exposure including risk and mortality estimates.

In general, the effects of non-physiological mechanisms in response to gB exposure (e.g., avoidance and its consequences) for fishes and mussels are poorly understood. Evidence exists from laboratory trials that some surrogate species may detect and avoid gB by elevating their position in the water column (Boogaard et al. 2016), but it is unclear whether at-risk species display similar responses. It is also unclear how these responses would differ (if at all) in a field setting or the consequences of elevation across the duration of the application cycle. Although avoidance may reduce the direct mortality pathway, displacement of species to suboptimal habitat may impair growth or survival through other mechanisms. In the case of small bodied fishes, the ability to avoid a large application area may not be feasible due to poor swimming ability. Mussels do not have the ability to rapidly avoid gB through movement, but may use valve-closure as an avoidance mechanism, thereby reducing filtering and resulting processes (e.g., feeding, excretion, reproduction). The consequences of avoidance behaviours (e.g., swimming, valve-closure) are poorly understood as they relate to potential effects on growth and survival for most fishes and mussels.

Both research documents focused on a single pathway of effect (direct mortality) to assess the effect of gB on fishes and mussels. However, relatively little is known about the indirect pathways that may impact fish and mussel species (Figure 2). For example, incomplete knowledge of food-web connections exist for most species of conservation concern making it extremely difficult to gauge the importance of indirect pathways relative to direct mortality. For some species, indirect pathways may elicit population responses such as for species with obligate species dependencies (e.g., Lake Sturgeon is a host fish for Hickorynut and both species have high risk to Bayluscide applications). Importantly, food-web effects could lead to beneficial outcomes for species at risk such as relaxing predation pressure on small fishes by reducing the abundance of non-lamprey predators or via the direct rescue from lamprey-induced predation and wounding on large-bodied species (e.g., Lake Sturgeon).

Almost all toxicity information in this science advisory process was based on surrogate values through taxonomic matching. For many species, the closest surrogate did not belong to the same genus and even species-specific differences in tolerance are likely to occur. This issue becomes of even greater importance for species such as Lake Sturgeon and Spotted Gar (*Lepisosteus oculatus*) where toxicity information was based on surrogates from different subclasses and infraclasses, respectively. As tolerance to gB is expected to vary across taxonomic groups based on physiological and behavioural responses to exposure, surrogate assignment has an important influence on estimates of relative risk and mortality for most species (Figure 6).

Although the risk of gB was evaluated based on patterns of past gB applications, the future spatial and temporal distribution of gB application is largely unknown due to uncertainty around Sea Lamprey population dynamics and the resulting need to assess and/or control the species in new localities. A flowchart has been provided to allow the results from Andrews et al. (2021) to be used in three circumstances. If similar application patterns are expected in the future, the baseline results from Andrews et al. (2021) will hold in the future (Figure 9, option 1) to describe the relative risk of mortality across fishes and mussels of conservation concern. However, if gB application is being considered for new ecosystems beyond those assessed in the study period (2011–2017), or effects on single ecosystems are of interest, maps from Andrews et al. (2021) can be used to determine the presence of a species in a given watercourse and relative risk can be estimated by multiplying the habitat (*H*) and toxicity (*T*) values of the species in question (Figure 9; option 2). The habitat and toxicity values are unaffected by spatial and temporal patterns of gB applications and therefore represent the stand-alone vulnerability of each species if applications occur within a watercourse containing the species. Alternatively, if the full suite of gB-related effects are of concern (e.g., indirect and direct pathways), habitat and toxicity values can be removed or down-weighted to rank species primarily on the basis of gB exposure (Figure 9; option 3).

All factors identified above (increased knowledge of population abundance, environmental concentrations of gB, species-specific toxicity, likelihood for and consequences of avoidance across sessile and non-sessile organisms, indirect physiological and non-physiological food-web effects, and effectiveness of potential mitigation measures) are important avenues for future research.

