

# **PFAS in New Hampshire Loon Eggs**



Photo credit: Brian Reilly

**Loon Preservation Committee  
Tiffany Grade, Squam Lakes Biologist  
Harry Vogel, Senior Biologist/Executive Director**

**December 2023**







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### Executive Summary

New Hampshire Department of Environmental Services and Loon Preservation Committee collaborated to evaluate the concentrations of PFAS in 144 inviable loon eggs from across the state to understand where significant bioaccumulation of PFAS is occurring in loons as well as potentially in fish and other organisms.

Key findings include:

- The geometric mean of total PFAS in eggs was  $306.3 \pm 28.2$  ng/g wet weight (ww), range: 85.6-1938.0 ng/g ww.
  - 26% of eggs exceeded the lowest observed effects level (LOEL) for PFOS in other bird species.
- Elevated levels of PFAS were found on Lake Winnepesaukee (mean:  $1033.4 \pm 108.0$  ng/g ww [range: 325.2-1938.0 ng/g ww]).
  - Of the 16 eggs tested, only one egg was below the PFOS LOEL in other bird species.
  - For each of the 9 loon territories tested, the average of eggs tested exceeded the LOEL.
- Elevated levels of PFAS were geographically concentrated in the southern (and especially southeastern) part of the state, including Winnepesaukee.
- 7:3 FTCA had a mean level of 32.6 ng/g ww, with a maximum of 174.0 ng/g ww. The egg with the maximum level was from Winnepesaukee.
  - The mean level of 7:3 FTCA in loon eggs was 12 times the highest level previously reported in bird eggs (2.7 ng/g ww) and the maximum loon egg was 64 times this level.
  - Toxicity of 7:3 FTCA in bird eggs is unknown, but there are indications in other biota that it may be more toxic than PFCAs.

Recommendations include:

- Fish sampling on Lake Winnepesaukee and other lakes with elevated levels of PFAS in loon eggs to identify possible risks to human health from consumption of fish from these lakes.
- Investigation of potential source(s) of PFAS in Lake Winnepesaukee.
- Further testing and monitoring of PFAS in loon eggs or other high-trophic level species to identify other lakes of concern and establish temporal trends.
- Research into the movement of PFAS through aquatic food webs.

## **Background**

Per- and polyfluoroalkyl substances (PFAS) have been used since the 1950s in stain repellants, surfactants, and fire-fighting foam, and have been found to persist in the environment and bioaccumulate in wildlife (Newsted et al. 2005, Verreault et al. 2005, T. Custer et al. 2010). Two major sub-groups of PFAS are the perfluoroalkyl carboxylates (PFCAs) and perfluoroalkyl sulfonates (PFASAs; Table 1). The latter category includes perfluorooctane sulfonic acid (PFOS), which has historically been considered the dominant type of PFAS found in wildlife tissues (Newsted et al. 2005). Data on the effects of PFOS on wildlife are contradictory (Wu et al. 2020, Custer et al. 2021), while knowledge of the effects of PFCAs and other types of PFAS analytes on wildlife is limited or lacking (Tartu et al. 2014, Eriksson et al. 2016, Custer 2021, Jouanneau et al. 2022). Growing awareness of the toxicity and persistence of these chemicals and their widespread distribution in the environment has led to increased concern about their effects on human and wildlife health.

As a long-lived species at the top of aquatic food webs, Common Loons (*Gavia immer*) are sensitive indicators of contaminants in aquatic ecosystems (Strong 1990, Evers 2006). Nutrients and contaminants deposited in eggs are primarily from dietary uptake in the weeks preceding egg laying (C. Custer et al. 2010; C. Custer, pers. com.; R. Letcher, pers. com.; LPC, unpubl. data) when loons are feeding in their territories (Gingras and Paszkowski 1999; LPC, unpubl. data). Therefore, contaminant levels in loon eggs are indicative of contaminants and potential risks to wildlife and human health in lakes or loon territories.

Following the Loon Preservation Committee's (LPC) discovery of elevated levels of PFAS in some loon eggs in New Hampshire (Grade and Vogel 2021), New Hampshire Department of Environmental Services (NHDES) conducted fish testing and issued PFAS fish consumption advisories on several lakes. NHDES and LPC collaborated to evaluate the concentrations of PFAS in 144 inviable loon eggs to understand where significant bioaccumulation of PFAS is occurring in fish and other organisms. Per the contract between NHDES and LPC, approved by Governor and Council December 21, 2022, this report details the PFAS concentrations of each egg for each congener, summarizes the range of PFAS concentrations found in the samples, and identifies locations at which PFAS concentrations exceed those known to affect the health, breeding success, or survival of other bird species. We also report on contaminant profiles, as different congener patterns could reflect different potential sources of contaminants (Chen et al. 2010, Su et al. 2017), different bioaccumulation or biotransformation patterns (Chen et al. 2010, Su et al. 2017), and different toxicities (Chen et al. 2010, Tartu et al. 2014).

## **Methods**

One hundred forty-four inviable loon eggs collected from failed nests between 2017-2022 were tested for PFAS to investigate recent PFAS levels in eggs. LPC collects inviable loon eggs under permits from United States Fish and Wildlife Service and New Hampshire Department of Fish and Game. Collection procedures were detailed in Grade and Vogel (2021); but, in brief, collected eggs were placed in sealed plastic bags and stored at -18°C.

Of the 144 eggs tested, 100 eggs were from 50 territories to allow for a paired sample of two eggs from separate years or clutches from a single loon territory within the study period. Fourteen eggs were tested to pair with eggs from a given territory previously tested by LPC within the study period. When eggs from the same territory were not available, we tested six eggs representing two separate loon territories on each of three lakes to allow for a paired sample from a given lake. We also tested 24 single eggs from a lake if a second egg was not available within the study time frame. This sample represents 77 lakes and 94 loon territories throughout New Hampshire.

We packed intact eggs in dry ice and shipped them to SGS AXYS Analytical Services Ltd. (Sidney, British Columbia, Canada) for homogenization and PFAS testing. A summary of analytical procedures at SGS AXYS are included in Appendix A. Eggs were tested for 40 PFAS congeners (Table 1). Lake or territory total PFAS and total PFCA values are averaged for all paired samples that have two eggs tested for all 40 PFAS congeners. Six eggs, previously analyzed by LPC, were tested when SGS AXYS was analyzing only 13 PFAS congeners. For these eggs, total PFAS and total PFCA levels are presented for only the egg tested for the 40 congeners. PFOS values are averaged for all paired samples, as PFOS was included among the analytes in LPC's earlier testing. An additional six eggs, previously tested by LPC in 2019 and 2020, were analyzed by SGS AXYS for 33 congeners. Total PFAS for these eggs was included in the averages for paired samples, despite being underestimated compared with eggs tested for 40 congeners. The seven congeners not included in testing of these eggs were 3:3 FTCA, 5:3 FTCA, 7:3 FTCA, PFEESA, PFMPA, PFMBA, and NFDHA. All eggs were adjusted for moisture loss, according to procedures in Grade and Vogel (2021). Results were corrected for blanks when blank values exceeded 5% of the lab result (see SGS AXYS 2023, cf. Hill et al. 2022).

## **Results and Discussion**

### *PFAS levels and profiles*

For the 144 eggs tested, the geometric mean of total PFAS was  $306.3 \pm 28.2$  ng/g ww (range: 85.6-1938.0 ng/g ww). The geometric mean of PFCA congeners was  $148.1 \pm 12.9$  ng/g ww (range: 28.3-839.7 ng/g ww; Table 1). Congeners detected in 100% of egg samples included PFNA, PFDA, PFUnA, PFDoA, PFTrDA, PFTeDA, PFOS, and PFDS. Additionally, PFOA, PFHxS, PFHpS, PFOSA, and 7:3 FTCA were detected in >90% of the samples (Table 1).

On average, PFOS was the dominant congener in the total PFAS profile ( $33.8 \pm 0.9\%$  [range: 8.4-64.9%]), followed by PFTrDA ( $14.6 \pm 0.4\%$  [range: 4.6-26.7%]), PFUnA ( $13.9 \pm 0.3\%$  [range: 3.2-23.1%]), and 7:3 FTCA ( $12.9 \pm 0.6\%$  [range: 0.0-36.7%]; Table 1). Overall, PFCAs accounted for  $49.4 \pm 0.8\%$  (range: 22.1-72.6%) of the total PFAS profile, and PFSAs accounted for  $35.2 \pm 0.9\%$  (range: 9.1-65.8%).

