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Maritimes Region **Science Response 2024/031**

LAKE UTOPIA RAINBOW SMELT, LARGE-BODIED POPULATION (LURS-LBP): UPDATED ABUNDANCE AND GENETIC ANALYSES

Context

Lake Utopia is part of the Magaguadavic River watershed in southwestern New Brunswick. The Rainbow Smelt (*Osmerus mordax*) living in Lake Utopia (LURS) represent one of the only three confirmed occurrences in Canada of co-existing genetically divergent smelt populations. Two populations of smelt co-exist in the lake: the small-bodied population (SbP) and the large-bodied population (LbP). The SbP has been protected under the *Species at Risk Act* (SARA) since 2003. The LbP was designated as Threatened by the Committee on the Status of Endangered Wildlife in Canada (COSEWIC 2008) because it is one of a sympatric species pair, it is endemic to a single lake with a small index of area of occupancy (6 km^2), it spawns in only three small streams, and it could quickly become extinct through degradation of spawning streams from increasing development around the lake shore and impacts of the dip-net fishery. Potential threats identified by COSEWIC (2008) were the introduction of exotic species and increasing eutrophication. Fisheries and Oceans Canada (DFO) undertook a Recovery Potential Assessment (RPA) for both LbP and SbP populations in 2010 (DFO 2011). Both populations were re-assessed as endangered by COSEWIC in 2018. The LbP was listed as endangered under SARA in 2019.

Sampling of the 2014 and 2017 LbP spawning runs were used to estimate population abundance and provide advice on an appropriate population size objective, as well as the likelihood that the population objective could be achieved under current levels of mortality (DFO 2018). However, the abundance estimates for the 2014 and 2017 spawning runs were uncertain as genetic analyses of the 2014 spawning run indicated a mismatch between expected genotype and the minimum fork length used to differentiate LbP from SbP spawners. No genetic analyses were available for the 2017 spawning run.

Recent genetic analyses of the 2017 LbP spawning run, as well as tissue sampling of SbP spawners in 2017 and 2018, provide an opportunity to review the minimum fork length that can be used to differentiate phenotypically between the LbP and SbP populations.

The objectives of this Science Response Process were to:

- Provide updated abundance estimates for LURS-LbP in 2014 and 2017 based on genetic analysis of these samples undertaken in 2019.
- Review the recommended minimum fork length criterion for distinguishing LURS-LbP from LURS-SbP.
- Discuss how the genetic analyses and updated abundance estimates affect the likelihood that the updated abundance objective for LURS-LbP can be achieved under current levels of mortality (actual mortality levels are not known).

This Science Response Report results from the Regional Science Response Process of November 19, 2019, and January 20, 2020, on Lake Utopia Rainbow Smelt, Large-bodied Population (LURS-LbP) Population Abundance and Allowable Harm Estimate.

Background

Although the two sympatric smelt populations in Lake Utopia are reproductively isolated and genetically distinct, the occurrence of hybrids indicates that gene flow occurs between the two populations (Bradbury et al. 2011). They are also distinguishable phenotypically by their choice of spawning stream, the timing of their spawning run, and morphological differences.

Historical data suggest that the LbP use the two largest streams, Mill Lake Stream and Trout Lake Stream-Spear Brook, for spawning, whereas Smelt Brook, Unnamed Brook, and Second (Scout) Brook are used by the SbP (COSEWIC 2008). The LbP spawns between late-March and mid-April, whereas the SbP spawns from mid-April until mid-late May.

The body forms of the two populations are distinguishable by their relative eye and jaw to body size ratios, number of gill rakers, and body size at maturity (Bradbury et al. 2011). Although COSEWIC (2008) characterized the LbP as spawners measuring 136–227 mm fork length (FL) and SbP as spawners measuring 73–136 mm FL, DFO adopted a minimum FL of 170 mm for the LbP, based on a larger sample size and evaluation of phenotypic and genotypic diversity in the two populations (Bradbury et al. 2011, Bradford et al. 2012).