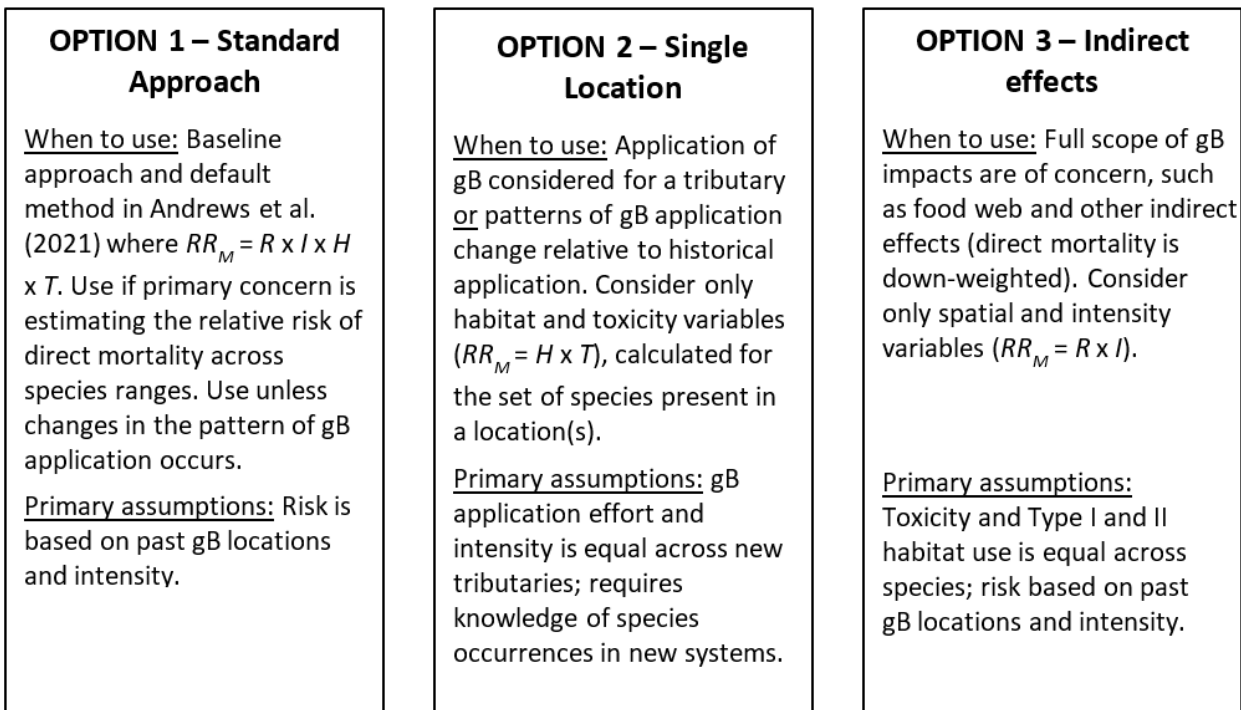


Figure 9: Framework for using risk assessment variables from Andrews et al. (2021) under different scenarios and assumptions.



## OTHER CONSIDERATIONS

### Mitigations and Alternatives

Models from Smyth and Drake (2021) indicated that gB-induced mortality has the potential to occur for certain fishes and mussels of conservation concern for a subset of Great Lakes tributaries (Detroit, St. Clair, Thames, and Sydenham rivers). However, the results of Andrews et al. (2021) indicate that the conditions for exposure and mortality exist throughout the Great Lakes basin and are not limited to the four focal rivers. Therefore, mitigation measures and alternatives may be warranted to reduce the likelihood of direct, gB-induced mortality on species of conservation concern. Moreover, because very little information is available about other pathways of effects (indirect pathways involving food-web components; non-physiological mechanisms such as avoidance), mitigation of those pathways may also be warranted in the absence of knowledge of their severity. A review of potential mitigation and alternative measures was conducted (Table 1), focusing on the potential benefits to species of conservation concern and key uncertainties. In some cases, mitigation measures have been quantitatively evaluated in Smyth and Drake (2021); whereas, in other cases, Table 1 presents measures that may reduce mortality but have not been quantitatively analyzed.

