USFWS Johnston Atoll National Wildlife Refuge

Crazy Ant Strike Team XX

Field Season Final Report

4 December 2020 – 1 June 2021



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Yellow Crazy Ant (YCA) Monitoring and Treatment

Yellow Crazy Ant (YCA) 50 m Monitoring Survey

The YCA 50 m Monitoring Survey is a monthly survey, which provides for the long-term monitoring of the presence or absence of YCA and other ant species (OAS) within the original YCA infestation area. Stations are positioned at the intersections of a 50 x 50 m grid laid across Johnston Island, with 314 stations total. Depending on the time of year, not all stations are surveyed due to nesting Sooty and Gray-back Terns and Black and Brown Noddies. Each station is baited with a pea-sized dollop of pureed Spam (1 can Spam: 1 cup water) placed on a 4 x 4 inch glazed ceramic tile. The survey begins before sunrise, at first light, and the surveyors return to each station 120 – 140 minutes following the initial baiting to count the number of YCA on each card, as well as the presence or absence of OAS. If a station has more than 40 individual YCA present on the tile, it is marked as ">40".

From November 2011 to January 2016, the YCA 50 m Monitoring Survey was conducted biweekly. However, in February 2016, the frequency of the survey changed to once every four weeks on the basis that the long term data would not be statistically compromised by reducing survey frequency, and other surveys had scheduling priority as the current low density demanded higher resolution surveys than were appropriate in the past.

CAST XX completed five YCA 50m Monitoring Surveys during their tour on Johnston (*Figure 1*). CAST XX did not detect YCA during any of the surveys. As of the final survey on 4 May 2021, YCA had not been detected in 48 consecutive 50m Monitoring Surveys. The last YCA detection via this survey was in January 2016.

Over the 50m surveys conducted by CAST XX, Other Ant Species (OAS) presence ranged between 73.1% - 92.7% of the stations. These percentages fall within the typical range found by other CAST crews, with the summer season usually having slightly higher OAS presence than the winter season. OAS presence has historically shown an inverse relationship to YCA presence. With the decline of YCA presence, the OAS presence has seemingly stabilized. However, there is a notable decline in OAS presence from January 2019 to June 2019. This may be explained by Hurricane Walaka that affected Johnston Atoll in September 2018. It should also be noted that the immediate effects of Hurricane Walaka were not recorded, with a gap in the 50m Survey data from July 2018 to January 2019.



Figure 1. YCA 50m Monitoring Survey results from June 2015 – April 2021. The shaded portion indicates CAST XX's tour on Johnston.



Figure 2. Average YCA 50m Monitoring Survey results from 2012 to 2021

Yellow Crazy Ant (YCA) Treatment Monitoring Survey

The Treatment Monitoring Survey provides data on the presence or absence of YCA and OAS within the YCA infestation area of Johnston Island. While initially a bi-weekly survey, survey priorities were shifted during CAST X to favor alternative survey techniques and the Treatment Monitoring Survey was conducted once every four weeks. Different from the 50m Monitoring Survey, the Treatment Monitoring stations target areas that are known to have YCA in order to determine more precisely how the YCA population is responding to bait application. During CAST XI the number of monitoring points was nearly doubled to increase the probability of detection.

There are currently 894 stations that are monitored over two consecutive days. Depending on the time of year, not all stations are surveyed due to nesting Sooty and Gray-back Terns and Black and Brown Noddies. Each station is baited with a pea-sized dollop of pureed Spam (1 can Spam: 1 cup water) placed on a 4 x 4 inch glazed ceramic tile. The survey begins before sunrise, at first light, and the surveyors return to each station 90 - 110 minutes following the initial baiting to count the number of YCA on each card, as well as the presence or absence of OAS. If a station has more than 40 individual YCA present on the tile, it is marked as ">40".

CAST XX completed five Treatment Monitoring Surveys. YCA were not detected at any of the stations during any of the surveys (*Figure 3*). OAS presence throughout CAST XX's tour has ranged from a low of 76.6% to a high of 95.9%. The average OAS presence over CAST XX was 84.6%, lower than most CAST crews and lower than CAST XIX's average of 93.5% (Dec 2019 – June 2020).

Similar to the 50m Monitoring Survey, OAS presence typically displays an inverse relationship to YCA presence. OAS presence levels have varied slightly over the years since YCA numbers declined in 2015. In May 2016, the percent of tiles that detected OAS temporarily declined immediately following the landscape level bait application of the infestation area. Additionally, the OAS presence dipped again temporarily from January to June 2019. As mentioned in the 50m Monitoring Survey results, this can most likely be attributed to the effects of Hurricane Walaka.



Figure 3. YCA Treatment Monitoring Survey results from Jan 2016 – May 2021. The shaded portion indicates CAST XX's tour on Johnston.

Hand Search Survey (previously Hand Search Tile Survey)

With the recent substantial drop in Yellow Crazy Ant (YCA) population across the island, survey methods such as the YCA 50m Survey and YCA Treatment Monitoring Survey lack the resolution to detect smaller densities of YCA. In order to detect these smaller populations, the Hand Search Survey was implemented during CAST XI's deployment. The purpose of the Hand Search Survey is to detect YCA within the Infestation Zone at as a high a resolution as possible. Each 50m grid square within the Infestation Zone is thoroughly searched by hand, for a minimum of one-person hour or until all possible nesting locations have been examined. Observers line up along the edge of a grid square, space themselves roughly equally across the diameter of the grid square, and advance in a single cardinal direction across the area where YCA were detected. In addition to hand searching, CAST XI originally implemented baited tiles within the squares they were searching. However, the tiles were not found to be effective as the YCA habitat was disturbed by the searching and YCA foraging behavior was altered. Therefore, CAST XI removed the baited tile element from the survey.

Observers thoroughly search the entire monitored area by hand, focusing on ideal YCA habitat such as the bases of trees, concrete edges, and under rocks and other debris. Thorough hand

searching techniques include but are not limited to uncovering vegetation stems, raking leaves or other debris, examining limbs and branches, and checking any cracks, cavities, or holes in the ground. Grid squares are searched in the morning hours, beginning shortly after sunrise and ending around 10:30 to avoid the heat of the day, when YCA are less active. If YCA are detected, nests are searched for and when found, flagged. The dominant and surrounding vegetation as well as a description of the nest location was recorded.

In order to declare YCA eradication from Johnston Atoll, it was determined that all 50m grid squares within the infestation zone needed to be hand searched four consecutive times without any YCA detections (via any survey method). In 2019, an Adaptive Hand Searching map was developed as most 50m grid squares had been searched and cleared four consecutive times. However, some 50m grid squares required an additional one to three additional hand searches. The Adaptive Hand Search map was updated in 2020 (Figure 4), removing the grid squares searched by CAST XIX and the remaining unsearched buffer squares- as these were used to add confidence, but not needed to declare eradication. The primary goal of CAST XX was to search and clear these adaptive hand searching grid squares.



Figure 4. The adaptive hand searching map, indicating the remaining number of times each 50m grid square still needed to be searched at the start of CAST XX.

On 7 April 2021 of the CAST XX's deployment, all 30 of the adaptive grid squares within the Infestation Zone were hand searched. All yellow squares in Figure 4 were cleared once, all orange squares were cleared twice and all red squares were cleared three times by CAST XX. No YCA were detected via hand searching or any other survey methods, therefore YCA eradication was declared.

During the first implementation of the Hand Search Survey, CAST XI detected YCA at 30 of the 367 surveyed grid squares surveyed (8%) through this survey. In the following deployment, CAST XII detected YCA at three of 156 hand searched grid squares (2%), CAST XIII detected YCA in two of the 247 grid squares hand searched (0.8%), and CAST XIV was the last crew to detect YCA via Hand searching, finding YCA at 4 of the 184 grid squares hand searched (2.2%). CAST XV proceeded to conduct Baited Area Monitoring (BAM) on these squares. These results are consistent with the idea that Hand Searching along with Treatment Monitoring is a fine scale method to detect YCA presence, followed by baiting methods to kill off the colony.