PFCAs accounted for >40% of the overall PFAS profile in 84.0% of the eggs tested (121 of 144 eggs) and for >50% of the overall profile in 43.1% of eggs (62 of 144 eggs; Fig. 1). PFSAs accounted for >40% of the overall profile in 34.7% of eggs (50 of 144 eggs) and for >50% of the overall profile in 9.0% of eggs (13 of 144 eggs; Fig. 2). This preponderance of PFCAs is

consistent with a trend towards increasing PFCAs and declining PFOS in bird eggs generally following the phase-out of PFOS (Custer et al. 2013, Eriksson et al. 2016).

This trend towards increasing PFCAs and declining PFOS was evident in LPC's previous testing in a dataset spanning 27 years on Squam Lake (1993-2019; Grade and Vogel 2021). The four loon eggs tested from Squam Lake in the present dataset fit this trend, with profiles that showed a higher proportion of PFCAs in the contaminant profile (60.0-67.7%) compared with PFASs (23.5-26.9%). Similarly, Little Squam Lake had the highest proportion of PFCAs in this dataset, with the 2022 egg containing 72.6% PFCAs and 9.1% PFASs.

Conversely, the egg from Arlington Mill Reservoir (Salem, NH; 2021) had the highest proportion of PFOS in the dataset (64.9%). This is expected, given the results of the Arlington Mill egg tested in 2019 by Loon Preservation Committee (Grade and Vogel 2021), which showed a notably high proportion of PFOS (75.9% when including the expanded suite of PFAS congeners vs. the more limited suite included in the analysis in Grade and Vogel 2021). Despite the continuing high proportion of PFOS in the 2021 egg, the decline from the egg tested in 2019 is notable. The distinct profiles of eggs from Arlington Mill may assist in identifying a source or sources of PFAS contaminants in these eggs (Grade and Vogel 2021).

The highest levels of PFCAs were distributed across the southern part of the state, particularly in the southeastern corner (Fig. 3), as well as in Lake Winnepesaukee (Fig. 4), which had the highest levels of total PFCAs in the state (geometric mean:  $474.6 \pm 48.1$  ng/g ww; individual egg: 839.7 ng/g ww). The preponderance of PFCAs in the southern half of the state is likely due to the higher density of human population and industry. Within total PFCAs, PFTrDA and PFUnA dominated the profile (29.2% and 28.1% respectively, Table 1). PFTrDA and PFUnA have similarly been found to be the dominant PFCA in freshwater piscivorous birds such as Herring Gulls, Caspian Terns, and Osprey, as well as in Northern Fulmars (Braune et al. 2013, Letcher et al. 2015, Eriksson et al. 2016, Su et al. 2017). In the present dataset, PFTrDA was the dominant PFAS congener in six of the loon eggs and one egg had PFUnA as the dominant PFAS congener. PFTrDA in plasma has been correlated with decreased corticosterones in Black-legged Kittiwakes (Tartu et al. 2014) and endocrine disruption in fish (Jo et al., 2014). Knowledge of the effects of PFCAs on wildlife is limited and merits further investigation (Tartu et al. 2014, Eriksson et al. 2016, Custer 2021).

7:3 FTCA, a breakdown product of fluorotelomer alcohols (Butt et al. 2014, Spaan et al. 2020), was detected in all eggs except for the two from Little Sunapee Lake (New London; Fig. 5) and accounted for  $12.9 \pm 0.6\%$  (range: 0.0-36.7%) of the PFAS profile. Mean levels of 7:3 FTCA were  $32.6 \pm 2.3$  ng/g ww, with levels exceeding 100 ng/g ww on Ayers Pond (Barrington), Jenness Pond (Northwood), and Kezar Lake (Sutton) and a maximum level of 174.0 ng/g ww on Lake Winnepesaukee (Fig. 6). To our knowledge, the highest level previously reported in bird eggs was 2.7 ng/g ww (Eriksson et al. 2016, Jouanneau et al. 2022). Although knowledge of effects of FTCA in birds are lacking, there are some indications that fluorotelomer acids may be more toxic than PFCAs (Phillips et al. 2007, Jouanneau et al. 2022).

7:3 FTCA was the dominant congener in 11 of the 18 eggs in which PFOS was not the dominant congener. These included eggs from Conway Lake (Conway), Dan Hole Pond (Tuftonboro), Big Diamond Pond (Stewartstown), Duncan Pond (Ossipee), Gunnison Lake (Goshen), Grafton Pond (Grafton), Great East Lake (Wakefield; both eggs tested), Hermit Lake (Sanbornton), Silver Lake (Harrisville), and Little Squam Lake (Ashland). To our knowledge, these are the first avian samples in which 7:3 FTCA was documented to be the dominant PFAS congener.

PFAS levels were generally consistent in the two eggs tested from a given lake or loon territory, as would be expected given that loons are feeding in their territories for 4-6 weeks preceding egg laying and nutrients and contaminants deposited in eggs are primarily from dietary uptake in the weeks preceding egg laying (C. Custer et al. 2010; C. Custer, pers. com.; R. Letcher, pers. com.). LPC's stable isotope analysis confirmed that the majority of nutrients in loon eggs come from freshwater sources (LPC 2013). Excluding four outliers discussed below, the geometric mean of differences between two eggs tested from a given territory for total PFAS was  $69.7 \pm 18.1$  ng/g ww (range: 4.2-664.5 ng/g ww). This is a  $19.2 \pm 2.7\%$  difference (range: 1.4%-83.8%), suggesting that loon eggs are good indicators of levels of PFAS on a lake/territory.

Notable exceptions to this low variability between eggs include Pleasant Lake (New London; +469.3% between 2020 and 2021), Christine Lake (Stark; +250.7% between 2020 and 2022), and Lake Winnepesaukee—Braun Bay (+109.5% between 2019 and 2021) and Varney Island (+106.9% between 2018 and 2019). LPC bands loons to identify individual birds; and, on Pleasant Lake, the long-term female, who was on territory in 2020, was evicted by a new female in 2021. Winnepesaukee (Varney Island) is also a case of a different female laying the two eggs. In the remaining two cases, the females in the pairs were unbanded or not positively identified. Overall, for the nine lakes/territories that are known to have had different females in the two sample years, the geometric mean of differences between two eggs tested from a given territory for total PFAS was  $168.0 \pm 78.9$  ng/g ww (range: 11.7-699.9 ng/g ww), a  $43.7 \pm 47.4\%$  difference (range: 4.7-461.9%). In contrast, among the eggs of the 12 lakes with the same female, the difference for total PFAS was  $48.9 \pm 35.8$  ng/g ww (range: 4.2-380.0 ng/g ww), a  $12.5 \pm 7.1\%$  difference (range: 1.4-83.8%).

The differences in PFAS levels between eggs when different females were in the territory and, in some cases, differences between years for the same females, suggest the importance of an increased understanding of the movement of PFAS in aquatic food webs, the possible impact of changes in fish populations and food availability on contaminant levels in higher trophic-level organisms, and maternal transfer of PFAS in birds. These differences may also reflect changes in PFAS levels in a lake environment between years. When examining percent change between the earlier and later eggs tested, there was no overall temporal signal, as would be expected over the short time period of this study. Excluding the outliers, there was an average increase of  $0.95\% \pm 4.5\%$  between the earlier and later eggs. However, contaminant levels increased substantially between the earlier and later eggs in the cases of all of the outliers. The reasons for these increases are unclear, although the presence of a known different female in the cases of Christine Lake and Winnepesaukee—Varney Island must be taken into account. The variability between some loon eggs and the increases in PFAS levels in later eggs seen in some of the

lakes/territories indicates the importance of continued monitoring to assess changes in PFAS levels in New Hampshire's lakes.

#### *Effects levels and PFOS levels by lake/loon territory*

PFOS has historically been considered the dominant type of PFAS found in wildlife tissues (Newsted et al. 2005). Although PFOS levels seem to be declining in bird eggs following its phase-out in 2002 (Custer et al. 2013, Ericksson et al. 2016), PFOS remains the best-studied PFAS analyte to date in wildlife. Despite this, data on effects are contradictory. While laboratory dosing studies have often set high threshold levels for reproductive impairment from PFOS (>5,000 ng/g ww; studies summarized in Custer et al. 2014, Su et al. 2017, Wu et al. 2020), two field studies investigating Tree Swallows found reduced hatching success at 150 ng/g ww (Custer et al. 2012, Custer et al. 2014). Another study found no reproductive effects of PFOS on Tree Swallows at levels up to 4.5 times higher than the 150 ng/g level (Custer et al. 2019). Given the contradictory nature of studies on the possible effects of PFAS on avian productivity (Wu et al. 2020, Custer 2021), the further uncertainty of possible effects on PFAS on loons specifically, and the complexities of assessing impacts in field studies due to co-occurring stressors including interacting effects of other contaminants (Braune et al. 2007, Bustnes et al. 2008, Letcher et al. 2010, Fischer et al. 2013, Bustnes et al. 2015, Huber et al. 2015), we use the level of 150 ng/g ww as the lowest observed effect level (LOEL) for the purposes of this report.

