This following except was taken from "Lake Wildwood 2019 Microbial Monitoring Program and Response to 2017 E. coli 0157:H7 Outbreak" beginning on p. 38

For additional information, please refer to the entire 63 page report located at:

On August 13, 2017, approximately one month after the LWW EcO157:H7 outbreak, 50- L beach water samples were concentrated by Nevada County Environmental Health personnel and shipped to the Centers for Disease Control laboratory in Atlanta GA for analysis. The samples were tested for the presence of EcO157:H7 and for microbial source tracking (MST) markers to help assess the probable source of the beach contamination. The genetic MST markers tested were human, deer and geese. Only goose markers were detected. This was the first evidence that geese may have played a role in the outbreak. Additional evidence was subsequently accumulated establishing the role of geese during the 2018 LWW monitoring and investigation conducted by Lake Wildwood Association (Yanko et al. 2019). Geese were documented to be the predominant source of shoreline contamination at the beaches. Additional evidence was developed that the geese were carrying EcO157, but it was not possible to determine where or how the geese became infected. One theory was that geese may pick up O157:H7 when foraging at nearby cattle pastures. Another possibility was that geese pick up STEC from water in the lake. In either event, a few infected resident geese could then continue to infect others in the flock by defecating on the park lawns where they regularly graze.

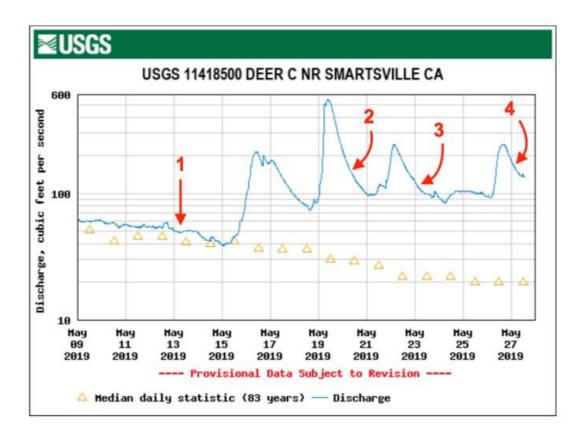
It was also reported in the 2018 report that Meadow Park Creek was chronically contaminated and the microbial contamination frequently included EcO157:H7 (Yanko et al., 2019). Based on the initial reports of creek contamination with STEC, Nevada County Lake Wildwood 2019 Microbial Monitoring Program and Response to 2017 E. coli O157:H7 Outbreak 39 Environmental Health (NCEH) coordinated with Centers for Disease Control (CDC) to analyze samples from Meadow Park Creek for Microbial Source Tracking (MST) markers to help assess the source of the contamination. Five samples including both water and sediment were collected by NCEH and shipped to CDC. These were tested for EcO157 and MST markers. Locations sampled were Meadow Park Beach shoreline, Meadow Park Creek at the Bridge, and Meadow Park Creek approximately 0.5 mile upstream (sediment only). CDC reported that E. coli O157:H7 was detected in water and sediment in the creek at the bridge location, and in the sediment at the upstream location. Isolates from those two locations had matching Pulse-Field Gel Electrophoresis (PFGE) patterns, and whole genome sequencing (WGS) demonstrated that the isolates were clonal, with 0-1 single nucleotide polymorphisms (SNPs) indicating a common source. E. coli O157:H7 was not detected in either water or sand at the beach location.

In the creek at Meadow Park Bridge, Human and Ruminant MST markers were detected in the water; Goose, Deer and Cow markers were not detected. The corresponding creek

sediment sample was negative for all of the animal markers and inconclusive for the human marker. The upstream sediment sample was negative for all markers, although it had been positive for O157:H7. At the Meadow Park Beach location, the water was positive for goose markers, but negative for all other markers, and sand was negative for all markers (cow not tested in the sand). The CDC report did not provide the qPCR data and did not provide any discussion or conclusions about the MST results. The full CDC report was included in the 2018 report, Appendix H (Yanko et. al. 2019). Without the qPCR results, it is difficult to assess the relative significance of the reported ruminant and human marker results.

In general, the CDC MST data appeared to be generally inconclusive. It is possible there could be some human contamination from septic systems outside of the LWW property or the sewer lines within LWW. That area of the sewage system was not examined after the outbreak because it was not located close to Commodore Beach where the outbreak occurred. Finding Ruminant markers, but not Deer, is surprising because the LWW Deer population is large and the Meadow Park stream flows for about a mile through a greenbelt area. There could be some ruminant animals such as goats or sheep in the properties outside of LWW producing the Ruminant signal. It also seemed unusual that the markers were detected in the creek water but not in the streambed sediments. The long term persistence of specific enteric pathogens compared to selected fecal MST markers in sediments is not well established. For the beach shoreline water sample, only goose markers were detected. That was consistent with the MST analyses conducted soon after the outbreak and the subsequent conclusions that geese were the primary source of contamination at the beaches.

Based on the earlier analyses discussed above, another effort was made to determine the source of contamination in Meadow Park Creek during 2019. Samples were collected and concentrated on-site by LWW project personnel. The sample filters were shipped to SCCWRP and analyzed for MST markers under the direction of Dr. John Griffith. Three samples were collected on May 27, 2019 and concentrated on site as described in the Methods Section of this report. The timing of the sampling was related to a series of spring rainstorms. Figure 8 shows a flow hydrograph for Deer Creek downstream from Lake Wildwood for a period of 18 days that Lake Wildwood 2019 Microbial Monitoring Program and Response to 2017 E. coli O157:H7 Outbreak 40 guided the Meadow Park Creek sampling. USGS maintains a flow measuring station downstream from LWW that reports real-time data. Similar flow data for the smaller Meadow Park Creek or Wildwood Creek that were sampled for MST were not available. The flows from these smaller watersheds would be substantially smaller than Deer Creek, but timing of the storm flows should be reasonably similar. Figure 8: Flow hydrograph for Deer Creek downstream from Lake Wildwood in May 2019.



Point #1 on Figure 8 is when the first EcO157 positive sample was collected. There had not been significant rainfall since about April 19-20 and flows were in a declining baseline condition, just slightly above the historic average (shown by the gold triangles). The first O157 positive was very weak, producing a barely perceptible signal on the Reveal test strip. A few days after that, a series of unseasonable spring storms started and flows rose quickly with each storm. Sample #2 was collected during the declining leg of the hydrograph the morning after the second storm, i.e. it was still in the increased flow period. That sample showed a very strong positive for O157 with the Reveal test. Sample #3 was the same scenario; the sample was collected the morning following a storm while still in the elevated flow stage, again producing a very strong positive for O157. That was the rationale for trying to capture the last of the forecast storms for source tracking analyses.

Samples were collected from Meadow Park Creek at the Bridge near where it enters the lake and about a mile upstream close to where the creek enters the LWW development. A third sample was collected from Wildwood Creek on the other side of the lake. That sample was also collected close to where the creek enters the LWW community. The MST markers utilized in this sampling are listed in Table 15.