CAST XX finished searching the core plots within the infestation zone of the adaptive hand search round and moved to searching 50m grid squares around camp (outside the infestation zone). The purpose of searching these squares was to increase the confidence interval when declaring eradication, as YCA once established at camp in the beginning years of the project. However, they had not been observed there since 13 August 2013. CAST XX did not detect any YCA in the 50m grid squares searched around camp (*Figure 5*).



Figure 5. The blue colored squares represent the grid squares searched around the CAST camp from 23 April to 23 May

All-Island Ant Survey

CAST XX conducted the 20th All-Island Ant Survey on 8, 10 & 11 February 2021. This survey provides biannual data on the presence or absence of YCA and OAS over a 50 m grid across Johnston Island. CAST XX successfully surveyed 1,047 of the 1,048 stations of the 50m grid (one station not surveyed due to nesting terns).

For the All-Island Ant Survey a plastic 15 mL centrifuge tube is wrapped in aluminum foil in order to provide a dark environment that the ants prefer. The night before the survey the tubes are baited with a small cube of Spam (about 1 cm³). The survey begins before sunrise, at first light (~0700 HST). The surveyors open the tube and place it at each of the individual stations of the 50 m grid. Surveyors return to each station 120 – 140 minutes after the initial placement to close and collect the tubes. Immediately after the survey, vials were examined for ants and recorded as containing YCA, OAS, or no ants. Vials with no ants were checked by at least two crew members to ensure that no ants were missed. Samples were frozen immediately to euthanize the ants following each day's survey and remained frozen for at least 48 hours before the ants were counted and identified. After the counting and identification, most of the ants were disposed of, keeping only samples for training purposes and for confirmation of rarer species by the entomologists in Honolulu. The resulting data gives a picture of the current ant population on Johnston Island and provides insight into how other ant species are impacted by the presence or absence of the Yellow Crazy Ant, as well as by bait application.

Out of the 1047 stations surveyed, no stations contained YCA (0%), 700 stations (66.9%) contained OAS, and 347 stations (33.1%) did not contain ants. YCA were last collected during this survey in January 2016. Of the 1048 stations typically sampled, one station was not accessible due to the nesting sooty tern colony.

Table 1. Species collected during the 20th All Island Ant Survey, 8, 10 & 11 February 2021. Note:"Unknown" and "Monomorium spp." were specimens that were damaged beyond speciesrecognition.

Species	Number of Stations	Total Number of Ants
Anoplolepis gracilipes (YCA)	0	0
Cardiocondyla emeryi	1	3
Cardiocondyla minutior	1	1
Cardiocondyla obscurior	2	3
Monomorium destructor	45	5132

Monomorium floricola	3	69
Monomorium liliuokalanii	1	80
Monomorium pharaonis	30	1253
Monomorium spp.	2	2
Ochetellus glaber	399	39261
Paratrechina longicornis	19	2089
Solenopsis geminata	133	16393
Tapinoma melanocephalum	29	2745
Tapinoma spp.	1	4
Tetramorium bicarinatum	11	1003
Tetramorium simillimum	58	803
Tetramorium spp.	1	1
Unknown	6	6
Stations with no ants collected	351	-
Grand Total		68,835

The number of ants collected by CAST XX was lower than normal, with 68,835 individuals counted and identified, not following previous season's trends. During the first All-Island Ant Survey collection after Hurricane Walaka (AIAS Survey 17), a total of 59,458 ants were identified and counted from the 498 vials containing ants. The number of ants have continued to increase since the hurricane in September 2018, with a total of 70,123 ants identified from the 631 vials containing ants on All-Island Ant Survey 18 and a total of 100,625 ants identified from the 831 vials containing tubes on All-Island Ant Survey 19. The temporary decrease in the number ants collected on AIAS Survey 17 is most likely explained by the impacts of Hurricane Walaka, as much of the habitat was damaged (i.e. uprooted trees). The reason for the most recent dip in the number of ants collected by CAST XX is unknown, but could be related to the weather as some of the collection days had heavy clouds.

The two most abundant species were *Ochetellus glaber* and *Solenopsis geminata*. In regards to the number of individual ants counted, the most abundant species was *O. glaber* (ants= 39,261; 57.0% / vials=399; 57.0% of all sampled vials) and the second most abundant was *S. geminata* (ants= 16,393; 23.8% of all ants counted / vials=133; 19.0% of all sampled vials). These two species have been the most abundant and widespread species on Johnston over at least the past 10 surveys (6 years).

Additionally, *Monomorium sechellense* was not collected during the 20th All-Island Ant Survey. This species was observed for the first time at one point on Survey 17 and seen again at one point on the 18th and 19th surveys.

All of the other ant species identified during this survey have been reported in previous surveys. Fourteen different species of ants were identified (Table 1). Two different species of ants were observed at 41 of the 831 stations containing OAS and three different species of ants were observed at one of the stations.



Figure 6. The count of stations at which eight influential ant species were identified over the past four All-Island Ant Surveys (Feb 2019, July 2019, Feb 2020, Feb 2021).

Of the eight influential ant species found on Johnston Island, *O. glaber* has increased the most in the number of stations the species was detected at since the last All-Island Ant Survey. The number of *S. geminata* decreased significantly since the last All-Island Ant Survey, while many of the other species remained relatively consistent. Of all the species identified, *M. pharaonis* and *M. destructor* were the most difficult to differentiate, especially in tubes containing few specimens. Samples from each species were sent back to Honolulu to confirm identification.



Figure 7. Distribution of ant species collected on Johnston Island on 8, 10 & 11 February 2021. Note: when there were two or more species of ants at the station, only the primary ant species at the station was included in this figure (i.e. Cardiocondyla minutior not shown as it was not the most abundant ant found at that point).

OAS Comeback Survey

Due to survey prioritization on CAST XX, the OAS Comeback Survey was not conducted. This survey monitors the response of other ant species (OAS) to the reduction of YCA within the infestation area. The survey prioritization was determined by management and biologists in Honolulu prior to the CAST XX deployment to ensure that all surveys needed to declare YCA eradication were able to be completed during this crew.

Non-Infestation Survey

Due to survey prioritization on CAST XX, the Non-Infestation Survey was not conducted. This survey monitors the response of other ant species (OAS) to the reduction of YCA within the infestation area.

Bait Application

No bait application occurred on CAST XX in response to YCA detections, as YCA were not detected using any of the various survey methods – Treatment Monitoring Survey, 50m Survey, or the Hand Search Survey.

Historically, CAST crews used bait containing the insecticide Safari (dinotefuran) delivered in water absorbing polyacrylamide ("hydrogel") crystals matrix to treat YCA. This was found to be the most effective form of bait application, followed by a cat food matrix.

All recipes using the insecticide Safari were mixed to have a final active ingredient concentration of 0.05%. The active ingredient concentration in the cat food recipe is based upon the final mass of bait. The concentration in the hydrogel recipe is based upon the volume of water used. The current recipes, per kilogram of bait mixed, are as follows:

Hydrogel: 780mL H20, 195g sucrose, 7.6g Large crystal hydrogel, 7.g6 Small crystal hydrogel, 2.0g Safari

Cat food: 250 g Friskies cat food, 250 mL Karo corn syrup, 500 mL water, 2.5 g Safari, xanthan gum

Seabird and Shorebird Monitoring

Mean Incubation Counts (MIC)

On Johnston Island, there are three groups of seabirds monitored by CAST crews. These groups, the Tropicbirds (*Phaethon*), the Boobies (*Sula*), and the Frigatebirds (*Fregata*), are monitored using routine nest counts. The timing of each of these mean incubation counts (MIC) is based on the mean incubation period of each species as described in *The Birds of North America* (Schreiber et al., 1996, Schreiber et al., 2009, Gauger et al., 2002). Tropicbirds are monitored every 43 days, boobies are monitored every 46 days and frigatebirds are monitored every 55 days. Additionally, Johnston Island is divided into 22 seabird sectors (*Figure 8*) to standardize survey areas and break the surveys into manageable regions.