Loon eggs containing levels of PFOS above the 150 ng/g ww LOEL are found in lakes in the southern half of New Hampshire and are most concentrated in the southeastern part of the state (Fig. 7), likely due to the increased human population density and presence of industry. Eggs with the highest levels of PFOS, exceeding 300% of the LOEL, were collected from Arlington Mill Reservoir (Salem) and Lake Winnepesaukee. Lakes/loon territories in which the average PFOS levels of the two eggs exceeded the LOEL include (Table 2): Arlington Mill Reservoir (Salem), Baxter Lake (Farmington), Halfmoon Lake (Alton), Jenness Pond (Northwood), Massabesic Lake (Auburn), Mendums Pond (Barrington), Mirror Lake (Tuftonboro), Spofford Lake (Spofford), Squam Lake—Heron Cove (Center Harbor), Sunapee Lake (Sunapee), Swains Lake (Barrington), Lake Umbagog—Sunday Cove (Errol), all nine territories tested on Lake Winnepesaukee (see below), and Winnisquam Lake—Three Islands (Laconia). Seven lakes/loon territories had one of the two eggs included in the sample exceed the LOEL, although the average of the two eggs was lower than the LOEL (Table 2): Ayers Pond (Barrington), Indian Pond (Orford), Pleasant Lake (New London), Squam Lake—Five Finger Point (Sandwich), Squam Lake—Kimball Island (Center Harbor), Squam Lake—Sturtevant Cove (Center Harbor), Webster Lake (Franklin).

Elevated levels of PFAS were documented from all nine territories tested from Lake Winnepesaukee (Fig. 8). Levels of PFOS in all territories tested exceeded the LOEL, with one territory (Spectacle Island, Moultonborough) exceeding 500% of the LOEL and three territories (Braun Bay, Moultonborough; Langdon Cove, Moultonborough; and Copps Brook, Tuftonboro) exceeding 400% of the LOEL. The highest level of PFOS documented in the 144 eggs tested for



this study was the egg from Braun Bay in 2021, which contained 1016.3 ng/g ww PFOS (678% of LOEL), although an egg from Arlington Mill Reservoir in 2019 (previously tested by LPC) had 1310.0 ng/g ww PFOS. The highest level of PFOS documented in a New Hampshire loon egg was 1400.0 ng/g ww from an egg at Canobie Lake (Windham) in 2016 (Grade and Vogel 2021). For eggs from Lake Winnepesaukee in the present dataset, only the egg from the Evergreen territory (Moultonborough) in 2021 did not exceed the LOEL for PFOS, with a level of 137.5 ng/g ww (92% of LOEL). The geometric mean of total PFAS for the 16 eggs tested from Lake Winnepesaukee was  $1033.4 \pm 108.0$  ng/g ww (range: 325.2-1938.0 ng/g ww).

In addition to the eggs included in the current dataset, LPC previously tested single eggs from two Winnepesaukee territories at Black Cove (2018; Meredith) and Breezy Island (2014; Meredith). PFOS levels were 905.0 ng/g ww (603% of LOEL) in the Black Cove egg and 834.0 ng/g ww (556% of LOEL) in the Breezy Island egg (Grade and Vogel 2021). Although the Breezy Island egg is outside the study period, the levels of PFOS in both of these eggs are in keeping with levels seen in the present study, and these territories are included in Figure 4 to provide a more complete sense of the distribution of elevated levels of PFOS/PFAS in Lake Winnepesaukee.

The reasons for the elevated levels of PFAS in loon eggs from Lake Winnepesaukee are unclear, particularly when considering the size and volume of the lake. Further study is needed to understand the source(s) of PFAS in Winnepesaukee.

### **Management Implications**

LPC's previous research and the results reported here indicate the importance of loons, a long-lived species at the top of aquatic food webs, as indicators of localized PFAS levels and the health of aquatic ecosystems (Strong 1990, Evers 2006). Further investigation is needed into the many broad questions regarding PFAS in aquatic environments and of the effects of PFAS on wildlife. Previous studies by LPC (Grade et al., in prep.) found no effects of PFAS on loon egg size or reproductive success; however, the small sample size and fewer congeners tested in this previous study may have limited our ability to detect potential effects. The results of this study suggest the importance of additional actions to evaluate the impacts of PFAS in New Hampshire for human and wildlife health. We recommend:

- 1) Fish sampling on Lake Winnepesaukee and other lakes with elevated levels of PFAS in loon eggs to identify possible risks to human health from consumption of fish from the lakes.
- 2) Investigation of the potential source(s) of PFAS in Lake Winnepesaukee.
- 3) Further testing of PFAS in loon eggs or other high trophic-level species, particularly in the southern half of the state, to identify other potential lakes of concern, as well as from additional loon territories/areas of Lake Winnepesaukee.

- 4) Ongoing monitoring of PFAS in loon eggs or other high trophic level species to identify temporal changes in levels of PFAS in New Hampshire's lakes.
- 5) Testing of historical loon egg samples for a greater number of PFAS analytes. Given that eggs tested prior to 2019 were analyzed for a limited number of analytes, testing of samples prior to that date for the broader suite of analytes will increase understanding of levels, temporal trends, and possible effects of PFCAs and 7:3 FTCA in loon eggs.
- 6) Research and investigations into the movement of PFAS through aquatic food webs, including differences in trophic levels and the breakdown of precursors into potentially more toxic and bioavailable substances.

### **Next Steps (LPC)**

The Loon Preservation Committee is researching the relationship between PFAS levels from eggs in this dataset and eggs previously tested by LPC on egg morphometrics and loon productivity for publication in a peer-reviewed journal. We will continue to test a limited number of inviable loon eggs from failed nests (4-6 per year) as funds allow for PFAS and a broad suite of legacy and emerging contaminants.

### **Acknowledgments**

This study was funded by New Hampshire Department of Environmental Services through the State of New Hampshire's Emerging Contaminants Fund. We thank Ken Edwardson and Jon Petali of NHDES for their assistance and support of this project and Ted Diers, David Neils, Tracie Sales, and Sara Steiner of NHDES for their advice, support, and efforts to identify contaminants of concern in New Hampshire. We also thank Caroline Hughes of Loon Preservation Committee for creating maps and providing other project support. This report references eggs previously tested by LPC, and we acknowledge funding and other assistance from many individuals and foundations that supported the collection, testing, and analyses of those eggs.

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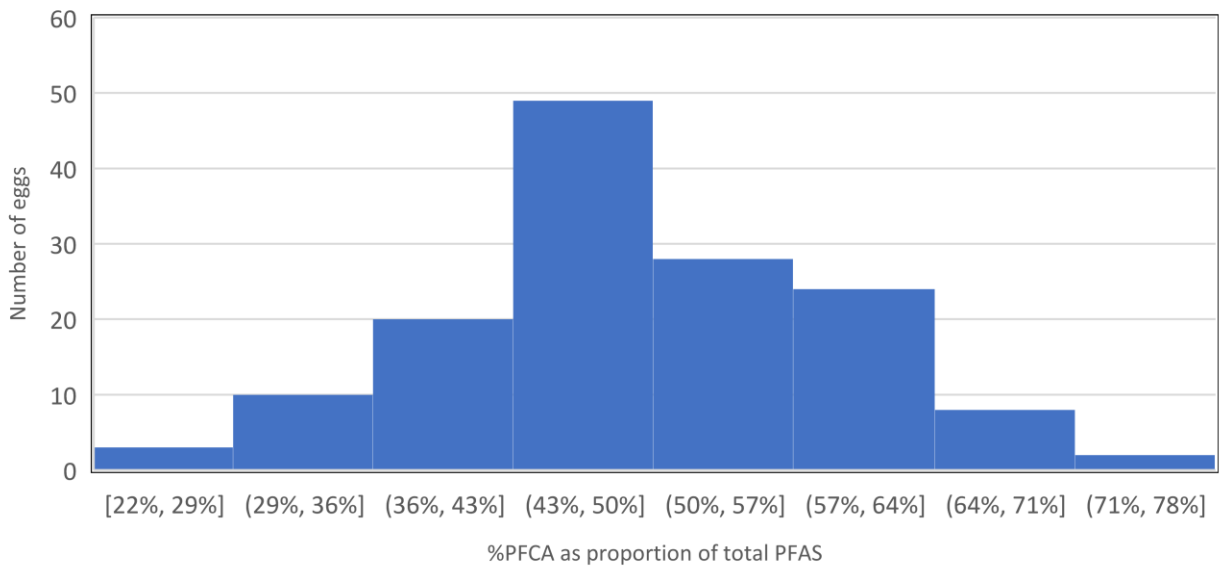


Figure 1: Histogram of percentage of PFCAs as proportion of total PFAS in loon eggs. PFCAs account for >40% of the total PFAS profile in 84.0% of eggs.