Should mitigation measures be pursued, it is recommended that they be accompanied by rigorous field testing to ensure intended benefits are realized and unintended outcomes minimized. More broadly, additional analytical research may be needed to understand how proposed mitigation measures may reduce the efficiency of Sea Lamprey assessment including how to maximize benefits to species of conservation concern while minimizing unintended consequences for Sea Lamprey assessment and control.

## CONCLUSIONS

High mortality events are possible under certain conditions, especially for native lampreys and SARA-listed freshwater mussels, and to a lesser extent for SARA-listed fishes. However, in most cases, gB applications are not expected to pose direct mortality. The risk assessment revealed a gradient of relative risk across species of conservation concern based on gB application locations, habitat features, and toxicity. Although gB applications occurred within the range of 36 fish and mussel species of conservation concern (including within areas currently or previously identified as critical habitat at the time of application for 6 fishes and 10 mussels) in the Great Lakes basin from 2011 to 2017, gB exposure is relevant for up to 27% of a species' distribution range and for less than 30% of gB application sites. Modeling indicates that high population reductions are possible for certain species under worst-case scenarios and therefore may warrant mitigation measures to avoid jeopardizing the survival or recovery of species. Mitigation measures such as changes to application site size and application frequency have model-based support to reduce the likelihood of extreme mortality events.

Table 1. Mitigations and alternatives to granular Bayluscide (gB) application in the Great Lakes basin, focusing on benefits and considerations for species of conservation concern.

Action	Benefit to Species of Conservation Concern	Considerations
Avoidance of watersheds containing species of conservation concern	Avoidance removes all negative direct and indirect, physiological and non-physiological mechanisms that lead to reduced viability of fishes and mussels of conservation concern.	<ul style="list-style-type: none"> <li>• Avoidance may lead to reduced effectiveness of Sea Lamprey assessment and control, which may result in negative effects for species that experience predation or wounding from Sea Lamprey (e.g., Lake Sturgeon).</li> <li>• The trade-off between avoidance and positive and negative effects of gB application among fishes and mussels of conservation concern is poorly understood (e.g., avoidance may benefit most species of conservation concern while negatively affecting those vulnerable to Sea Lamprey predation like Lake Sturgeon). These issues may be of lesser importance in watersheds that lack species susceptible to Sea Lamprey predation.</li> </ul>
Reducing realized concentrations of gB in the aquatic environment	Potential to reduce mortality and other direct and indirect pathways (including sub-lethal effects) to fishes and mussels of conservation concern.	<ul style="list-style-type: none"> <li>• The maximum concentration at which toxicity is negligible for non-target species is unknown given multiple plausible direct and indirect pathways.</li> <li>• Requires large investment and research effort to evaluate effectiveness on non-target species.</li> <li>• Uncertainty exists about the fate of gB in the aquatic environment at current application rates, which would need to be resolved to demonstrate a meaningful reduction in realized concentration.</li> <li>• The trade-off between reducing realized concentrations and positive and negative effects among fishes and mussels is poorly understood, particularly for species experiencing Sea Lamprey predation like Lake Sturgeon.</li> </ul>
Reducing the frequency with which gB is applied in a particular area	Potential to reduce mortality and other direct and indirect pathways (including sub-lethal effects) to fishes and mussels of conservation concern.	<ul style="list-style-type: none"> <li>• Relationship between application frequency and population effects is non-linear and highly dependent on assumed population abundances (see Smyth and Drake [2021]), which are poorly understood for most species of conservation concern.</li> <li>• The trade-off between reducing the frequency of application and positive and negative effects among fishes and mussels is poorly understood, particularly for species experiencing Sea Lamprey predation like Lake Sturgeon.</li> </ul>
Reducing size or number of gB application sites	Potential to reduce mortality, especially rare, high-abundance mortality events (Smyth and Drake 2021). Potential to reduce other direct and indirect pathways (including sub-lethal effects) to fishes and mussels of conservation concern.	<ul style="list-style-type: none"> <li>• Does not eliminate the risk of mortality to fishes and mussels of conservation concern.</li> <li>• Relationship between treatment plot size/number and mortality is non-linear (Smyth and Drake 2021).</li> <li>• The trade-off between reducing size/number of gB application sites and positive and negative effects among fishes and mussels is poorly understood, particularly for species experiencing Sea Lamprey predation like Lake Sturgeon.</li> </ul>