CAST XX surveyed seabird sectors using the following method; crew members dispersed at equidistant lengths in parallel to each other at one of the boundaries of a sector, then, maintaining the same pace, they each walk a straight line transect to the opposite end of the sector. The distance between crew members is determined by the density of vegetation of the transect. Each crewmember is responsible for counting the active nests directly in the transect they walk in order to avoid double counts of birds. The crew repeats this method going from one boundary of the sector to the opposite boundary until the entire area has been covered. From such surveys, crews can determine how many birds are actively reproducing on the island and which months have the highest levels of reproduction.



Figure 8. The 22 seabird survey sectors on Johnston Island.

Brown Booby, Red-footed Booby, and Masked Booby Mean Incubation Counts

Brown Boobies (Sula leucogaster, BRBO), Red-footed Boobies (Sula sula, RFBO), and Masked Boobies (Sula dactylatra, MABO) exhibit asynchronous and non-seasonal breeding behavior on Johnston Atoll. Active booby nests are counted once every mean incubation period (46 days for Red-footed Boobies and 43 for Brown Boobies and Masked Boobies) in an attempt to monitor the total breeding population on the island. Due to the historically low numbers of Brown Boobies and Masked Boobies that nest on Johnston, all three Booby species are surveyed within the same mean incubation period as the Red-footed Boobies: 46 days. All 22 seabird sectors were surveyed for nests via wide, straight-line transects and recorded as having an egg or a chick. In all surveyed sectors, chicks were classified along a growth-stage spectrum with stage 1 being a small, completely naked chick and stage 9 being a fully feathered, nearly fledged chick (Figure 9). Nests were determined as unconfirmed when observers could see nesting material and a brooding adult, but the presence of an egg or chick could not be verified. Most of the unconfirmed nests were too high off the ground to see inside the nest or to tip the adult. Some unconfirmed nests were inaccessible due to the nesting Sooty Tern colony, typically located in the northeast corner of Johnston Island.

(n)	Naked
(2)	Down appearing on back
(3)	Down beginning to cover whole body
(4)	All downy approx. 1/2 adult size
(5)	All downy approx. adult size
(6A)	Primaries and rectrices visible; no scapulars
(6B)	Scapulars visible; down still present on back
(6C)	Scapulars unite in midline; down still on wings
σ	Wings and back almost clear of down; thick down remaining on head, neck, flanks, belly and
	breast variable
(8)	Fully feathered but not flying; thin or wispy down may persist on radio-ulna, nock and flanks
(9)	Fully feathered

Figure 9. Staging guide to all of the booby species during each MIC since 2011

CAST XX conducted three Mean Incubation Counts of the Booby populations on Johnston Island in January 2021, March 2021 and April 2021. The surveys usually took between three to four days to complete and were conducted approximately 46 days apart. As in all past years of CAST surveys, the Red-footed Booby was the most common nesting species on Johnston Island. CAST XX observed a record high of 5,350 active RFBO nests in January 2021, compared to the previous high of 4,394 observed active nests in April 2019 (*Figure 10*). The March 2021 recorded the second highest RFBO nesting numbers (n=5,259), but also had fewer unknown nests as the observer could more easily identify chicks than eggs in higher up nests.

The seasonal fluctuations in the number of active RFBO nests during CAST XX's tour were not quite consistent with historical data from previous CASTs. Typically, peak nesting for RFBOs will occur March or April each year, not January. However, March did still have high nesting numbers. There was a 33.0% increase in the maximum number of active RFBO nests between Spring 2020 and Spring 2021; the largest increase seen in throughout the CAST project. Additionally, there was a 17.9% increase from the previous record high in Spring 2019 (n=4,394). It is possible that the increase from 2020 to 2021 was uncharacteristically large due to the El Nino Southern Oscillation (ENSO) cycle, which is often associated with the skipping of a breeding season in Boobies and other long-lived birds (Nelson 1969, Erikstad et al. 1998, Cubaynes et al. 2010).



Figure 10. Total number of active Red-footed Booby nests on Johnston Island, 2015-2021. Note the gap in data collected from July 2018 to January 2019 due to the Hurricane Walaka evacuation, and the gap from April 2020 to January 2021 due to Covid-19.



Figure 11. Active Red-footed Booby nests counted in each seabird sector during the peak nesting MIC for RFBO (January 2021) conducted by CAST XX, totaling 5,350 active nests. The numbers within the parentheses indicate the number of active Brown Booby nests counted in each seabird sector during the peak nesting MIC for BRBO (April 2021) conducted by CAST XX, totaling 21 active nests.

Brown Booby active nests are much less common than RFBO active nests on Johnston Island, with a previous high of 13 active nests most recently observed in February 2019. CAST XX observed a record high of 21 BRBO nests on Johnston Island MIC in April 2021. Additionally, twelve BRBO nests were observed during the January 2021 MIC, followed by 19 BRBO nests observed during the March 2021 MIC.

CAST XX observed one active Masked Booby nest on Johnston Island during their deployment in Seabird Sector 14. However, after the adult incubated the eggs for more than 80 days, this active nest was considered failed and the adult eventually left the nest. Anecdotally, adult MABOs were regularly observed flying over the eastern side of the island or perched along the seawall. Less frequently, juvenile MABOs were also observed, indicating they were actively breeding on the outer islands.

Red-tailed and White-tailed Tropicbird Mean Incubation Count

Red-tailed Tropicbirds (Phaethon rubricauda, RTTR) and White-tailed Tropicbirds (Phaethon lepturus, WTTR) are surveyed for breeding activity on Johnston Atoll in nine sectors (2, 3, 4, 5, 9, 13, 16, 17, and 20) every 43 days (Figure 12) to coincide with the mean incubation period for Tropicbirds. The three outer island sectors are not surveyed. Sectors are surveyed for nests via loose, straight-line transects and recorded as having an egg or chick. Chicks are classified along a ten-category growth stage spectrum with 1 as a small chick completely covered in down and 10 being a fully feathered, nearly fledged chick.

CAST XX only completed one Tropicbird MIC due to survey prioritization. This survey encompassed all 22 Seabird Sectors in March 2021, during peak nesting season. All Tropicbird MIC data is detailed in the *All-Island Red-tailed and White-tailed Tropicbird Survey* below.

All-Island Red-tailed and White-tailed Tropicbird Survey

Once a year, since February 2012, CAST crews have completed an All-Island Red-tailed and White-tailed Tropicbird Survey including all 22 of the seabird sectors on Johnston Island (Figure 12). This survey is conducted as close to the annual peak in nest activity as possible so a minimum population size on Johnston Island can be extrapolated. Based on the peak breeding season from previous years, as well as monitoring both the routine Tropicbird MIC data and activity at the Reproductive Plots, it was determined that this MIC would fall as close to peak nest activity as possible. Other than the additional Seabird Sectors monitored, the survey protocols and staging of chicks is the same as the regularly conducted Tropicbird MIC.

The All-Island Tropicbird MIC was conducted between 22-27 & 29-31 March 2021; all 22 seabird sectors across Johnston Island were surveyed. On Johnston Island **a total of 12,963 active RTTR nests** were counted (*Figure 12*). Of the total active nests, 5,700 (44.0%) contained eggs and 7,256 (56.0%) nests contained in various stages of development.

Seven nests were recorded as unconfirmed as observers were unable to reach the nest with a tipping stick to confirm chick or egg presence. The unconfirmed nests were due to their location within extremely dense vegetation surrounded by an overwhelming presence of wasps. The density of vegetation on Johnston Island (primarily thickets of *Leucaena leucocephala* and *Pluchea indica*) is notably increasing each year. Because of the vegetation growth and increasing number of wasp nests, the margin of error has almost certainly increased as well, erring on the side of missing nests.



Figure 12. Active Red-tailed Tropicbird nests counted in each seabird sector from 22-27 & 29-31 March 2021, during peak nesting season. Across all sectors a total of 12,956 active nests were recorded.