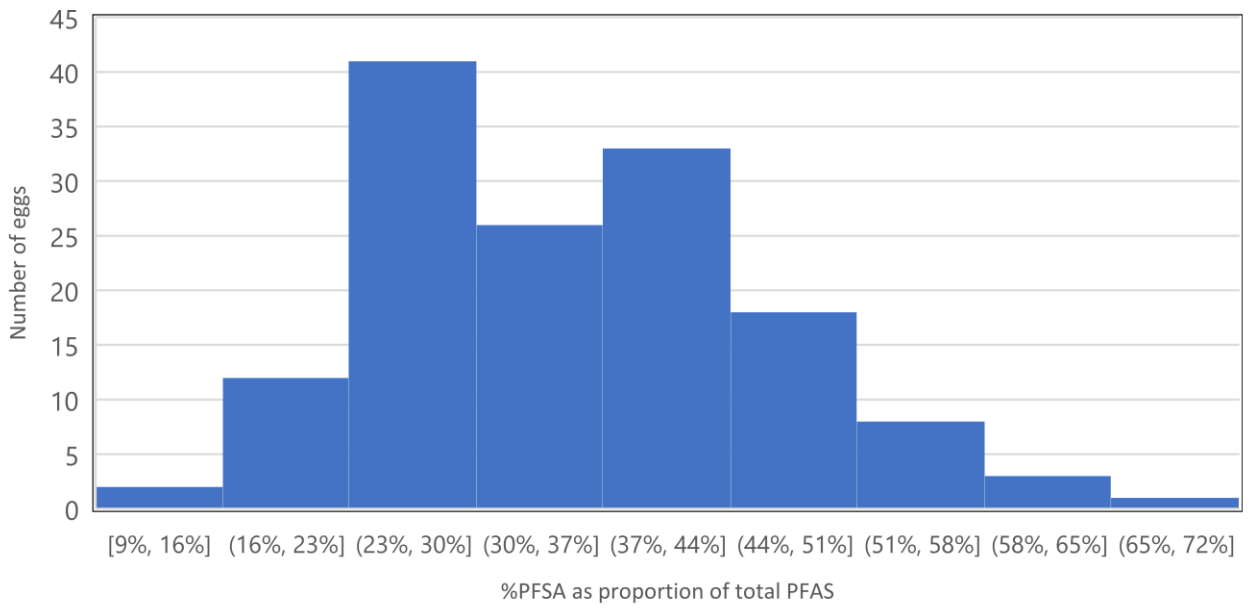


Figure 2: Histogram of percentage of PFSAs as proportion of total PFAS in loon eggs. PFSAs account for >40% of the total PFAS profile in 34.7% of eggs.

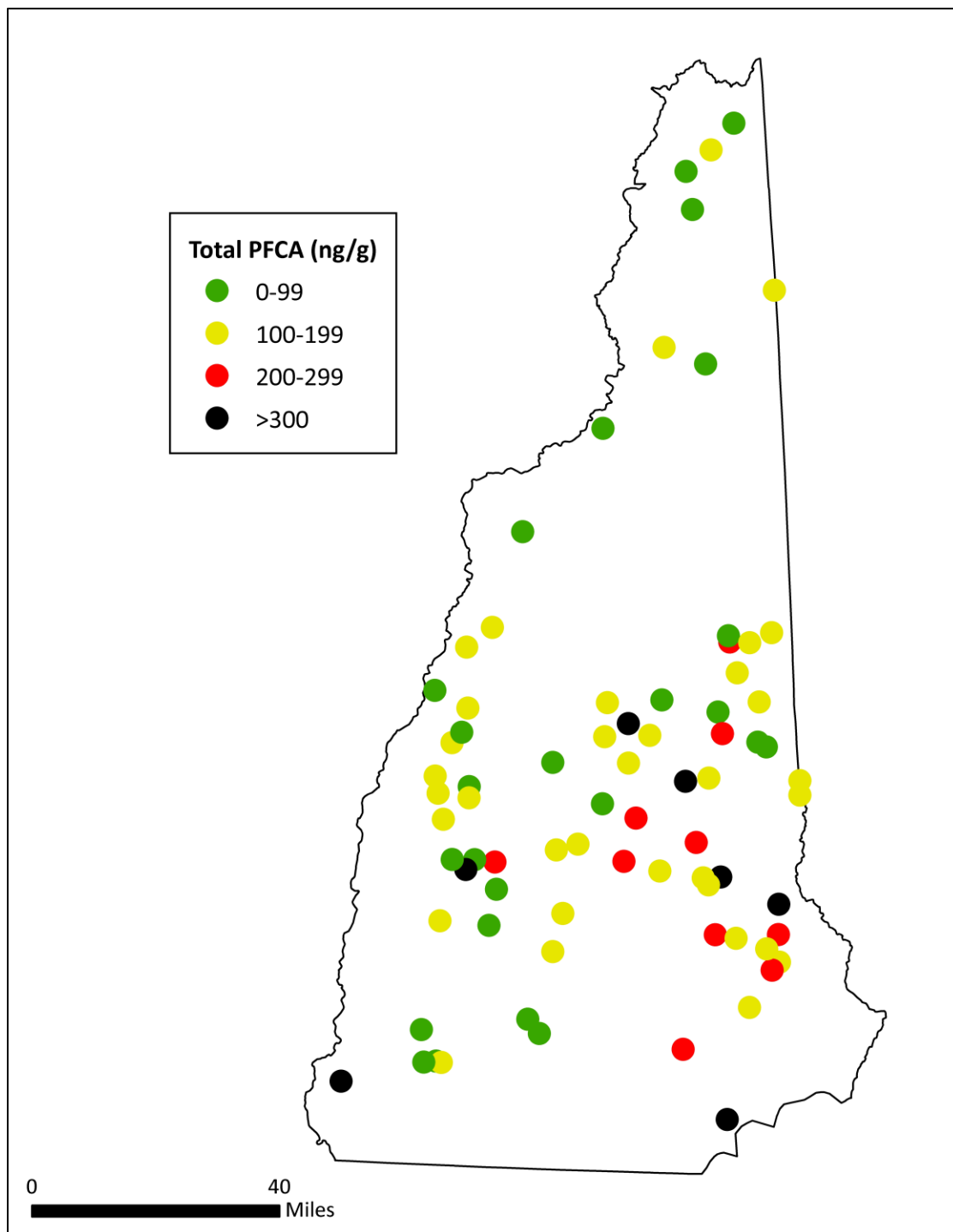


Figure 3: Distribution of total PFCA levels in loon eggs in New Hampshire (ng/g ww). The highest levels of PFCAs were found in Lake Winnepesaukee (see Figure 4).