Action	Benefit to Species of Conservation Concern	Considerations
Move location of application sites to areas outside or downstream of critical habitat	Potential to decrease direct and indirect pathways to fishes and mussels of conservation concern, particularly when applications are located downstream of occupied habitat.	<ul style="list-style-type: none"> <li>• The distribution of fishes and mussels of conservation concern is poorly known; assumes range boundaries known with precision.</li> <li>• May not reduce indirect effects (requires better understanding of food web linkages).</li> </ul>
Salvage/exclusion of mussels or fishes of conservation concern prior to gB application.	Decreases the number of fishes and mussels of conservation concern within application area. Potential to reduce direct and indirect pathways.	<ul style="list-style-type: none"> <li>• Removal sampling and salvage is often incomplete due to gear selectivity; fishes and mussels of conservation concern are likely to remain in application site and experience gB exposure.</li> <li>• Deepwater mussels are extremely challenging to sample and relocate</li> <li>• Potential mortality or harm to fishes and mussels can occur during capture and relocation (e.g., consequences for growth or survival).</li> <li>• Mobile species can return to application area prior to gB treatment.</li> </ul>
Offset impacts to non-target species through habitat restoration or other feasible means	An offset such as habitat restoration may increase the availability or quality of habitat, thereby increasing the viability of non-target fishes or mussels	<ul style="list-style-type: none"> <li>• Effectiveness of offsetting for fishes and mussels of conservation concern is highly uncertain. Certainty can be increased by implementing offset in advance and validating effectiveness</li> <li>• Species in question may not be habitat limited, so habitat-related offsets may not provide benefit to species. Feasible offsets may not exist for the species in question.</li> <li>• Physical habitat manipulations may be insufficient to produce net benefit for species if the application of gB has the potential to extirpate fishes or mussels from the system.</li> </ul>
Application of gB after August 1 <sup>st</sup> or seasonally outside of reproductive periods for a given species	Avoids harm to sensitive life stages (e.g., spawning, young of year) for many fish and mussel species	<ul style="list-style-type: none"> <li>• Does not eliminate the risk of mortality to fishes and mussels of conservation concern.</li> <li>• Currently a lack of knowledge about how the timing of application leads to mortality or other effects on fishes and mussels.</li> <li>• Unknown if seasonal adjustment of application imposes other trade-offs or unexpected consequences.</li> </ul>

## LIST OF MEETING PARTICIPANTS

Name	Organization/Affiliation
Dave Andrews	DFO – Science
Andrew Drake	DFO – Science
Eric Smyth	DFO – Science
Jason Barnucz	DFO – Science
Kelly McNichols-O'Rourke	DFO – Science
Lynn Bouvier (Chair)	DFO – Science
Olivia Sroka (Rapporteur)	DFO – Science
Todd Morris	DFO – Science
Tom Pratt	DFO – Science
Lisa Wren	DFO – Fish and Fish Habitat Protection Program
Alan Rowlinson	DFO – Sea Lamprey Control Centre
Bruce Morrison	DFO – Sea Lamprey Control Centre
Fraser Neave	DFO – Sea Lamprey Control Centre
Mike Steeves	DFO – Sea Lamprey Control Centre
Shawn Robertson	DFO – Sea Lamprey Control Centre
Tonia Van Kempen	DFO – Sea Lamprey Control Centre
Amy Boyko	DFO – Species at Risk Program
Becky Cudmore	DFO – Species at Risk Program
Shelly Dunn	DFO – Species at Risk Program
Michael Siefkes	Great Lakes Fishery Commission
Kim Fredericks	United States Geological Survey
Michael Boogaard	United States Geological Survey
Theresa Newton	United States Geological Survey
Ryan Prosser	University of Guelph
Margaret Docker	University of Manitoba
Nick Mandrak	University of Toronto