The original All-Island Tropicbird Survey conducted in February 2012 recorded 5,212 active nests, marking Johnston Island as the largest known RTTR breeding colony in the world. The trend shows an increasing number of active nests (*Figure 13*). From the All-Island MIC in 2020 to 2021, the number of active RTTR nests increased by 19.0% (from 10,884 to 12,956 active nests), demonstrating that Johnston Island remains a crucial and robust breeding site for Red-tailed Tropicbirds.



Figure 13. Total number of egg nests and total active nests (combined egg and chick nests) recorded during each All-Island Tropicbird MIC throughout the CAST project.

In addition to the Red-tailed Tropicbirds, 19 White-tailed Tropicbird active nests were counted. Of these nests, 14 contained eggs and five contained chicks. White-tail nests were found in Seabird Sectors 8, 9, 10, 11, 18, 20, 21 & 22.

Great Frigatebird Mean Incubation Count (MIC)

As Great Frigatebirds (Fregata minor) exhibit asynchronous and aseasonal breeding behavior on Johnston Atoll, the number of active Great Frigatebird (GRFR) nests are counted and staged once every mean incubation period (55 days), with the goal of monitoring the total breeding population.

Frigatebird Chick Stages
A1: Naked
A2: All Downy
B: Scapulars erupted, <3cm
C: Scapulars >3cm, primaries and secondaries not erupted
D: Primaries and secondaries erupted; rectrices erupted, <3cm
E: Rectrices >3cm; down just clearing head and wing
F: Down half gone, clear from head and wing
G: Down on throat and Breast only
H: Traces of down only
I: Fully feathered; no down

Figure 14. Staging guide to the Great Frigatebird nests surveyed during each MIC since 2011.

CAST XX successfully surveyed all 22 seabird sectors of the island for active Great Frigatebird nests on three occasions, in December 2020, February 2021 and April 2021. This survey was always completed in less than one workday. Notably, a significant increase and record high number of active nests (n=228) was documented in February 2021 (*Figure 15*). Following previous nesting behavior, all of the Great Frigatebirds nests observed by CAST XX on Johnston Island were located on the eastern section of the island - primarily in Seabird Sectors 14 and 15, with the occasional nest in Seabird Sectors 1 and 16 (*Figure 16*). Additionally, primarily nests with eggs were observed in December and nests with chicks were then primarily observed in February and April.

CAST XX also completed a Great Frigatebird MIC on the outer islands in December 2020 and made anecdotal observation in May 2021 - results are summarized in the "Outer Island" section of the report below. It should also be noted that between August 2013 and March 2016, no active GRFR nesting was observed on Johnston Island. However, during this period, Johnston Island typically had large numbers of adult and juvenile frigatebirds present, as breeding took place on the outer islands.



Figure 15: This graph shows all active Great Frigatebird nests surveyed during each MIC since 2011. Note: There is a six-month gap in observation between May 2020 and December 2020, and a five-month gap in observations between July 2019 and January 2020.



Figure 16. Active Great Frigatebird nests counted in each seabird sector during the peak nesting MIC by CAST XX (February 2021). Across all sectors a total of 228 active nests were recorded and notes were made about the locations of adults that exhibited roosting or breeding behaviors.

Tropicbird Reproductive Plots

During their deployment, the members of CAST XX conducted weekly Tropicbird Reproductive Plot Surveys to monitor nesting success of the island's Red-tailed Tropicbird and White-tailed Tropicbird. These surveys were conducted both inside and outside of the YCA infestation area in an attempt to observe the effect of YCA presence on nesting success of Red-tailed and Whitetailed Tropicbirds on the island. The Reproductive Plot Surveys began in April 2014.

Reproductive Plot Surveys are conducted in ten 2,500 m² plots around the island. Nine of these plots are inside existing 50m grid squares and the tenth is made up of sections of four 50m grid quadrants (Figure 17). Each week CAST members visited their assigned plots and catalogued nest locations and the presence of an egg or a chick. Similar to the Tropicbird MIC surveys,

Reproductive Plot Surveys record the developmental stage of chicks, but also monitor overall nest success by taking into account chick mortality and fledgling success for each nest.



Figure 17. Location of the Tropicbird Reproductive Plots on Johnston Island

Due to malfunctions with the Microsoft Access databases, the RTTR Reproductive Plots could not be analyzed for each crew. Initial notes have been temporarily made until the database issues can be resolved with the assistance of a database manager

From summarized notes of each week's observations on CAST XX, a total of 692 nests were laid during the 2021 hatch year (i.e., 19 November 2020 to 17 May 2021). Of these, nests, 443 of these nests reached a terminus (fledging or failure); 38.4% (n=170) of nests have successfully fledged, while 61.6% (n=273) of nests have failed (i.e. "egg gone" "dead chick" or "chick gone"). Additionally, 255 nests were left as incomplete lifecycle observations. Many of the failed nests seems to be due to the predators on Johnston Island. The Hawaiian Short-eared Owl likely killed many early-staged chicks. In addition, 5S plot had many missing chicks during the weeks that the Peregrine Falcon and Northern Harrier were observed in that region of the island. This data will be further analyzed once the issues with Access are resolved.

Shorebird/Non-Pelagic Bird Survey

An all-island Shorebird and Non-Pelagic Bird Survey is conducted biweekly. Four transects (Figure 18) are walked east-to-west by four people in unison, and in radio contact to avoid missing or double-counting birds. All shorebirds and non-pelagic birds are recorded for each transect. This survey takes approximately 90 minutes to complete the ~2-mile-long routes. Surveys began between 7:00 and 8:00 HST.

Non-pelagic birds are defined as birds that do not depend on marine resources for survival. Surveys were performed during the USFWS's tenure on the active US Air Force Johnston Atoll installation from 1991 to 2001. They ceased upon base closure and resumed in 2010 with the establishment of the CAST program. Due to growth of forests on the island following abandonment, survey routes were changed in 2011 to improve visibility for counts (Figure 18). Regular radio checks are necessary to maintain pacing of surveyors and avoid re-counting of birds. Survey transects are approximately 2 miles (3.1 km) long, typically taking 75 minutes to complete. Seven common species are normally recorded: Bristle-thighed Curlew (Numenius tahitiensis), Pacific Golden Plover (Pluvialis fulva), Wandering Tattler (Heteroscelus incanus), Sanderling (Calidris alba), Ruddy Turnstone (Arenaria interpres), Cattle Egret (Bubulcus ibis), and Hawaiian Short-eared Owl (Asio flammeus sandwichensis). Other species of interest are recorded if observed.



Figure 18. Shorebird/Non-Pelagic Bird Survey transects on Johnston Island surveyed since 2011

CAST XX performed 10 surveys from 8 January 2021 to 14 May 2021. For 6 out of 10 surveys, Pacific Golden Plovers were the most common shorebird. Numbers of the three most common shorebirds, the Ruddy Turnstone, Pacific Golden Plover and Bristle-thighed Curlew, increased over the winter season (*Figure 19*). A decline in these species was observed in late April and May, which is consistent with the seasonal observed by previous CASTs. The Hawaiian Shorteared Owl, Wandering Tattler and Sanderling were least common species observed, with maximum sightings of two, seven and twelve birds, respectively.



Figure 19. Total number counted per survey of each common shorebird/non-pelagic species during CAST XX on Johnston Island. Abbreviations: Bristle-thighed Curlew (BTCU), Pacific Golden Plover (PGPL), Ruddy Turnstone (RUTU), Wandering Tattler (WATA), Sanderling (SAND), Hawaiian Short-eared Owl (SEOW).

Overall, the average number of Cattle Egrets sighted during each Shorebird and Non-Pelagic Bird Survey has been generally increasing throughout the CAST project (*Figure 20*). A slight decrease in the average was observed in 2018 and again in 2021. CAST XX recorded the lowest average since 2014. Anecdotally, the CAST XX crew observed more Cattle Egret carcasses than normal, with some that appeared to be predated upon. The decrease may a result of the vagrant birds of prey (Northern Harrier and Peregrine Falcon) observed in the first few months of the CAST XX season. Cattle Egret eggs and chicks were still seen on Johnston Island. Additionally, adult Cattle Egrets have continued to predate upon tern and tropicbird eggs.