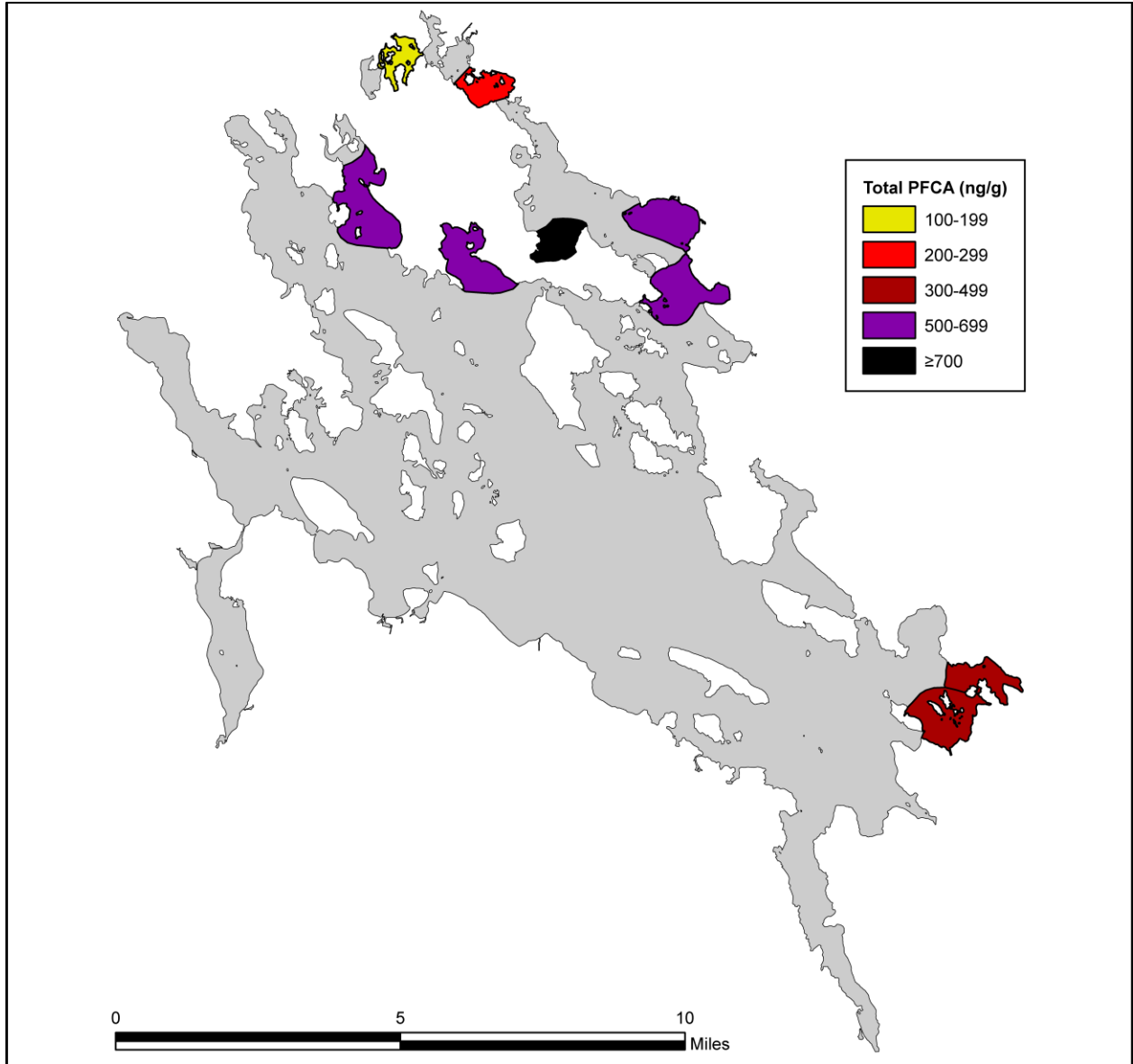


Figure 4: Distribution of levels of total PFCAs in loon eggs in Lake Winnepesaukee. The maximum concentration of 839.7 ng/g ww documented in this dataset was from an egg at Braun Bay, Winnepesaukee (Moultonborough).

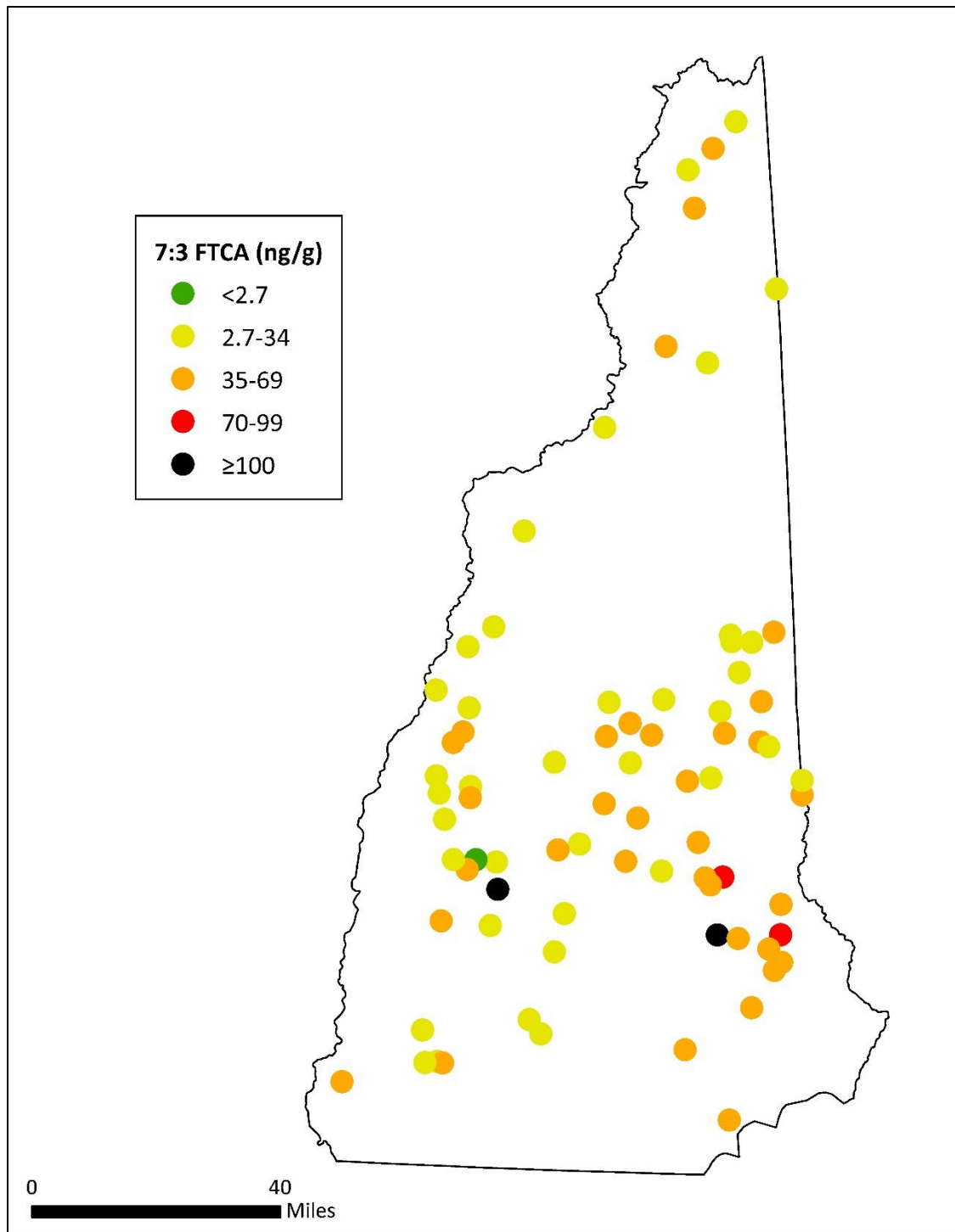


Fig. 5: Distribution of 7:3 FTCA levels in loon eggs in New Hampshire. To our knowledge, the highest level of 7:3 FTCA previously documented in bird eggs was 2.7 ng/g ww (Eriksson et al. 2016, Jouanneau et al. 2022). Levels of 7:3 FTCA in individual eggs from Ayers Pond (Barrington), Jenness Pond (Northwood), Kezar Lake (Sutton), and Lake Winnepesaukee exceeded 100 ng/g ww.