Using genetic analyses of tissue samples, Lake Utopia smelt can be assigned to the LbP or SbP genotypes based on the proportion (q-value) of their genome that is characteristic of that population. Individuals whose genetic background is primarily derived from a single population have a q-value approaching one for that population, while an individual with mixed ancestry will have an intermediate value. Historical genetic analyses indicate that presumed SbP individuals had q-values greater than 0.9 SbP genotype (Bradbury et al. 2011). Presumed LbP individuals showed more hybridization, requiring a q-value equal to or greater than 0.7 LbP genotype to be classified as LbP population (Bradbury et al. 2011).

The genetic analyses of smelt collected during the 2014 and 2015 spawning runs in Lake Utopia streams confirmed the presence of both the SbP and the LbP populations (DFO 2018). Both populations displayed increased hybridization compared to smelt sampled from 1990–2010 (DFO 2018), particularly the LbP. Most smelt spawning in Smelt and Second brooks could be characterized as SbP because all the fish sampled had q (SbP) values greater than 0.8. Hybridization was more pronounced in smelt collected from Mill Lake Stream, where individuals could only be characterized as the LbP by accepting a minimum q-value of 0.63 (LbP) genome.

More than 90% of 378 spawning smelt sampled during the early 2014 Mill Lake Stream spawning run were smaller than 170 mm FL, indicating that this criterion was ineffective for characterizing LbP spawners. DFO (2018) proposed 143 mm FL, the minimum length of spawners exhibiting greater than 63% LbP genome. A second spawning run sampled in Mill Lake Stream in late April 2014 was a mix of smelt with a characteristic SbP genotype and smelt with an intermediate or hybridized LbP genotype (DFO 2018).

Mark-recapture events were conducted over two nights during the 2017 Mill Lake Stream spawning run (DFO 2018), but no genetic analysis was available at that time to categorize the proportion of the run as LbP, SbP, or hybridized. DFO (2018) used the proposed minimum 143 mm FL, estimating consecutive nightly spawner abundance of 5,654 (April 13) and 10,917 (April 14).

Analysis and Response

Tissue Sampling During 2017 and 2018 Spawning Runs

In 2017, eight fin clips were collected during two mark-recapture events on the Mill Lake Stream spawning run (Table 1). Eight fin clips were also collected during the SbP spawning run in Second Brook. Four length-stratified sampling events took place in April and May 2018: 95 fin clips on April 15 and 9 fin clips on April 23 in Mill Lake Stream; four fin clips on May 3 during the spawning run in Smelt Brook; and 90 fin clips during the spawning run in Second Brook (May 3-7). The samples taken from Second Brook in 2018 coincided with an attempted mark-recapture study, only one night of which was successful.

Table 1. Lake Utopia Rainbow Smelt samples collected in 2017 and 2018 from large-bodied population (Mill Lake Stream) and small-bodied population (Second and Smelt brooks) spawning streams.

Spawning Stream	Date	Number Sampled	Fork Length Range (mm)
Mill Lake Stream	April 14, 2017	8	103–157
Second Brook	May 5, 2017	8	101–131
Mill Lake Stream	April 15, 2018	95	124–262
Mill Lake Stream	April 23, 2018	9	122–144
Smelt Brook	May 3, 2018	4	113–120
Second Brook	May 3-7, 2018	90	93–150

Fin clips from all sampling events were preserved separately in ethanol and labelled to correspond to information regarding the fish's sex and length. Tissue analyses and interpretation of the results were conducted by Dr. Paul Bentzen and Dr. Tony Einfeldt of the Marine Gene Probe Laboratory (Dalhousie University, Halifax, NS).

Observations During 2018 Spawning Run in Mill Lake and Trout Lake Streams

Visual observations were conducted for Mill Lake Stream and Trout Lake Stream according to the methodology described in MacDonald and Burbidge (2017). Each stream was divided into zones and smelt were counted from the shore. Sampling of Trout Lake Stream was enhanced by installing egg mat collectors near the culvert that were checked twice per week. A gill net with one inch stretched mesh was deployed for 30 minutes at midnight on April 12 and April 15 at the culvert. No eggs were observed on the collectors, and no smelt or other species were caught in the gill net.