Figure 20. Average number of Cattle Egrets counted during each Shorebird/Non-Pelagic Bird Survey from 2010-2021.

Birding Highlights and Anecdotal Observations

Other Seabird and Regularly Observed Non-Pelagic Species

In addition to the seabirds regularly monitored through mean incubation counts (tropicbirds, boobies, and frigatebirds), there are several other seabird species that nest on Johnston Island. The other seabird species of note include Sooty Terns, Gray-backed Terns, White Terns, Black Noddies, Brown Noddies, and Wedge-tailed Shearwaters. Additionally, the Hawaiian Short-eared Owl and Cattle Egret, both monitored in the Shorebird/Non-pelagic Bird Survey, have been observed nesting on Johnston Island.

Sooty Terns (Sterna fuscata oahuensis, SOTE)

CAST XX mapped the Sooty Tern Colony on Johnston Island on a weekly basis when the SOTEs were present. Both the colonies on the outer islands and Johnston Island can have various breeding times throughout the year. CAST XX first observed the colony on Johnston Island on 24 April 2021. Thousands of Sooty Terns were seen on the ground, courting, finding nesting locations and copulating. By mid-May the colony had established itself with thousands of breeding pairs on the northeastern corner of the island. Throughout the month of May, the colony spread to Seabird Sector 14 at the east end of the runway. From mid-to-late May, approximately half of the colony were made during the month of June due to the demobilization of the Johnston camp, but it suspected that the colony would still grow slightly in size.

Additionally, the CAST XX crew participated in a Sooty Tern feather collection study for a PhD student in France, using seabirds as indicators for oceanic mercury levels. The goal was to collect approximately five breast feathers from 30 adults and 30 chicks. CAST XX collected 30 adult feathers ensuring samples were taken from different individuals. Morphometric data was also recorded for the study. The 30 chicks were unable to be sampled, as they were still too young and downy or feathers still in pin.



Figure 21. The Sooty Tern extent of colony on Johnston Island observed by CAST XX

Gray-backed Terns (Sterna lunata)

On 24 April 2021, the Gray-backed Terns were also first observed nesting in smaller groups scattered around the island. Throughout the month of May, they were observed on the Tide Station Pier with a few individuals incubating eggs. Additionally, they were seen sporadically around the edges of the Sooty Tern colony displaying courtship behaviors, but were not seen with eggs prior to the CAST XX departure.

White Terns (Gygis alba)

Although a formal survey has never taken place, it seems as though the number of White Terns on Johnston Island has decreased over the CAST project. This is most likely due to predation as the adult carcasses are frequently found with typical signs of Hawaiian Short-eared Owl predation. Only four White Terns were confirmed to be incubating eggs on CAST XX. However, none of these eggs hatched before the CAST demobilization.

Black Noddies (Anous minutus melanogenys)

In April, the Black Noddies began gathering nesting material and displaying courtship behaviors. In May, nests had been built in trees along the northern shore between the wharf and Tide Station Peninsula, at the Internet Café, and at the eastern edge of Seabird Sector 13.

Brown Noddies (Anous stolidus pileatus)

In late May, the Brown Noddies began to lay eggs on Johnston Island. Two small colonies, each with approximately 20 nests, were formed along the seawall of Seabird Sector 3. In the past, Brown Noddies have also nested on the Tide Station Peninsula and along the eastern edge of the runways.

Wedge-tailed Shearwaters (Puffinus pacificus)

In April 2021, the Wedge-tailed Shearwaters returned to Johnston Island. CAST XX first heard their distinct calls at night and were regularly seeing them within a few days at dusk and at night. Courtship behavior was just starting to be observed in May, along with freshly dug burrows. In addition to the burrows mapped in 2017, CAST XX also made anecdotal observations of where the burrows were newly observed (*Figure 22*). It also seemed that the freshly dug shearwater burrows could possibly be denser than those mapped in 2017 and they may have spread out from those locations slightly.



Figure 22. Wedge-tailed Shearwater burrows from 2017 with anecdotal notes from 2021.

Hawaiian Short-eared Owls "Pueo" (Asio flammeus sandwichensis)

No Pueo nests were observed during the CAST XX deployment. Previous nests were typically detected on the ground in shallow nest bowls within pluchea shrubs. Detections usually occurred during tropicbird MICs or when adults were seen flushing from a location. Although no nests were detected, at least eight adults were seen at one time on Johnston Island. As this species is quite elusive, they are not frequently observed during the Shorebird and Non-Pelagic Bird Survey. The exact number of individuals is difficult to estimate, but most likely at least 15 and up to 40 individuals.

Additionally, Pueo were observed predating on many of the seabird species on Johnston Island. CAST XX observed instances where they were flushed while feeding and found many carcasses where the cause of death was characteristic of the way the Pueo feed (the spine and head ripped from the body). It appeared their prey primarily consisted of young Red-tailed Tropicbird chicks, Sooty Terns, Gray-backed Terns and White Terns. On a few occasions, both Brown Noddy and Black Noddy carcasses were also observed.

Observed Vagrants

Laughing Gull (Leacophaeus atricilla)

This gull was spotted on the southwest end of the island, near the Shark Chute and the old shooting range. It was only seen during the CAST XX drop-off and throughout the month of December 2020. Other vagrant laughing gulls have been spotted on Johnston Island in the past.



Figure 23. Laughing Gull (Photo credit: Ryan Rash).

Peregrine Falcon (*Falco peregrinus*)

This species was also spotted on the southwest end of the island, near the Shark Chute and old shooting range. The peregrine was observed predating upon early-staged tropicbird chicks. It was first observed in December 2020 and was last seen in early February 2021. A Peregrine Falcon was also seen in on Johnston Island from November 2017 through January 2018.



Figure 24. Peregrine Falcon (Photo Credit: Ivan Vicente)

Northern Harrier (*Circus hudsonius*)

The harrier was observed for the first time during the Shorebird and Non-Pelagic Bird Survey at the end of South Shore Transect on 20 January 2021. It was subsequently sighted over the next few weeks on at least four other occasions, all in southwest region of Johnston Island. Four of the CAST crew members were able to observe and confirm the species. This observation by CAST XX appears to be the first recorded instance of the Northern Harrier by a CAST crew. Although it was not directly observed doing so, it is suspected to have been predating on the Red-tailed Tropicbird chicks. During the time of the Northern Harrier and Peregrine Falcon observations on Johnston Island, many dead Red-tailed Tropicbird chicks were found in the region of the island where they were seen. Additionally, the dead carcasses found appeared to have different predation marks than what is typically seen from the Hawaiian Short-eared Owl.



Figure 25. Northern Harrier spotted on the multiple occasions at the southwest end of the island. (Photo credit: Ryan Rash)

Marine Monitoring

Sea Turtle Survey

The objective of the Sea Turtle survey is to monitor the status of Green Sea Turtles and to detect any major changes in their presence over time. Each survey was performed for one hour, every other week and required simultaneous observations by surveyors atop the east and west sides of Johnston Island's JACADS peninsula. Each observer counted the number of turtles from their respective sides of the peninsula being mindful not to count the same individual twice.

Ten Sea Turtle Surveys were conducted during CAST XX's deployment with a mean of 9.0±1.8SD turtles observed per survey, compared to a mean of 10.5±5.2SD turtles observed per survey

since January 2012 (n = 206). Like past crews, CAST XX noticed a difference in the number of sea turtles seen from the west side of the JACADS peninsula compared to the east side of the peninsula (*Figure 26*). CAST XX observers saw a mean of 3.30 turtles on the west side compared to a mean of 5.50 turtles on the east side. Since January 2012, observers have seen a mean of 3.40 turtles on the west side and 7.02 turtles on the east side. Unlike the two previous winter crews (CAST XVII and CAST XIX), CAST XX saw a fairly consistent number of turtles throughout the season rather than more turtles in late spring compared to the winter and early spring. Additionally, surveys were conducted on a fairly even mix of sunny and cloudy days with varying wind speeds and tidal conditions. These conditions did not appear to have any major effect on turtle sightings.



