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PFAS Surveillance within a Highly Militarized Island: A Case Study of Okinawa, Japan

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Description of sampling and extraction protocols

Bottles were submerged and rinsed twice with surface water prior to collection at each site. The four field blanks were prepared in the laboratory by transferring 250 mL of Optima water (Fischer Scientific) into clean HDPE bottles and then transported into the field during sampling events. At four randomly selected sites, field blanks were opened and transferred into another clean HDPE bottle. These field blanks were then stored and processed with the collected surface water samples. All solvents and reagents used for water extraction were Optima LC-MS grade and were purchased from Fischer Scientific. In short, all samples (surface water, field blanks, and quality control samples) were gravimetrically weighed, then spiked with 25 µL of an isotopically labeled PFAS internal standard mixture (made by dilution of 2 mL Wellington Laboratories MPFAC-24ES in 25 mL of Optima methanol). This mixture contained 19 isotopically labeled PFAS (dilution and spiked concentrations can be found in Supplemental Table ST3). Additionally, between 100-150 µL of glacial acetic acid was used to acidify each sample to a pH of 5. Samples were then extracted via solid phase extraction using Strata-XL-AW 100 µm Polymeric Weak Anion 500mg/6mL cartridges (Phenomenex, Torrance, CA). The cartridges were attached to a 24-port vacuum manifold and each cartridge was pre-conditioned with 4 mL of 0.3% ammonium hydroxide in methanol, 3 mL of methanol, and 3 mL of acetic acid/ammonium acetate buffer solution. Following cartridge conditioning, samples were loaded and extracted using a low vacuum – passing at 1-2 drops per second. The mass of the empty sampling bottles were recorded to normalize the final volume of each sample extracted. After loading the sample onto the cartridge, a washing step of 4 mL of acetic acid/ammonium acetate buffer was applied to each cartridge. Using full vacuum, cartridges were dried for 10 minutes to remove all water. Elution of PFAS was achieved with 2 mL of methanol and 6 mL of 0.3% ammonium hydroxide in methanol, followed by a drying step under full vacuum for 15 minutes. The eluent was then concentrated to 1 mL under ultra-high purity nitrogen gas. A 200 µL aliquot of each concentrated extract was transferred into polypropylene autosampler vials and stored at -20 °C until analysis.

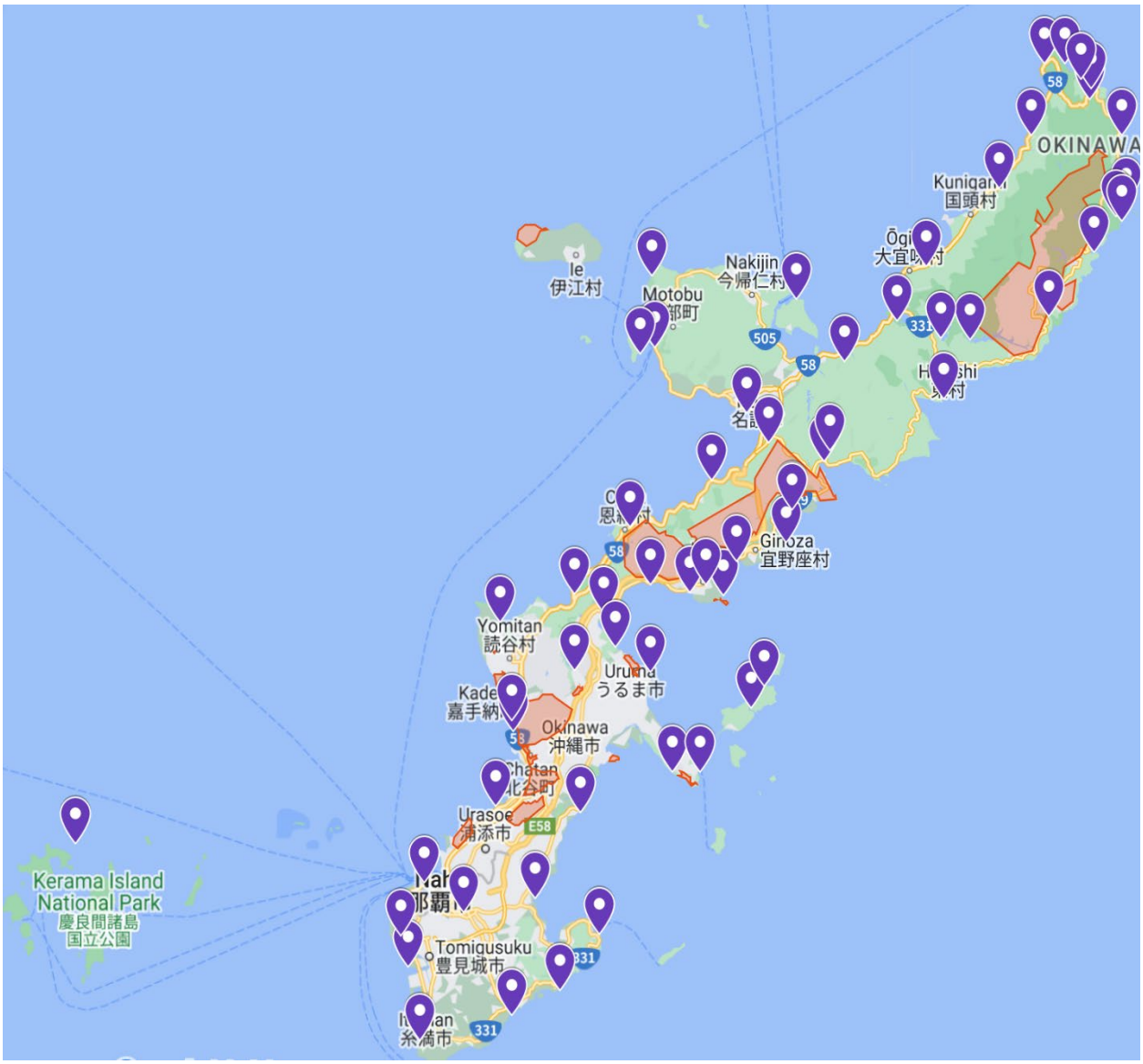
Description of predictive heat map generation:

The dataset containing 61 Okinawan PFAS sampling records with latitude and longitude coordinates was ingested into a geographic information system (GIS). ESRI's ArcGIS Pro (AGP) version 3.1.3 was used to ingest the spatially enabled data table and create a GIS PFAS point shapefile. The shapefile was then exported and projected from latitude and longitude into UTM Zone 52N, Japanese Geodetic Datum 2011. AGP was used to explore the data's spatial autocorrelation. The exploration revealed there was a slight correlation between the data value and its location and that the data was clustered (Moran's Index 0.076198, z-score 1.799058, p-value 0.072009). The point shapefile and AGP's Inverse Distance Weighting (IDW) interpolation program, using default parameters, produced a predictive PFAS surface. The IDW predictive surface best fit the observed values. Political boundaries for Japan were obtained from the Geospatial Information Authority of Japan website, The Global Map of Japan (https://www.gsi.go.jp/kankyochiri/gm_japan_e.html) (Geospatial Information Authority of Japan (GSI), 2016). The boundary was then spatially buffered 450m in order to encompass all but one of

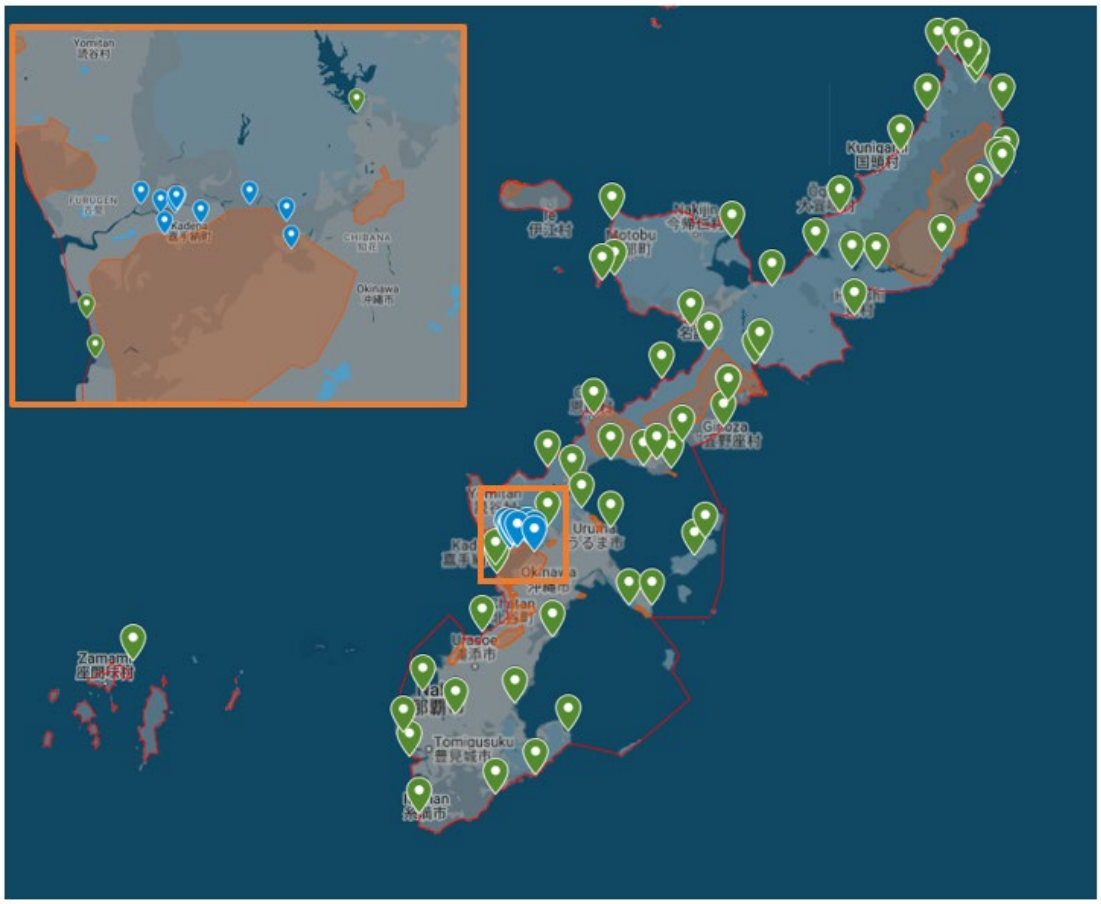
the distant offshore data points, Kerama Islands. A separate buffer was created for the point and added to the political boundary layer. The predictive surface was then clipped to the GIS shapefile. The predictive surface symbology used 10 classes and Natural Breaks (Jenks). The PFAS shapefile symbology also used Natural Breaks, but only 5 classes.

Description of other Okinawa PFAS studies