Genetic Analyses of the 2017 and 2018 Spawning Runs

Figure 1 shows the smelt sampled in 2017 and 2018 ordered along the horizontal axis first by their spawning location and timing and then by increasing body length, while the vertical axis indicates increasing q-value (LbP) genotype. All smelt collected from Second and Smelt brooks consistently exhibit less than 0.3 (LbP) genotype, that is, q-values of greater than 0.7 (SbP) genotype. Genotypes of smelt from the early Mill Lake Stream run were more variable (q-values of 0.3 to 0.8 (LbP) genotype), as were smelt from the later Mill Lake Stream run (q-values of 0.3 to 0.6 (LbP) genotype). There is a tendency for the proportion of LbP genotype to increase with body size. Greater variability in the genome of smelt from Mill Lake Stream compared to smelt

from Second and Smelt brooks indicates that gene flow is occurring between the two populations, mainly from the SbP to LbP population, and to a greater degree in Mill Lake Stream than Second and Smelt brooks.

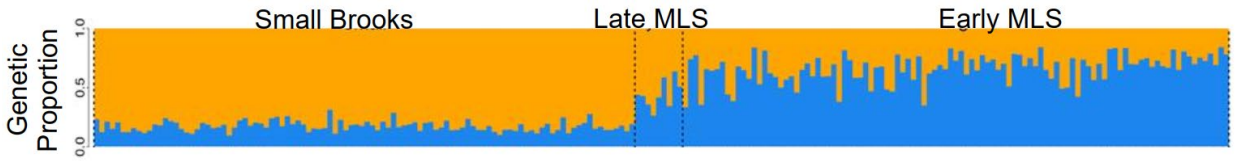

Figure 1. Proportion of genotype characteristic of large-bodied population (LbP; blue) in Lake Utopia Rainbow Smelt collected from Lake Utopia streams in 2017 and 2018. Each bar represents a sample arranged from left to right on the x-axis from earliest to latest sampling events and by increasing body length. Small brooks: fish collected from Smelt and Second brooks; Late MLS: fish collected during the second observed run in Mill Lake Stream on April 23, 2018; Early MLS: smelt collected during early run in Mill Lake Stream in 2017 and 2018.

Figure 2 shows the relationship between spawner body length and the proportion of LbP genome. A gap is evident in the distribution of samples in Figure 2 at the 130 mm FL separating samples collected in the early Mill Lake Stream sampling and samples collected in Second and Smelt brooks. Using the 130 mm FL to attribute the samples to the LbP or SbP, identifying smelt with FL 130 mm or less in length as SbP and larger fish as LbP, results in a miscall rate of 4-10% (Figure 2).

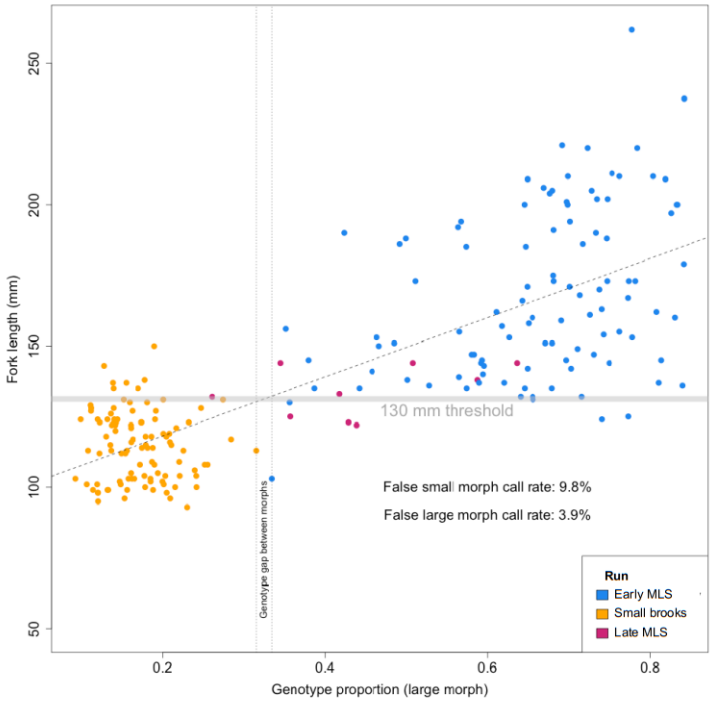

Figure 2. Relationship between proportion of large-bodied smelt genome and fork length (mm) of smelt collected from Lake Utopia streams in 2017 and 2018 (slope indicated by dashed line). The grey horizontal line is proposed as the threshold for classifying smelt as the Large Bodied Population. Small brooks: fish collected from Smelt and Second brooks; Late MLS: fish collected during the second observed run in Mill Lake Stream on April 23, 2018; Early MLS: smelt collected during early run in Mill Lake Stream in 2017 and 2018.