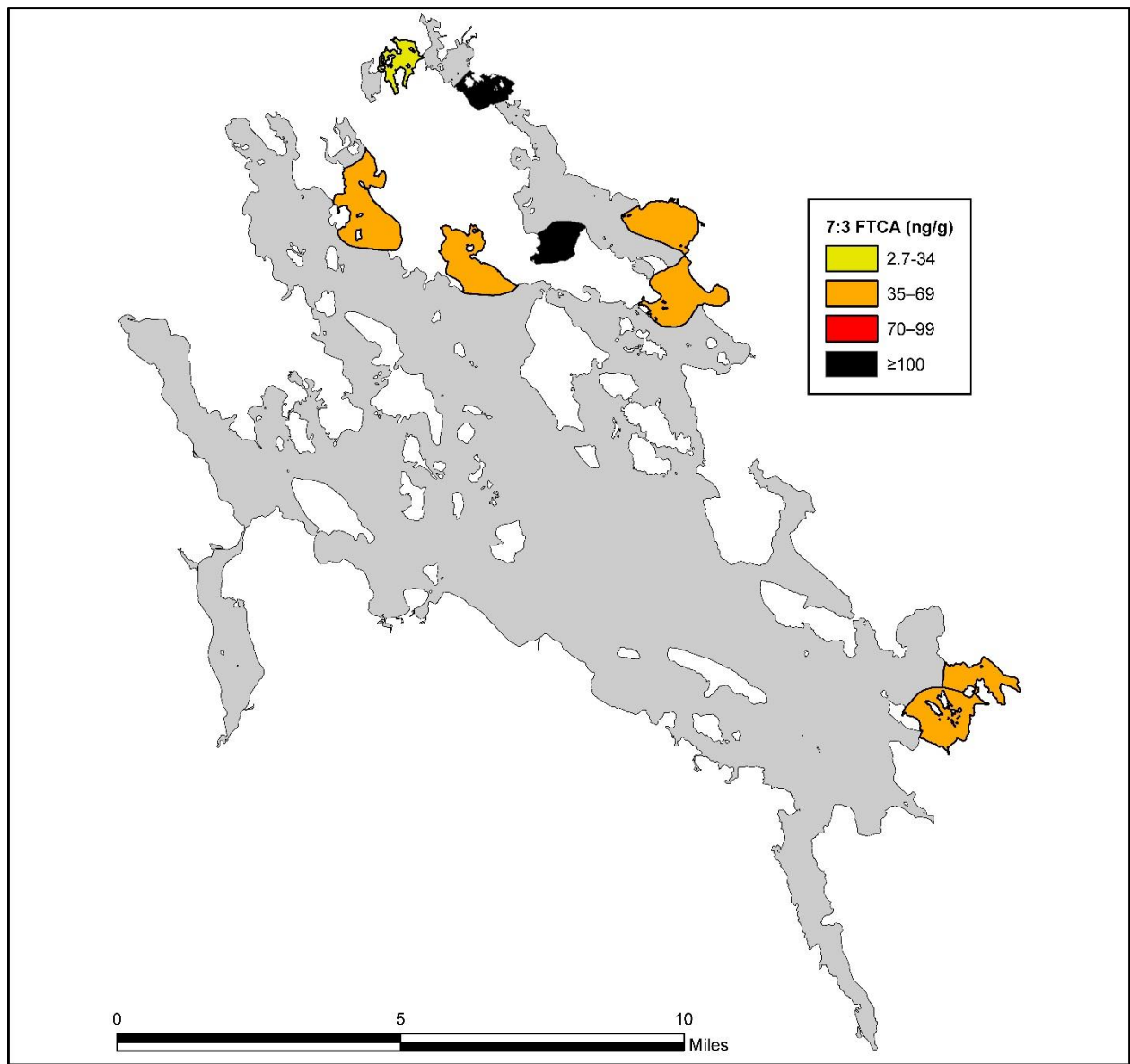


Figure 6: Distribution of levels of 7:3 FTCA in loon eggs in Lake Winnepesaukee. No eggs from Lake Winnepesaukee were below the previously-documented maximum level of 2.7 ng/g ww for 7:3 FTCA in a bird egg (Jouanneau et al. 2022). The maximum concentration of 174.0 ng/g ww documented in this dataset was from an egg at Lincoln Island, Winnepesaukee (Moultonborough).

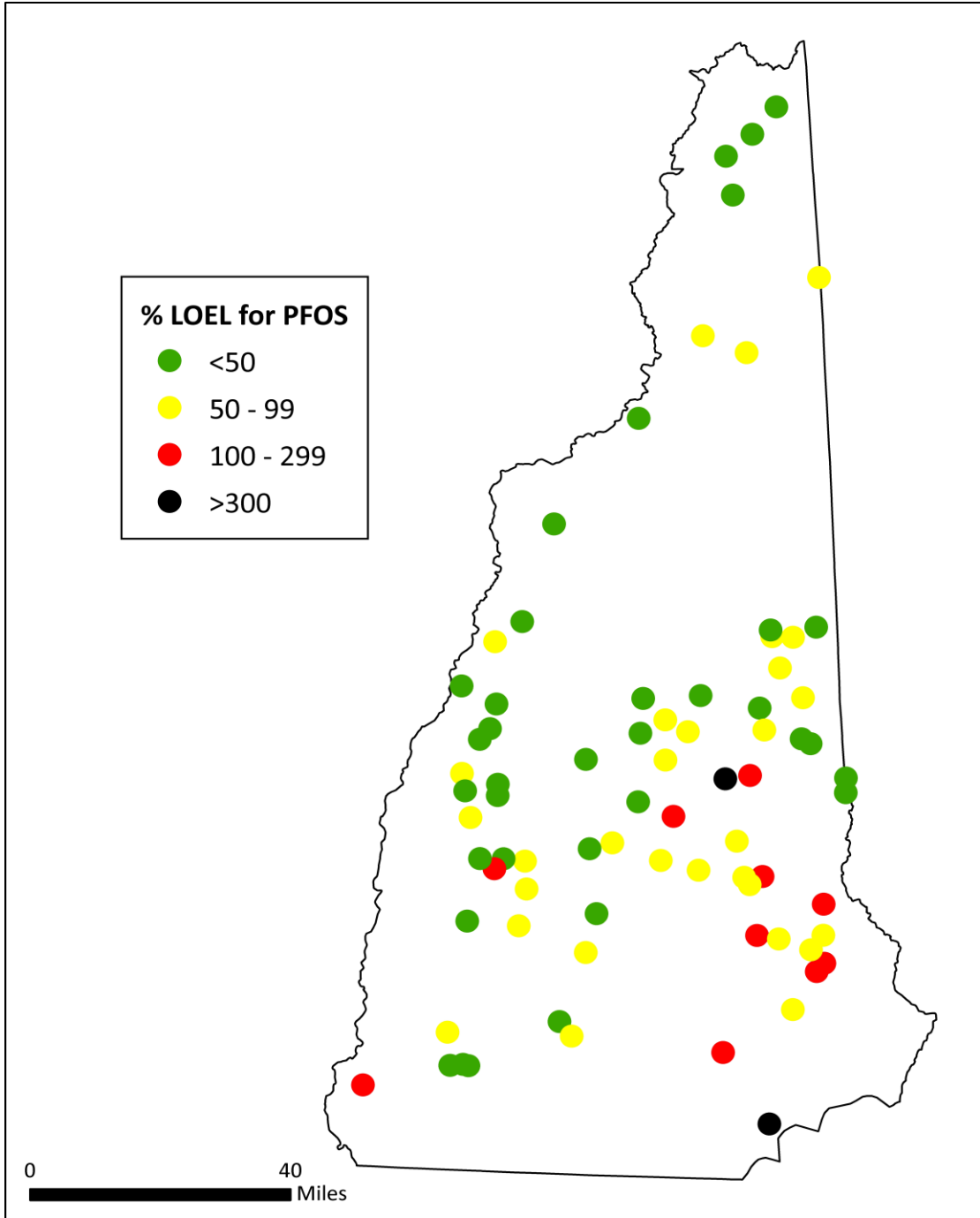


Figure 7: Distribution of PFOS levels in loon eggs across New Hampshire as a percentage of a lowest observed effect level (LOEL) of 150 ng/g ww.