## SOURCES OF INFORMATION

This Science Advisory Report is from the February 28–March 1, 2019 regional peer review meeting on Information on the Potential Harm to Fish and Mussel Species at Risk (SAR) from Bayluscide Applications. Additional publications from this meeting will be posted on the [Fisheries and Oceans Canada \(DFO\) Science Advisory Schedule](#) as they become available.

Adair, R.A., and Sullivan, P. 2004. Standard Operating Procedures for Application of Lampricides in the Great Lakes Fishery Commission Integrated Management of Sea Lamprey (*Petromyzon marinus*) Control Program. U.S. Fish and Wildlife Service and Fisheries and Oceans Canada Report SLC 04-001.10: x +49 p.

- Allred, S.S., Woolnough, D.A., Morris, T.J., and Zanatta, D.T. 2020. Status update for native mussels in the Detroit River. *In* Proceedings of the 2019 Canadian Freshwater Mollusc Research Meeting: December 3-4, 2019, Burlington, Ontario. *Edited by* T.J. Morris, K.A. McNichols-O'Rourke, and S.M. Reid. Can. Tech. Rep. Fish. Aquat. Sci. 3352: viii + 34 p.
- Neave, F.B., Bravener, G.A., and Mandrak, N.E. 2007. [Conservation status report for Silver Lamprey \(\*Ichthyomyzon unicuspis\*\)](#). Can. Sci. Advis. Sec. Res. Doc. 2007/043. vi + 52 p.
- Andrews, D.W., Smyth, E.R.B., Lebrun, D.E., Morris, T.J., McNichols-O'Rourke, K.A. and Drake, D.A.R. 2021. [Relative Risk of Granular Bayluscide Applications for Fishes and Mussels of Conservation Concern in the Great Lakes Basin](#). DFO Can. Sci. Advis. Sec. Res. Doc. 2021/034. viii + 174 p.
- Bills, T.D., and Marking, L.L. 1976. Toxicity of 3-trifluoromethyl-4-nitrophenol (TFM), 2',5-dichloro-4'-nitrosalicylanilide (Bayer 73), and a 98:2 mixture to fingerlings of seven fish species and to eggs and fry of Coho Salmon. U.S. Fish and Wildlife Service, Investigations in Fish Control 69: 24 p.
- Boogaard, M.A., Erickson, R.A., and Hubert, T.D. 2016. Evaluation of avoidance behavior of Tadpole Madtoms (*Noturus gyrinus*) as a surrogate for the endangered Northern Madtom (*Noturus stigmosus*) in response to granular Bayluscide®. U.S. Geological Survey Open-File Report 2016-1130: 11 p.
- Dawson, V.K. 2003. Environmental fate and effects of the lampricide Bayluscide: a review. J. Great Lakes Res. 29(Suppl.1): 475–492.
- DFO. 2014. [Guidance on Assessing Threats, Ecological Risk and Ecological Impacts for Species at Risk](#). DFO Can. Sci. Advis. Sec. Sci. Advis. Rep. 2014/013. (*Erratum*: June 2016)
- Eshenroder, R. L. 2014. The role of the Champlain Canal and Erie Canal as putative corridors for colonization of Lake Champlain and Lake Ontario by sea lampreys. Trans. Am. Fish. Soc. 143(3): 634–649.
- Gilderhus, P.A. 1979. Effects of granular 2',5-dichloro-4'-nitrosalicylanilide (Bayer 73) on benthic macroinvertebrates in a lake environment. Great Lakes Fishery Commission Technical Report 34: 5 p.
- Great Lakes Fishery Commission. 1956. Annual Report of the Great Lakes Fishery Commission for 1956. Great Lakes Fishery Commission, Ann Arbor, MI. 36 p.
- Hubert, T.D. 2003. Environmental fate and effects of the lampricide TFM: a review. J. Great Lakes Res. 29 (Suppl. 1): 456–474.
- Larval Assessment Task Force. 2012. Larval assessment sampling protocol for non-wadable waters of the Great Lakes and its tributaries. Great Lakes Fishery Commission Internal Report, Ann Arbor, MI. 22 p.
- Marking, L.L., and Hogan, J.W. 1967. Toxicity of Bayer 73 to fish. Investigations in Fish Control 19, U.S. Fish and Wildlife Service, Washington, D.C. 13 p.
- Newton, T.J., Boogaard, M.A., Gray, B.R., Hubert, T.D., and Schloesser, N.A., 2017. Lethal and sub-lethal responses of native freshwater mussels exposed to granular Bayluscide (R), a sea lamprey larvicide. J. Great Lakes Res. 43(2): 370–378.
- Scholefield, R.J., and Seelye, J.G. 1992. Toxicity of 2',5-dichloro-4'-nitrosalicylanilide (Bayer 73) to three genera of larval lampreys. Great Lakes Fishery Commission Technical report 57: 6 p.