Updated 2017 Nightly Spawner Abundance Estimates

Abundance estimates of smelt in the 2017 Mill Lake Stream spawning run were adjusted by a ratio of 0.83 in DFO (2018), because it was assumed that fish less than 143 mm FL were SbP spawners. While few of the 2017 spawners were genotyped, the genetic analyses described here indicate that all of the 2017 and 2018 spawners larger than 130 mm FL could be classified as LbP, with a miscall rate of 4%. Using a minimum 130 mm FL as an indicator of population identity, 95% of the individuals marked-recaptured during the 2017 Mill Lake Stream spawner abundance study (DFO 2018) were LbP (Figure 3). Therefore, the nightly LbP spawner estimates for the two nights sampled in 2017 are 6,319 (April 13) and 12,201 (April 14) (Table 2).

Using a minimum 130 mm FL to differentiate LbP, 99% of the early April 2014 spawning run sampled in Mill Lake Stream were LbP (Figure 3). Nightly LbP spawner estimates over the five nights with mark-recapture events ranged from 1,707 to 23,421 (Table 2).

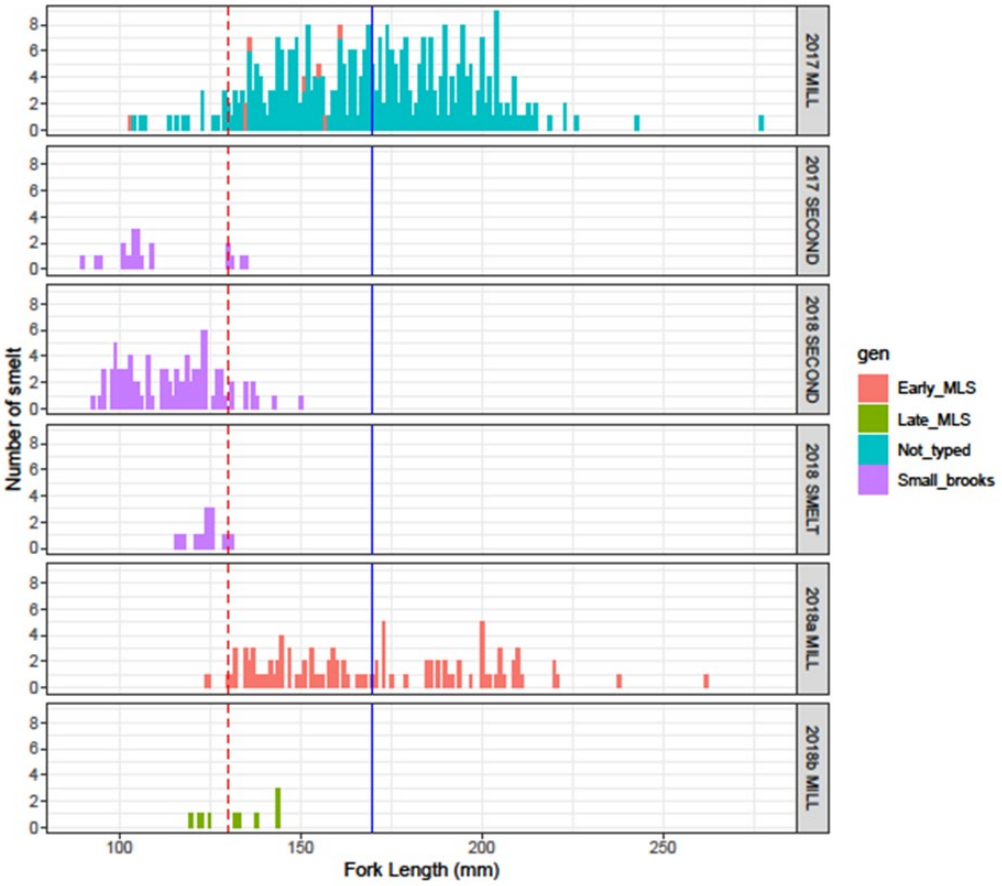

Figure 3. Length frequency distributions of smelt spawning runs sampled in 2017 and 2018. First panel: Mill Lake Stream (MLS), April 2017; second panel: Second Brook, 2017; third panel: Second Brook, 2018; fourth panel: Smelt Brook, 2018; fifth panel: Mill Lake Stream, early April 2018; sixth panel: Mill Lake Stream, late April 2018. Colours (gen) indicate locations where genotyped smelt were sampled: purple – Second and Smelt brooks; pink – Mill Lake Stream run (early April 2018); green – Mill Lake Stream run (late April 2018); blue – Mill Lake Stream run not genotyped (early April 2017). Vertical lines indicate minimum fork length used to differentiate large-bodied smelt population: Solid blue line: 170 mm length currently used; dashed red line: 130 mm length based on recent genetic analyses.

Table 2. Abundance estimates of spawning Lake Utopia Rainbow Smelt in Mill Lake Stream in 2014 and 2017 (DFO 2017, 2018). Number of large-bodied population spawners (LbP) estimated by applying a minimum fork length (FL) of 130 mm.

Date	Estimated Number Spawners	95% Confidence Intervals	Proportion >130 mm FL	Number of LbP Spawners
April 3, 2014	1,724	812-3,641	0.99	1,707
April 4, 2014	3,082	1,453-6,512	0.99	3,051
April 5, 2014	14,542	12,051-18,514	0.99	14,397
April 6, 2014	12,058	9,561-16,099	0.99	11,937
April 10, 2014	23,658	13,965-41,156	0.99	23,421
April 13, 2017	6,652	5,200-9,200	0.95	6,319
April 14, 2017	12,843	9,600-19,800	0.95	12,201

Visibility Ratio Based on the 2017 Mill Lake Stream Spawning Run

The number of fish visually observed on April 13, 2017, in Mill Lake Stream using the method outlined in MacDonald and Burbidge (2017) was 3,500 compared to a mark-recapture estimate for the same night of 6,652 spawners (DFO 2018). Using the ratio of these two numbers as an index of visibility indicates that 53% of the smelt in the stream are visible to someone observing the run from the bank compared to the numbers that would be estimated using a mark-recapture event. This ratio is recommended for estimation of spawning run size in years without mark-recapture events.