Figure 26. Green Sea Turtles observed from the JACADS Peninsula on Johnston Island from January 2012 to May 2021. Note: there is a gap in the data from September to December 2018 due to the Hurricane Walaka Evacuation and from May 2020 to January 2021 due to Covid-19.

NOAA Marine Debris Shoreline Survey

Marine debris has become one of the most widespread pollution problems in the world's oceans and waterways today. The NOAA Marine Debris Program (MDP) serves as a centralized marine debris resource within NOAA, coordinating and supporting activities within NOAA and with other federal agencies.

Standing-stock studies provide information on the amount and types of debris on the shoreline. Debris within discrete transects at the shoreline site is tallied during standing stock surveys. This is a quick assessment of the total load of debris and is used to determine the density (# of items per unit area) of debris present. Debris density reflects the long-term balance between debris inputs and removal and is important to understanding the overall impact of debris. Prior to deployment, NOAA trains outgoing CASTs for the monitoring of marine debris on Johnston Island. Because of the possible contaminants found on Johnston Island and the limited sandy shoreline, USFWS opted for a Standing-Stock Study to be conducted once a month on the South beach of Johnston Island. This data is reported to NOAA to be accumulated in their longstanding database.

Due to survey prioritization on CAST XX, the NOAA Marine Debris Shoreline Survey was not conducted.

Marine Survey

The objective of the Marine Survey on Johnston Atoll is to establish and maintain a relative index of fish species and numbers on the reef habitats surrounding Johnston Atoll. Two transects are currently monitored: East Camp and West Camp. The East and West Camp transects, running perpendicular to the north shore, are both swum during one survey. These transects have been monitored since 2012. The South Shore Transect, which was monitored from February 2014 to August 2016, runs parallel to shore for approximately 300 meters. The Camp transects are surveyed every week, unless conditions do not allow for the survey to be conducted or ant or bird surveys take precedence. On both transects, individual fish within one meter of the transect line are counted and identified to species.

The habitat of Camp Reef is characterized by coral outcroppings and ridges separated by sandybottomed canyons. As with most shallow locales around Johnston, the dominant coral species is Acropora cytherea, and other corals such as Montipora spp. and Pocillopora spp. are common. This location is one of the more sheltered areas around the island with respect to wave action and circulation. Both transects begin in the coral rubble near the seawall and extend north over the reef for their duration, however, the East transect hugs the eastern reef edge, where the reef opens into a little basin that sits between the main Camp Reef and a smaller patch reef still farther to the east. Sandy bottoms make up a greater portion of the West transect.

During CAST XX's deployment, 17 marine surveys were conducted at both Camp Reef locations. A total of 6,547 individuals were counted across 17 families and 51 distinct species. The two Camp Reef transects are comparable in both abundance and diversity (Table 2).

Table 2. Summary of total sums and average fish counts observed during Marine Surveys duringCAST XX's tour on Johnston Island.

Transect	Total Count	Average Individuals	Species Observed	Families observed
West	3357	198	45	15
East	3190	188	49	16
Camp Reef			51	17

Like previous CAST crews, the population composition is similar across both transects and differences in species composition are minimal (*Figures 27 & 28*). The most common species belong to the Parrotfish, Goatfish, Butterflyfish, Wrasse and Surgeonfish families on both transects (refer to Appendix A for scientific names and common names).



Figures 27 & 28: Relative composition of the six primary fish families on each transect from January 2021 to May 2021. All other families comprise of less than 3% of species observations.

Vegetation Management

Klu Survey

Klu (*Acacia farnesiana*) is a thorny invasive weed that fails to provide suitable nesting habitat for seabirds. CAST XX continued Klu monitoring and management in the five sites regularly searched by previous CASTs: south of the Internet Café, north of the Tennis Courts, the far Western Shore, the large Gravel Pile on the southern edge of Seabird Sector 17, and Corner Beach in Sector 4 (*Figure 29*). Sites were visited once a month, spending a minimum of one person-hour in each location searching for plants and seedlings.

CAST XX conducted six surveys with a total of 30 person-hours searching for Klu. A singular specimen was detected at the Internet Café site on 5 January 2021, during the first Klu survey of CAST XX. The plant was approximately 1.5m in height with no seeds present. The entire plant was able to be pulled up and then burned, following the SOP. No other Klu plants were detected at the other four sights. This was the first Klu detection since CAST XVI, on 25 July 2018. CAST XVI detected their one ~1.25m Klu stand during the RTTR MIC in the approximate location that the CAST XX Klu stand was also detected at.



Figure 29: Five locations surveyed for the removal of Klu by CAST XX, showing the one Klu detection at the Internet site.

Garlon Application

In 2017, a permit was acquired for herbicide application in order to clear any vegetation that reached nuisance levels on Johnston Atoll. In several locations across Johnston Island, especially the designated landing area surrounding the runway, common Ironwood (Casuarina equisetifolia), Indian Fleabane (Pluchea indica), Kou (Cordia subcordata), Sea Grape (Coccoloba uvifera) and Haole Koa (Leucaena leucocephala) have reached nuisance levels. During the CAST XII- CAST XIII changeover, CAST XIV – CAST XVI changeover and CAST XVI redeployment, a team of sawyers and swampers felled many ironwoods from both sides of the runway. CAST XIII, CAST XIV, and CAST XV had also cleared problematic vegetation. CAST crews continued the initiative by cutting and treating all main branches with Garlon 4-Ultra (triclopyr), while adhering to all quidelines posted in the Garlon Application SOP. In addition to the runway, CAST crews treat woody vegetation threatening the structural integrity of camp bunkers and the vegetation surrounding the "Internet Café" area. On Johnston, the survey routes—which are not treated with Garlon—are managed per survey on a continual basis, as to not impact the integrity of data collected. These include the multiple ant survey routes that are impacted by the evergrowing flora of the island. This process of trail cutting is crucial to the completion of these surveys in a timely and effective manner.

The Garlon 4 Ultra was mixed to the manufacturer's recommended ratio: 1 part Garlon 4 Ultra concentrate to four parts MSO (activator/oil). From which, CAST XIII derived the following recipe:

1 liter herbicide = 200 mL Garlon 4 Ultra, 800 mL MSO, and 1 – 2 mL of dye

On February 14 & 16, CAST XX cleared the sides of the runway of invasive vegetation and applied Garlon 4 Ultra to prevent regrowth. CAST XX treated a total of 9.16 acres, using a total of 8000mL of Garlon mixture, containing 1600mL of Garlon and 6400mL of activator. Per the manufacturer's label, there is 6045mL/L of active ingredient. Total active ingredient applied on those dates mentioned equaled 4826mL at a rate of 526.86 mL per acre applied.

Total Acre (ac)	Rate per Acre (L/acre)	Total Herbicide (mL)	Active Ingredient (mL/L)	mL Active Applied	mL Active Ingredient /Acre
9.16 ac	0.87L/ac	8000mL	6045mL/L	4826mL	526.86mL/ac

Table 3. Total acreage and Garlon active ingredient applied during CAST XX



Figure 30. Garlon 4 Ultra application along the runway

Trail Maintenance

Clearing of vegetation along the YCA 50m Monitoring Survey, YCA Treatment Monitoring Survey, and All-Island Ant Survey routes were a priority upon CAST XX's arrival to Johnston as a crew was not on island to maintain the routes from June 2020 to December 2020. After the routes were established, maintenance was conducted as needed, mostly after rainy period. At the end of the CAST XX deployment, the camp was broken down and all tiles and most flagging were pulled from these survey locations.