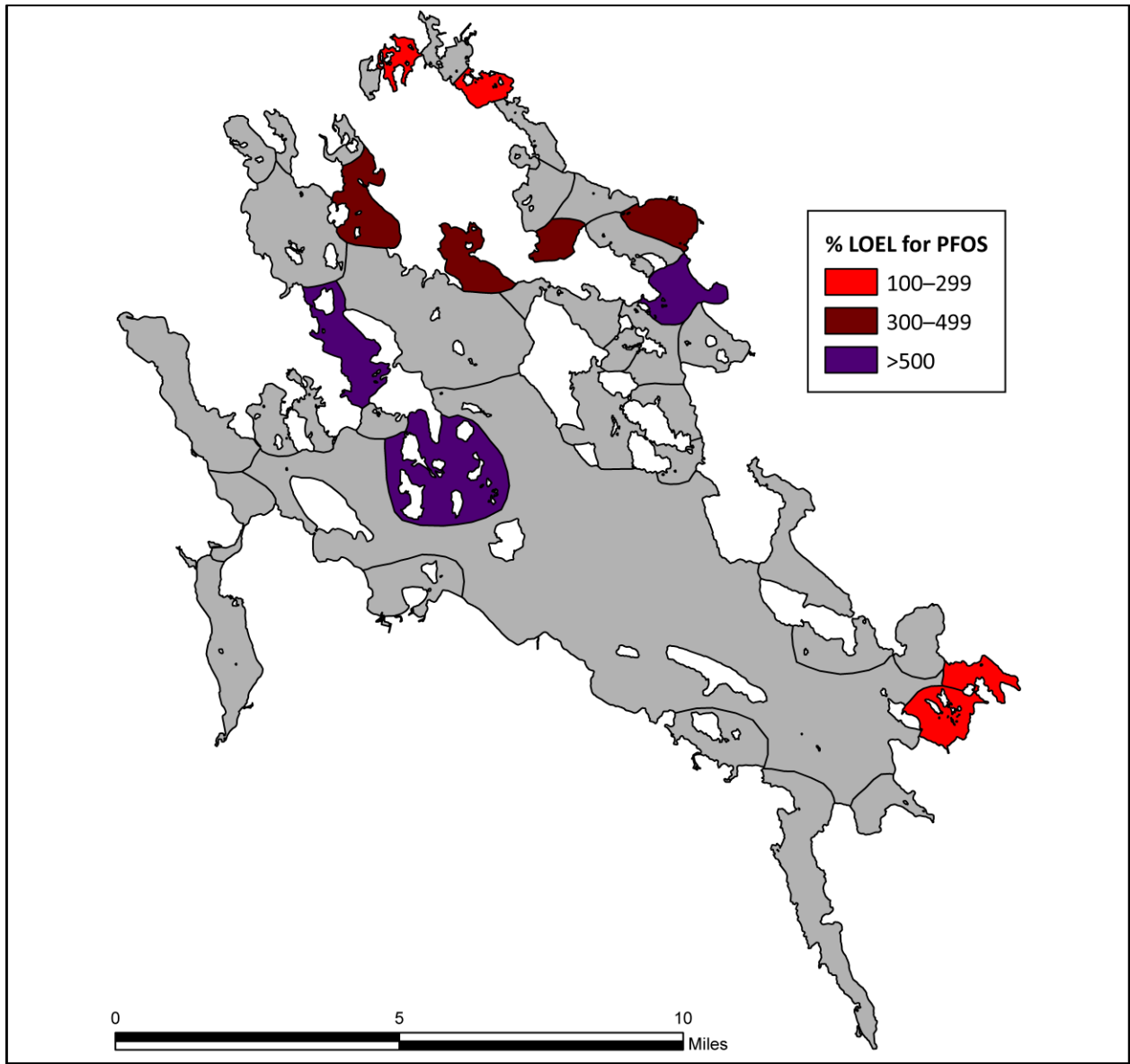


Figure 8: Distribution of PFOS in tested loon territories on Lake Winnepesaukee as a percentage of a lowest observed effect level (LOEL) of 150 ng/g ww. In addition to eggs from the current dataset, the map also includes single eggs from two territories tested by the Loon Preservation Committee in 2014 and 2018.

Table 1: PFAS levels in Common Loon eggs from New Hampshire lakes (geometric mean  $\pm$  standard error [range]). All results are reported in nanograms/gram wet weight. ND = Non-detect. If  $>50\%$  of data is less than the reporting limit, only the range is reported (Schmutz et al. 2009). -- = detection in  $<50\%$ , M = median for detection  $>50\%$  and  $<85\%$ . The geometric mean and standard error are reported if detection is  $>85\%$ .

PFAS analyte group	Congeners	Levels	% Total PFAS	% Total PFCA	% Detection	
Perfluoroalkyl carboxylates (PFCAs)	PFBA	-- (ND-4.4)	$0.2 \pm 0.1$ (0.0-3.4)	$0.5 \pm 0.1$ (0.0-7.9)	26.4	
	PFPeA	-- (ND-0.3)	$0.0 \pm 0.0$ (0.0-0.1)	$0.0 \pm 0.0$ (0.0-0.2)	0.7	
	PFHxA	-- (ND-2.3)	$0.0 \pm 0.0$ (0.0-0.9)	$0.1 \pm 0.0$ (0.0-2.2)	16.0	
	PFHpA	-- (ND-0.7)	$0.0 \pm 0.0$ (0.0-0.3)	$0.1 \pm 0.0$ (0.0-0.5)	37.5	
	PFOA	$0.7 \pm 0.1$ (ND-4.2)	$0.3 \pm 0.0$ (0.0-1.9)	$0.6 \pm 0.1$ (0.0-5.9)	92.4	
	PFNA	$2.8 \pm 0.3$ (0.6-17.7)	$1.0 \pm 0.1$ (0.2-3.4)	$2.1 \pm 0.1$ (0.4-7.0)	100	
	PFDA	$13.4 \pm 1.9$ (1.6-145.5)	$4.6 \pm 0.1$ (1.0-9.4)	$9.6 \pm 0.3$ (3.6-22.3)	100	
	PFUnA	$40.8 \pm 4.2$ (5.1-290.0)	$13.9 \pm 0.3$ (3.2-23.1)	$28.1 \pm 0.5$ (14.6-42.2)	100	
	PFDoA	$27.9 \pm 2.8$ (3.7-162.4)	$9.5 \pm 0.2$ (3.8-18.4)	$19.1 \pm 0.3$ (12.1-26.9)	100	
	PFTTrDA	$42.6 \pm 3.1$ (9.1-196.3)	$14.6 \pm 0.4$ (4.6-26.7)	$29.3 \pm 0.4$ (15.5-42.9)	100	
	PFTeDA	$15.1 \pm 1.1$ (4.3-69.3)	$5.2 \pm 0.1$ (2.2-12.2)	$10.6 \pm 0.3$ (5.1-24.2)	100	
	Perfluoroalkyl sulfonates (PFSAs)	PFBS	ND	0.0		0.0
		PFPeS	ND	0.0		0.0
PFHxS		$0.3 \pm 0.0$ (ND-3.3)	$0.2 \pm 0.0$ (0.0-3.8)		97.2	
PFHpS		$0.3 \pm 0.1$ (ND-5.6)	$0.1 \pm 0.0$ (0.0-0.4)		92.4	
PFOS		$98.7 \pm 14.7$ (19.0-1016.3)	$33.9 \pm 0.9$ (8.4-64.9)		100	
PFNS		M = 0.1 (ND-1.9)	$0.0 \pm 0.0$ (0.0-0.1)		66.0	
PFDS		$2.6 \pm 0.5$ (0.4-39.9)	$1.1 \pm 0.1$ (0.2-5.7)		100	
PFDoS		-- (ND-0.3)	$0.0 \pm 0.0$ (0.0-0.1)		9.0	
Fluorotelomer sulfonates	4:2 FTS	ND	0.0		0.0	
	6:2 FTS	-- (ND-7.6)	$0.3 \pm 0.1$ (0.0-6.1)		17.4	
	8:2 FTS	-- (ND-1.8)	$0.0 \pm 0.0$ (0.0-0.6)		4.2	

PFAS analyte group	Congeners	Levels	% Total PFAS	% Total PFCA	% Detection
Perfluorooctane sulfonamides	PFOSA	0.4 ± 0.0 (ND-1.9)	0.2 ± 0.0 (0.0-0.8)		98.6
	N-MeFOSA	-- (ND-0.1)	0.0 ± 0.0 (0.0-0.1)		2.1
	N-EtFOSA	ND	0.0		0.0
Perfluorooctane sulfonamidoacetic acids	MeFOSAA	-- (ND-0.7)	0.0 ± 0.0 (0.0-0.1)		28.5
	EtFOSAA	-- (ND-0.2)	0.0 ± 0.0 (0.0-0.0)		8.3
Perfluorooctane sulfonamidoethanols	N-MeFOSE	-- (ND-1.6)	0.0 ± 0.0 (0.0-0.6)		1.4
	N-EtFOSE	M = 1.4 (ND-46.5)	1.9 ± 0.3 (0.0-15.2)		57.6
Ether carboxylates	HFPO-DA	ND	0.0		0.0
	ADONA	ND	0.0		0.0
	NFDHA	ND	0.0		0.0
	PFMPA	ND	0.0		0.0
	PFMBA	-- (ND-0.3)	0.0 ± 0.0 (0.0-0.2)		1.4
Fluorotelomer carboxylates	3:3 FTCA	ND	0.0		0.0
	5:3 FTCA	ND	0.0		0.0
	7:3 FTCA	32.6 ± 2.3 (ND-174.0)	12.9 ± 0.6 (0.0-36.7)		98.6
Ether sulfonates	PFEESA	-- (ND-0.4)	0.0 ± 0.0 (0.0-0.3)		2.8
	9Cl-PF3ONS	-- (ND-0.7)	0.0 ± 0.0 (0.0-0.1)		0.7
	11Cl-PF3OUdS	-- (ND-0.5)	0.0 ± 0.0 (0.0-0.1)		1.4
	<b>Total PFCA</b>	<b>148.1 ± 12.9</b> <b>(28.3-839.7)</b>	<b>49.4 ± 0.8</b> <b>(22.1-72.6)</b>		
	<b>Total PFAS</b>	<b>306.3 ± 28.2</b> <b>(85.2-1938.0)</b>			

Table 2: Total PFAS, total PFCA, and PFOS levels by loon territory or lake. Bolded PFOS values indicate a lake average >150 ng/g ww, the lowest observed effect level (LOEL) used in this report for negative health or reproductive effects. Italicized PFOS values indicate that one of the two samples from a given lake exceeded 150 ng/g ww, although the lake average of two eggs did not exceed the LOEL. All values are in nanograms/gram wet weight.