In 2020, Mitchell et al reported on PFAS data collected by the local government, which only monitored for the presence of three PFAS: perfluorooctanoic acid (PFOA), perfluorooctanesulfonic acid (PFOS), and perfluorohexanoic acid (PFHxA). This data reported on 7 sampling locations, congregated around Kadena Airforce Base, with anywhere between 4 and 50 sampling events at each site over a 12-month period. The goals of these reports were to monitor possible PFAS-contamination leaks from military bases into the surrounding water systems (Mitchell, 2020). Of note, elevated concentrations of perfluorooctane sulfonic acid (PFOS) and perfluorooctanoic acid (PFOA) were detected in Okinawa water systems near Kadena Airforce Base at concentrations up to 120 ng/L (individually and/or combined PFOS/PFOA) in drinking water, 2,000 ng/L in springs used for irrigation, and approximately 2-4 times the national average in blood levels (Mitchell, 2020). These values exceeded the drinking water advisory limit set by the Japanese government of 50 ng/L (combined PFOS/PFOA) set forth in 2020. Another study by Yukioka et al. in 2020 also examined the presence of PFAS on Okinawa through nontargeted suspect screening, in drinking water, river water and groundwater (n=18 total) and found concentrations of PFOS and perfluorohexane sulfonic acid (PFHxS) to be between 65-196 ng/L and 88-444 ng/L, respectively (Yukioka et al., 2020).



Supplemental Figure SF1: Map of Okinawa, Japan showing the 61 sites where surface water was collected. Map made using Google My Maps.



Supplemental Figure SF2: Map of Okinawa, Japan showing our 61 surface water collections (green pins) in relation to the eight river and groundwater sites collected by Yukioka et al. 2020 (blue pins). Red boundaries indicate military bases and/or training areas. Map made using Google My Maps.