In addition to survey trail maintenance, CAST XX assisted with clearing routes for detector dog surveys, regularly maintained the roads on island, cleared the top of the Ant Cave to maintain structural integrity, cleared paths and vegetation around the US Air Force monitoring wells and maintained the runway (detailed in the Garlon Application section above).

Other Work

Tide Station Maintenance

The University of Hawaii monitors a tide station on Johnston Island to study sea level changes because of climate change. This research is being conducted in the same manner in over 80 other places around the world to provide data from a wide range of oceanic locations. The instruments and monitoring equipment, including two underwater tidal switches and a solar panel that powers a transmitter, are cleaned regularly to ensure proper data collection.

During the CAST XX deployment in December 2020, new transmitting equipment and batteries were installed. The tide station previously stopped transmitting data during CAST XIX. A piece of transmitting equipment malfunctioned, causing an approximate ten-month gap in data collection. After the installation by CAST XX, the tide station functioned properly for the entirely of the deployment. Additionally, Jason Klem, a University of Hawaii Tide Station Monitor, serviced the equipment in May 2021. Equipment was updated and replaced at the original Tide Station Peninsula and a new tide monitoring setup was installed along the western edge of the Wharf.

Weather

A number of basic weather conditions are measured and monitored regularly on Johnston Island. A rain gauge is located on the east side of camp and checked each morning, and a Kestrel unit mounted on a tripod near the runway collects temperature and wind data every hour on the hour.

During CAST XX's deployment the rain gauge was monitored daily, but the Kestrel was not functional as the time would become increasingly less and less accurate every hour. Therefore, only rain data was collected on CAST XX.

Table 4. Summary of rain gauge data for Johnston Atoll, January-May 2021.

Month	January	February	March	April	May (1 st -23 rd)
Rain (mm)	48.15mm	46.85mm	126.75mm	18.50mm	14.75mm

Outer Island Monitoring – December 2020

Mean Incubation Counts

Many of the seabird species exhibit asynchronous and aseasonal breeding behavior on Johnston Atoll. The number of active nests on the outer islands (North Island, Sand Island, East Island) of Johnston Atoll are typically monitored once every six months, as they are only accessible by boat, with the goal of monitoring the total breeding population of each species on each island. In 2020, the outer islands were only monitored once.



Figure 31. Shows the number of active nests of each seabird species surveyed on North Island, Sand Island and East Island on 12-13 December 2020.

In addition to the species charted above, an estimate of Sooty Tern eggs and chicks was made for each island. Sand Island had one egg and no chicks, East Island had three eggs and no chicks, and North Island had ~5,140 eggs and four chicks. Additionally, two Brown Noddy eggs were observed on East Island.

Ant Surveys

A collection survey to determine if YCA are present on the outer islands was done on North Island, Sand Island, and East Island on 12 & 13 December 2020. Fifty centrifuge tubes containing a spam cube were placed along transects on each island and then collected an hour later. Of the 150 tubes collected, no YCA were detected on any of the outer islands.

Camera Trap Notes

Sand Island:

Sand Island West #1 Camera: 24% full SD card, 17,661 pictures, full battery

- SD card and batteries changed
- New desiccant added
- Settings remain no motion sensor, 1 picture/30minutes on 24hrs interval
- Measured and painted markers that were already in place

Sand Island West #2 Camera: did not turn on, very rusty inside and bottom contacts for the batteries were unusable

- Collected SD card
- Replaced camera, solar panel and battery pack with the new camera that was meant to be deployed on the eastern end of Sand Island

Sand Island East: 26% full SD card, 17,666 pictures, full battery

- SD card and batteries changed
- New desiccant added
- Settings remain no motion sensor, 1 picture/30minutes on 24hrs interval
- Measured and painted markers that were already in place

East Island:

East Island West Camera: No longer functioning. Wiring torn and infested with ants (*Ochetellus glaber*). Camera was not replaced.

East Island SE Corner Camera: 14% full SD card, 17,665 pictures, full battery

- SD card and batteries changed
- New desiccant added
- Settings remain no motion sensor, 1 picture/30minutes on 24hrs interval
- Measured and painted markers that were already in place

North Island:

North Island Camera: 31% full SD card, 17,664 pictures, full battery

- SD card and batteries changed
- New desiccant added
- Settings remain no motion sensor, 1 picture/30minutes on 24hrs interval
- Added, measured and painted markers

Outer Island Monitoring – May/June 2021

Bird Observations

East Island: 27 May 2021

BRNO: on eggs and chicks of all stages present SOTE: a few on eggs but mostly loafers GBTE: on eggs, some chicks present, lots of loafers MABO: on eggs and chicks of various stages present BRBO: on eggs and chicks of various stages present WTSH: adults seen in burrows but no eggs or chicks seen RUTU seen around shoreline

North Island: 29 May 2021

BRNO: on eggs and early stage chicks present SOTE: a few with eggs and a few early stage chicks, lots of loafers present BRBO: late stage chicks present MABO: mostly late stage chicks present, one nest with young chick and one with eggs RFBO: late stage chicks present GBTE: all stage chicks present WTSH: adults present, no eggs or chicks seen GRFR: late stage chicks present PGPL and RUTU seen around shoreline

Sand Island: 28 May 2021

BRNO: on eggs and some early stage chicks present SOTE: on eggs and some early stage chicks present BRBO: chicks of all stages present but a lot of late stage chicks (6 and up) on causeway MABO: on eggs and some mid stage chicks present WTSH: mating RFBO: on eggs and chicks of all stages present GBTE: on eggs CHSH: present no eggs or chicks seen GRFR: lots of chicks in all stages present

Camera Trap Notes

Johnston Island

Wharf 1:

• Strapped to a wooden post near the NE corner of the wharf. Set to take one picture/hr, sunrise to sunset.

Wharf 2:

• Strapped to an Ironwood tree on the southern edge of the wharf (almost directly south of Wharf 1). Set to take one picture/hour, sunrise to sunset.

Sooty Tern Colony:

• Strapped to a large ironwood tree on the easternmost edge of the runway. Set to take one picture/hour, sunrise to sunset.



East Island

SE corner:

- Memory card 3% full, batteries full
- Number of pictures: 3998
- Memory card replaced with 64GB card, Lithium batteries and desiccant packets replaced.

SW corner:

- This site had no camera from last maintenance trip
- New camera was placed on post with a new lithium batteries and a 64GB SD card placed in camera

North Island

North:

- Memory card 7% full, batteries full
- Number of pictures: 8059
- Memory card replaced with a 64GB card and lithium batteries and desiccant packets replaced.

Sand Island

Sand West 1:

- Memory card 5% full, batteries full
- Number of pictures: 8021
- Memory card replaced with a 64GB card and lithium batteries and desiccant packets replaced.

Sand West 2:

- Memory card 6% full, batteries full
- Number of pictures: 8017
- Replaced camera, had to detach from solar panel and external battery, got new 32GB SD card, new Lithium batteries and new desiccant packets

Sand East:

- Memory card 6% full, batteries full
- Number of pictures: 8013
- Memory card replaced with 64GB card, lithium batteries and desiccant packets replaced.

Detector Dog Surveys

Project Lead and Summarization by Aisha Rickli-Rahman

After 3 years of having crews search diligently for YCA on Johnston, hand-searching surveys were turning up zero YCA. In an effort to bolster our confidence that YCA was no longer present, we contracted with Country Canine LLC to deploy two scent detection dogs to survey the infestation area for YCA. The canines, Solo and Guinness, are conservation dogs trained to detect many different odors. They spent approximately one year training on the scent of YCA to assist on this project. Not only did Solo and Guinness learn the scent of YCA, they were also trained to discriminate between the scent of YCA and other ants, such that they ignored the scents of any other any species. They were also trained to alert on the scent of YCA in places where they could not actually reach the ants i.e. cracks in the concrete, thick vegetation, etc. Handlers Kyoko Johnson and Michelle Reynolds worked with project lead and bio shadow Aisha Rickli-Rahman as well as another bio shadow, Keely Hassett to complete this survey work in December of 2020.