Lake (Territory)	Town	Total PFAS	Total PFCAs	PFOS
Arlington Mill Reservoir <sup>‡</sup>	Salem	1394.5	352.8	<b>999.5</b>
Ayers Pond	Barrington	454.9	206.3	<i>145.1</i>
Balch Pond	Wakefield	242.5	113.8	74.6
Baxter Lake	Farmington	585.3	306.4	<b>205.9</b>
Bolster Pond	Sullivan	198.5	76.9	85.3
Bow Lake	Strafford	314.7	179.1	77.5
Broad Bay*	Ossipee	333.0	170.6	94.4
Cedar Pond <sup>†</sup>	Milan	158.6	35.0	77.7
Childs Bog <sup>†</sup>	Harrisville	250.5	131.1	67.6
Christine Lake	Stark	265.7	111.2	<i>95.7</i>
Clark Pond	Canaan	170.1	72.1	47.9
Clement Pond	Hopkinton	292.1	167.6	98.7
Connecticut Lake, First*	Pittsburg	246.4	126.9	64.3
Connecticut Lake, Second <sup>†</sup>	Pittsburg	224.4	91.6	63.9
Conner Pond	Ossipee	131.4	88.3	25.3
Conway Lake	Conway	250.6	154.4	46.6
Dan Hole Pond	Tuftonboro	390.2	240.3	78.1
Deering Reservoir <sup>†</sup>	Deering	143.3	85.2	40.1
Diamond Pond, Big <sup>‡</sup>	Stewartstown	114.1	50.8	34.9
Duncan Pond <sup>†</sup>	Ossipee	193.7	74.0	43.0
Eastman Pond <sup>†</sup>	Grantham	257.9	100.9	122.0
Goose Pond	Canaan	240.0	111.4	65.0
Grafton Pond	Grafton	220.6	111.4	58.2
Great East Lake	Wakefield	285.0	147.3	49.0
Gunnison Lake	Goshen	233.7	126.2	62.0
Halfmoon Lake <sup>†</sup>	Alton	586.0	326.7	<b>174.0</b>
Hermit Lake	Sanbornton	126.9	57.4	30.7
Highland Lake*	Andover	281.4	141.2	73.8
Indian Pond	Orford	302.7	148.5	<i>112.3</i>
Iona Lake <sup>†</sup>	Albany	160.0	77.8	45.6
Jeness Pond <sup>†</sup>	Northwood	654.6	296.4	<b>199.6</b>
Kanasatka Lake	Moultonborough	344.9	157.1	138.4
Kezar Lake	Sutton	318.7	84.5	116.5
Knowles Pond	Northfield	380.0	216.8	112.6
Lake Francis (South Cove)	Clarksville	107.9	41.0	45.4
Lake Francis (Deadwater Stream)*	Clarksville	206.9	91.0	54.6
Ledge Pond <sup>†</sup>	Sunapee	136.1	85.4	19.0



Lake (Territory)	Town	Total PFAS	Total PFCAs	PFOS
Little Squam Lake	Ashland	248.3	158.2	29.6
Martin Meadow Pond <sup>†</sup>	Lancaster	113.2	48.9	45.5
Mascoma Lake <sup>‡</sup>	Enfield	257.0	131.3	88.8
Massabesic Lake <sup>*</sup>	Auburn	783.7	281.7	<b>405.5</b>
Mendums Pond <sup>‡</sup>	Barrington	438.4	251.7	<b>161.1</b>
Mirror Lake	Tuftonboro	545.1	173.0	<b>327.5</b>
Newfound Lake	Bristol	133.6	73.9	38.0
Nippo Pond	Barrington	320.4	147.1	120.5
Pawtuckaway Lake	Nottingham	282.8	126.2	120.0
Pea Porridge Pond, Middle	Madison	298.4	146.2	116.4
Pearl Lake <sup>†</sup>	Lisbon	93.3	51.0	32.5
Perch Pond	Campton	203.2	128.3	49.2
Pleasant Lake <sup>*</sup>	Fracestown	135.6	84.0	75.7
Pleasant Lake	New London	386.3	203.6	<i>145.1</i>
Post Pond <sup>†</sup>	Lyme	148.5	69.5	72.4
Red Hill Pond <sup>‡</sup>	Sandwich	177.6	96.1	65.5
Reservoir Pond <sup>†</sup>	Dorchester	167.1	113.7	39.3
Shellcamp Pond <sup>†</sup>	Gilmanton	268.6	109.5	138.6
Silver Lake <sup>†</sup>	Harrisville	85.2	28.3	20.9
Silver Lake	Madison	267.3	161.2	76.5
Smith Pond	Enfield	172.7	105.1	39.6
Spectacle Pond <sup>†</sup>	Enfield	193.8	86.3	72.4
Spofford Lake <sup>†</sup>	Spofford	761.6	341.0	<b>345.8</b>
Squam Lake (Five Finger Point) <sup>‡</sup>	Sandwich	518.7	357.7	<i>119.4</i>
Squam Lake (Heron Cove) <sup>*</sup>	Center Harbor	442.5	281.9	<b>172.5</b>
Squam Lake (Kimball Island) <sup>‡</sup>	Center Harbor	511.8	340.6	<i>129.7</i>
Squam Lake (Sturtevant Cove) <sup>**</sup>	Center Harbor	403.2	273.0	<i>135.4</i>
Sunapee Lake	Sunapee	538.7	314.3	<b>167.5</b>
Sunapee Lake, Little	New London	104.6	59.9	40.2
Suncook Lake, Lower	Barnstead	325.3	158.5	95.5
Suncook Lake, Upper	Barnstead	356.3	156.4	89.8
Sunset Lake <sup>†</sup>	Alton	400.8	249.2	90.0
Swains Lake <sup>†</sup>	Barrington	451.2	165.8	<b>203.0</b>
Tarleton Lake <sup>†</sup>	Piermont	278.8	189.8	63.7
Todd Lake	Newbury	178.4	66.2	84.1
Umbagog (Rapid River)	Errol	162.2	88.1	56.0
Umbagog (Sunday Cove)	Errol	465.0	259.0	<b>165.6</b>
Waukegan Lake	Meredith	319.1	159.6	115.0
Webster Lake	Franklin	246.9	108.2	<i>115.5</i>
White Pond <sup>†</sup>	Ossipee	85.6	28.3	37.8

Lake (Territory)	Town	Total PFAS	Total PFCA <sub>s</sub>	PFOS
Whitton Pond <sup>†</sup>	Albany	375.6	215.8	116.0
Winnepocket Lake <sup>†</sup>	Webster	269.1	177.1	67.4
Winnepesaukee (Blueberry)	Moultonborough	1205.7	572.0	<b>550.8</b>
Winnepesaukee (Braun Bay)	Moultonborough	1431.6	636.4	<b>728.2</b>
Winnepesaukee (Copps Brook)	Tuftonboro	1262.5	569.0	<b>614.2</b>
Winnepesaukee (Evergreen) *	Moultonborough	325.2	155.0	<b>168.4</b>
Winnepesaukee (Langdon Cove) ‡	Moultonborough	1531.4	745.7	<b>699.8</b>
Winnepesaukee (Lincoln Island)	Moultonborough	703.3	297.0	<b>266.3</b>
Winnepesaukee (Spectacle Island)	Moultonborough	1535.7	647.4	<b>812.1</b>
Winnepesaukee (Varney Island)	Wolfeboro	1004.8	486.5	<b>435.5</b>
Winnepesaukee (Worcester)	Wolfeboro	948.3	489.5	<b>376.0</b>
Winnisquam Lake (Winnisquam)	Belmont	253.0	131.2	84.2
Winnisquam Lake (Three Islands)	Laconia	579.6	268.8	<b>229.8</b>
Woodward Pond	Roxbury	204.1	96.2	73.0

\*Eggs from two separate loon territories.

<sup>†</sup>Values from single egg.

<sup>‡</sup>Average values include one egg previously tested by LPC within study period and not part of present sample.

\*Average value for PFOS includes one egg previously tested by LPC within study period and not part of the present sample. Total PFAS and total PFCA levels represent single egg from present sample.

## **Appendix A: Laboratory Analytical Methods**

Eggs were analyzed at SGS AXYS Analytical Services, Ltd. (Sidney, BC, Canada) using SGS AXYS Method MLA-110. For all analyses, <sup>13</sup>C-labelled surrogate standards were added to the samples prior to extraction. Blanks, spikes, and duplicates were analyzed for quality assurance and control for each batch.

After spiking the samples with surrogate standards, samples were extracted using methanolic potassium hydroxide solution, then with acetonitrile, and then again with methanolic potassium hydroxide solution. Supernatants from each treatment were combined and treated with ultra-pure carbon powder before being evaporated, diluted with water, and cleaned by solid phase extraction. The extract was analyzed using ultrahigh performance liquid chromatography with a reversed phase C18 column coupled to a triple quadrupole mass spectrometer (LC-MS/MS). The analysis was run in Multiple Reaction Monitoring in negative electrospray ionization mode. Final sample concentrations were determined by isotope dilution/internal standard quantification. Minimum reporting limits for perfluorinated organic compounds ranged from 0.07-5.69 ng/g ww. The average percent recovery was 50.2% (range: Non-quantifiable-241%).