Figure 4. Location of zones monitored in Mill Lake Stream (from MacDonald and Burbidge (2017).

Observations During the 2018 Spawning Run

Smelt were first observed in Mill Lake Stream on April 9 (Table 3), when they were seen in all three marked zones. Zone A is from the shoreline to the downstream end of the culvert; Zone B is between the upstream ends of the culverts and the riffle area below the falls; and, Zone C is from the riffle area to the outflow from Mill Lake (Figure 4; MacDonald and Burbidge 2017). The size of the run increased to greater than 3,500 fish six days after smelt were first observed, with the highest numbers occurring between 2400 hr and 0200 hr. Smelt passed through the main culvert and congregated in Zones B and C. The water level was high enough for Zone C to be accessible.

Using the visibility ratio to indicate the numbers of smelt that would have been estimated by mark-recapture events, the number of smelt ranged from 511 to 6,736 over the six days of observation (Table 3).

Table 3. Visual estimates of Lake Utopia Rainbow Smelt in Mill Lake Stream in April 2018; zone with highest numbers of smelt, nightly total maximum numbers of fish, and time of the peak numbers.

Date	Time (hr)	Zone of Maximum Abundance	Maximum Numbers Observed	Numbers Estimated by Visibility Ratio
April 9	0130	A: Riffles to Mill Lake outflow	271	511
April 11	0130	A: Riffles to Mill Lake outflow	1,250	2,358
April 12	2400	B: Above culverts to riffles	370	698
April 15	0130	C: Riffles to Mill Lake outflow	3,570	6,736
April 19	2400	C: Riffles to Mill Lake outflow	420	793
April 23	0200	C: Riffles to Mill Lake outflow	819	1,545

Sources of Uncertainty

Few genetic samples were available for the 2017 spawning run in Mill Lake Stream. The run is presumed to have been LbP spawners because the length frequency of the spawners and the timing of the run were similar to the 2014 and 2018 spawning runs. The overall population abundance of the LbP in any year cannot be determined because sampling does not take place on every night of the spawning run and the proportion of the whole population spawning on any given night is unknown. Additionally, the contribution by Trout Lake Stream or Spear Brook is not known. Spear Brook is difficult and, at times, dangerous to access, so observations were not attempted in 2017 or 2018. Trout Lake Stream was monitored on the same schedule as Mill Lake Stream, but no visual observations of smelt were made in either year. Therefore, uncertainty remains about whether smelt use locations other than Mill Lake Stream to spawn and the genetic composition of spawning runs in these locations.