Figure 32. Aisha mapping the route surveyed by Kyoko and Solo (photo by Tor Johnson)

Each day the two teams would go to different pre-determined areas (determined by factors such as wind and other weather, priority level based on previous YCA detections, amount of vegetation that had been cleared prior to surveys, etc.) and survey for about 3 hours in the morning then again for about 2-2.5 hours in the evening. The two teams together surveyed for over 84 hours and covered a total of 73% of the treatment area (infestation area + buffer), a total of 141 acres (130 of which were within the treatment area), and tracked a total of 110

miles, combined. After all of this, no live YCA were detected. Guinness' average detection distance was 8.7 meters and Solo's was 4.5 meters. The dog's average detection distances in the environmental conditions on Johnston were determined by hiding training aids at the end of nearly every survey. Training aids included metal tea balls with dead ants contained inside, YCA scented gauze and YCA scented coffee filters. Training aids were left in various different areas in different types of habitat and at varying distances within vegetation. When the dog indicated it was "on scent", that spot was marked. Changes in behavior such as a quick change in walking direction, a sharp turn of the head, lifting the head or dropping it to the ground to sniff or a change in the dog's breathing pattern were often the best way to know if the dog was "on scent." When the dog eventually found and alerted on the training aid successfully (which they did every time), the distance between the training aid and the location where the dog was first "on scent" was measured, which provided the detection distance. A minimum of 10 different detection distances were calculated for each dog, and then averaged to give the average detection distance for the environmental conditions on Johnston Island. The average detection distance for each dog was then applied to each of their tracks, which allowed for the calculation of the total survey coverage.



Figure 33. Detector dog survey tracks from 4-17 December 2020.



Figure 34. Infestation Zone coverage on Johnston Island based on detector dog survey tracks from 4-17 December 2020.

Both Solo and Guinness did an incredible job and their handlers Kyoko and Michelle were also incredibly hard working as were the two bio shadows, Keely and Aisha. One major task that made this project possible was all of the trail clearing that was done by the dog teams, the CAST crew, other expedition members and even at times with the help of the boat crew. The work would also not have been possible if the non-native Golden Paper Wasp, which is very prevalent on Johnston Island, was actively nesting and therefore much more aggressive. Thankfully, all odds were in the favor or the project and it went incredibly well, demonstrating that it is definitely possible for detector dogs to be deployed on our remote Pacific island refuges, as long as certain very important factors are considered in advance and more importantly, planned for.



Figure 35. from left to right – Michelle Reynolds (dog handler), Guinness, Keely Hassett (USFWS), Aisha Rickli-Rahman (USFWS), Kyoko Johnson (dog handler) and Solo (photo by Tor Johnson)



Figure 36. Keely, Michelle and Guinness at work (photo by Tor Johnson)

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Appendix A

Marine Species Database:

Family	Genus	Species	Common Name
Chaetodontidae	Chaetodon	auriga	Threadfin Butterflyfish
Chaetodontidae	Chaetodontidae	trifascialis	Chevron Butterflyfish
Chaetodontidae	Chaetodon	citrinellus	Speckled Butterflyfish
Chaetodontidae	Chaetodon	lunulatus	Oval Butterflyfish
Chaetodontidae	Chaetodon	ornatissimus	Ornate Butterflyfish
Chaetodontidae	Chaetodon	ephippium	Saddleback Butterflyfish
Chaetodontidae	Chaetodon	quadrimaculatus	Fourspot Butterflyfish
Chaetodontidae	Chaetodon	multicinctus	Multiband Butterflyfish
Chaetodontidae	Forcipiger	flavissimus	Common Longnose Butterflyfish
Chaetodontidae	Chaetodon	unimaculatus	Keyhole/Teardrop Butterflyfish
Scaridae	Chlorurus	spilurus	Bullethead Parrotfish
Scaridae	Scarus	psittacus	Palenose Parrotfish
Scaridae	Scarus	rubroviolaceus	Ember Parrotfish
Scaridae	Scarus	dubius	Regal Parrotfish
Scaridae	Chlorurus	perspicillatus	Spectacled Parrotfish
Scaridae	Calotomus	carolinus	Stareye Parrotfish
Acanthuridae	Acanthurus	nigroris	Blueline Surgeonfish

Acanthuridae	Acanthurus	triostegus	Convict Tang
Acanthuridae	Acanthurus	achilles	Achilles Tang
Acanthuridae	Acanthurus	olivaceus	Orangeband Surgeonfish
Acanthuridae	Acanthurus	blochii	Ringtail Surgeonfish
Acanthuridae	Naso	lituratus	Orangespine Unicornfish
Acanthuridae	Naso	unicornis	Bluespine Unicornfish
Acanthuridae	Ctenochaetus	strigosus	Goldring Surgeonfish
Acanthuridae	Zebrasoma	flavescens	Yellow Tang
Acanthuridae	Zebrasoma	veliferum	Sailfin Tang
Mullidae	Mulloidichthys	flavolineatus	Square-spot Goatfish
Mullidae	Mulloidichthys	vanicolensis	Yellowfin Goatfish
Mullidae	Parupeneus	cyclostomus	Blue Goatfish
Mullidae	Parupeneus	multifasciatus	Manybar Goatfish
Mullidae	Parupeneus	insularis	Island Goatfish
Mullidae	Parupeneus	pleurostigma	Sidespot Goatfish
Labridae	Thalassoma	duperrey	Saddle Wrasse
Labridae	Thalassoma	quinquevittatum	Five-stripe Wrasse
Labridae	Thalassoma	lutescens	Sunset Wrasse
Labridae	Thalassoma	trilobatum	Christmas Wrasse
Labridae	Epibulus	insidiator	Slingjaw Wrasse
Labridae	Oxycheilinus	unifasciatus	Ringtail Wrasse
Labridae	Stethojulis	balteata	Belted Wrasse
Labridae	Gomphosus	varius	Bird Wrasse
Labridae	Coris	gaimard	Yellowtail Coris
Labridae	Thalassoma	purpureum	Surge Wrasse
Labridae	Thalassoma	ballieui	Old Woman Wrasse
Pomacentridae	Abudefduf	sordidus	Blackspot Sergeant Major
Pomacentridae	Dascyllus	albisella	Domino Damselfish
Pomacentridae	Plectroglyphidodon	johnstonianus	Blue-eye Damselfish
Balistidae	Rhinecanthus	aculeatus	Lagoon Triggerfish
Balistidae	Melichthys	niger	Black Triggerfish
Balistidae	Melichthys	vidua	Pinktail Triggerfish
Kyphosidae	Kyphosus	sandwicensis	Gray Chub
Kyphosidae	Kyphosus	vaigiensis	Brassy Chub
Tetraodontidae	Arothron	meleagris	Spotted Puffer
Tetraodontidae	Canthigaster	jactator	White Spotted Toby
Ostraciidae	Ostracion	meleagris	Spotted Boxfish
Ostraciidae	Ostracion	whitleyi	Whitley's Boxfish
Zanclidae	Zanclus	cornutus	Moorish Idol
Carangidae	Caranx	melampygus	Bluefin Trevally
Carangidae	Scomberoides	lysan	Leatherback

Carangidae	Caranx	sexfasciatus	Big-Eyed Jack
Carangidae	Carangoides	orthogrammus	Yellow-Spotted Jack
Lutjanidae	Aphareus	furca	Forktail Snapper
Carcharhinidae	Carcharhinus	amblyrhyynchos	Grey Reef Shark
Fistulariidae	Fistularia	commersonii	Cornetfish
Monacanthidae	Cantherhines	dumerilii	Barred Filefish
Synodontidae	Synodus	variegatus	Reef Lizardfish
Holocentridae	Sargocentron	spiniferum	Saber/Longjaw Squirrelfish
Aulostomidae	Aulostomus	chinensis	Trumpetfish
Kuhlidae	Kuhlia	sandvicensis	Hawaiian Flagtail
Sphyraenidae	Sphyraena	Barracuda	Great Barracuda