- Slade J.W., Adams, J.V., Cuddy, D.W., Neave, F.B., Sullivan, W.P., Young, R.J., Fodale, M.F., and Jones, M.L. 2003. Techniques and methods for estimating abundance of larval and metamorphosed sea lampreys in Great Lakes tributaries, 1995-2001. *J. Great Lakes Res.* 29(Suppl.1): 130–136.
- Smith, B.R., and Tibbles, J.J. 1980. Sea Lamprey (*Petromyzon marinus*) in Lakes Huron, Michigan, and Superior: history of invasion and control, 1936-1978. *Can. J. Fish. Aquat. Sci.* 37(11): 1780–1801.
- Smyth, E.R.B., and Drake, D.A.R. 2021. [Estimating the Mortality of Fishes and Mussels of Conservation Concern Resulting from Bayluscide® Applications within four rivers of the Huron-Erie Corridor](#). DFO Can. Sci. Advis. Sec. Res. Doc. 2021/035. xi + 198 p.
- USFWS (United States Fish and Wildlife Service) and DFO (Fisheries and Oceans Canada). 2016. Procedure for application of Bayluscide 3.2% granular Sea Lamprey larvicide for assessment or control applications. Technical Operating Procedures TOP017.11: 7 p.

### THIS REPORT IS AVAILABLE FROM THE:

Center for Science Advice (CSA)  
Ontario and Prairie Region  
Fisheries and Oceans Canada  
501 University Crescent, Winnipeg, Manitoba, R3T 2N6

Telephone: (204) 983-5232  
E-Mail: [csas-sccs@dfo-mpo.gc.ca](mailto:csas-sccs@dfo-mpo.gc.ca)  
Internet address: [www.dfo-mpo.gc.ca/csas-sccs/](http://www.dfo-mpo.gc.ca/csas-sccs/)

ISSN 1919-5087  
ISBN 978-0-660-38488-7 Cat. No. Fs70-6/2021-016E-PDF

© Her Majesty the Queen in Right of Canada, 2021



Correct Citation for this Publication:

DFO. 2021. Science Advice on the Potential Harm of granular Bayluscide Applications to Fish and Mussel Species at Risk. DFO Can. Sci. Advis. Sec. Sci. Advis. Rep. 2021/016.

*Aussi disponible en français :*

*MPO. 2021. Avis scientifique sur les dommages potentiels des applications de Bayluscide granulaire pour les espèces de poissons et de moules en péril. Secr. can. de consult. sci. du MPO. Avis sci. 2021/016.*