Adopting a minimum 130 mm FL to distinguish the LbP from the SbP is a 24% decrease from the 170 mm FL established by DFO (2011), but consistent with observations of the Mill Lake Stream spawning run. COSEWIC (2008) adopted a minimum 136 mm FL, based on Curry et al. (2004) observation of 120–260 mm smelt during the 1999 and 2000 runs, as well as genetic analyses by Taylor and Bentzen (1993). Given the current overlap in length frequencies between the SbP and LbP populations, minimum FL is not useful for indicating to which

population an individual belongs. The genetic analyses reported in DFO (2018), and this report indicate that timing and location of the spawning run should be used as the first indicators of population identity, followed by body length, and corroborated by genetic data when available.

As more life-history information is collected through the annual sampling of smelt runs in Lake Utopia spawning streams, the earliest spawning run appears to occur in Mill Lake Stream. These smelt are a broad range of sizes and ages, including some very large fish, with variable genotypes, but more characteristic of the LbP than the seasonally later runs that occur in Second, Smelt, and Unnamed brooks. In years when sampling of Mill Lake Stream was extended into late April and May, a second run of smelt with a smaller body size and a genome more characteristic of SbP (the 2014 spawning run), or hybrids (the 2018 spawning run), has been observed. Hybridization between the two populations is occurring and is impacting the LbP. This is evidenced by the declining characteristic minimum FL observed in this study and previous work (Bradbury et al. 2011, DFO 2018), the difficulty in assigning a characteristic genotypic proportion to the LbP, and the occurrence of hybridized smelt in the early Mill Lake Stream spawning run.

Recommendations

It is recommended that timing and location of the spawning run should be used first as indicators of population identity, followed by FL, and corroborated by genetic data when available.

A decrease in the minimum FL used to categorize LbP from 170 mm to 130 mm is recommended, based on genetic analyses of the 2017 and 2018 spawning runs in Mill Lake Stream, Second, and Smelt brooks.

Further biological sampling (e.g., non-lethal fin clips) is required to assess the following: the degree of hybridization between the two smelt populations; the use of Mill Lake Stream by SbP; and to identify the mechanisms promoting hybridization. Sampling should be length-stratified and temporally-stratified to ensure that changes in the character of the spawning run can be detected.

Conclusions

Nightly abundance estimates for the 2014 LbP spawning run in Mill Lake Stream are 1,707–23,421, based on five mark-recapture events. The nightly spawner estimates for the two nights sampled by mark-recapture in 2017 are 6,319 on April 13 and 12,201 on April 14.

The updated recovery abundance objective for the LbP of 5,000 individuals (DFO 2018) occupying Mill Lake Stream on at least one night was met in 2014 and 2017, based on mark-recapture estimates. Although the maximum number of spawners visually observed in 2018 was 3,570, applying a visibility ratio to account for the smelt present, but not detectable to an observer, increases the nightly count to greater than 6,000 smelt. The maximum nightly spawner abundance estimates have decreased over the last four years from 23,421 in 2014, to 12,201 in 2017, and to 6,736 in 2018.

The maximum level of allowable harm that the LbP can sustain without jeopardizing survival or recovery cannot be quantified given the unpredictability of the annual spawning run size occupying Mill Lake Stream. Spawning runs exceeding the recovery abundance target in 2014, 2017, 2018, and the presence of multiple year classes in the 2017 spawning run, are encouraging signs that the population is at, or above, the abundance objective threshold.

Recent monitoring efforts indicate that Mill Lake Stream is the only stream known to be used by the LbP. Smelt have not been observed in Trout Lake Stream since 2012, despite increased sampling effort in 2018. The declining minimum FL required to distinguish the LbP smelt, the weakening genetic differentiation between the two populations, and the increase in the proportion of hybrids suggest that the rate of hybridization between the two populations is increasing and/or the selective pressures removing hybrids (Bradbury et al. 2011) are decreasing.

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