ARTIFICIAL BROOD PARASITE TECHNIQUE FOR REARING ABANDONED EGGS OF THE ENDANGERED LITTLE TERN *STERNULA ALBIFRONS*

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ABSTRACT

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The Little Tern *Sternula albifrons* is a ground-nesting species classified as Endangered on the Japan Ministry of the Environment's Red Data List. It may abandon its eggs owing to disturbances, a factor having a negative impact on its population trends. Successful artificial nurturing of these eggs could aid in the recovery of the species. To address this issue, I developed a 'brood parasite' technique. The process involves taking abandoned eggs from a nest, incubating them in a facility until one day before hatching, and then covertly placing the protected eggs into other nests within the colony. By these means, foster parents and chicks can establish their post-fledging relationship. Herein, I provide an overview of this technique.

Key words: abandoned eggs, colonial bird, endangered species, ground nesting, *Sternula albifrons*

INTRODUCTION

The Little Tern *Sternula albifrons* lays its eggs directly on coastal beaches and riverbeds, in nests having clear vistas to detect predators (Ehrlich et al., 1994). Because their nests are visible, predators can easily locate eggs and chicks (Japan Ministry of the Environment, 2014). Additionally, high water or tides may lead to flooding (O'Connell et al., 2014), reducing breeding success. The species is listed as Endangered on the Red List of Japan by the Ministry of the Environment (Category II). The unstable nesting environment of the Little Tern, the presence of predators, and human disturbance may be factors in egg abandonment, which can be a frequent occurrence within breeding colonies.

One possible strategy for restoring the Little Tern population is to utilize the abandoned eggs. In 2001 and 2002, abandoned eggs that were found submerged were put into incubators until hatching (Chiba City Zoological Park, 2023). A similar program has been in place for Piping Plovers *Charadrius melodus* since 1992 (Powell et al., 1997), and the method has contributed to population recovery (Roche et al., 2008). However, no cases have been confirmed of captive-reared Little Terns maturing and returning to Japan the following year. Artificial incubation and release programs have also been conducted for the closely related Least Tern *Sternula antillarum* in the USA (Faulkner, 2009), but with no confirmed cases of released individuals surviving into the following year. Unlike Piping Plovers, Little and Least tern chicks and juveniles continue to be fed by their parents after leaving the nest (Faulkner, 2009). It is thought that juvenile Little Terns gradually learn to catch fish on their own in the short period after fledging during which they continue to live with their parents. Since artificially born chicks do not have the opportunity to watch their parents catch food, they may have less ability to forage than individuals who have spent time with their parents. Thus, they may have difficulty surviving to the next year.

In Japan, many Little Tern colonies are situated in areas that are visible to the public, such as coastal beaches, and regular monitoring by researchers is not allowed because of this visibility. There is an issue with egg abandonment, and O'Connell et al. (2015) proposed a method for artificially fostering abandoned Little Terns eggs by placing them in nests with eggs of the same hatching date, which were found by frequent monitoring. However, in Japan, there is a strong preference for minimizing human intrusion into bird colonies, and because researcher presence in colonies is usually visible to the public, on-site monitoring of colonies is discouraged. Thus, information on the daily age of individual eggs, obtained through frequent entry into the colony, is unavailable. In addition, there is often depredation by crows *Corvus* spp. on disturbed colonies, which creates more incentive to minimize entry by researchers into colonies.

Therefore, I developed a technique to address the inability to estimate the age of eggs in the colony and to minimize predation by crows. This included determining the status of the eggs (abandoned or not), collecting the abandoned eggs, and placing them in an incubation facility. Once they had developed to the point just before hatching, I placed eggs into the nests of Little Terns in the colony, where they were raised by and having foster parents raise the eggs.

METHODS

Discovery of abandoned eggs and artificial incubation

On 03 June 2019, I conducted a single preliminary survey at a colony of about 50 Little Tern nests on a natural sandy beach in Oamishirasato City, Chiba Prefecture (35°49′N, 140°43′E). To ensure that the location of the nests could be determined from outside the colony, I marked nest locations with 18-cm bamboo strips placed in the sand 20–30 cm from each nest.

I conducted observations from outside the colony. If I determined that parents were not incubating their eggs, I collected them to avoid the risk of the eggs dying due to overheating when the surface temperature of the sand reached 50 °C (Andes, 2018; Kasielka, 2009). Little Tern parents temporarily leave the nest to mob when a natural predator (or researcher) approaches, and to avoid mistakenly interpreting the eggs as abandoned during such an episode, I chose to continue observation for a specific period when the parents were not in the nest. Koyama (2018) found that when Little Terns temporarily leave the nest for mobbing, only 4% of them do not return for more than 20 minutes. Based on these data, eggs in nests where the parents were away for more than 30 minutes were considered abandoned. I also decided to search for abandoned eggs in the morning, before the sunlight made the sandy ground surface too hot. Little Terns usually lay one egg per day and begin incubation after reaching a full clutch (2–3 eggs). I accounted for the possibility that the unincubated eggs were abandoned and collected them, placing commercial Japanese Quail *Coturnix japonica* (hereafter, quail) eggs of similar size and pattern in the nests. If the parent birds were not observed incubating the quail eggs for more than five days, it was determined that Little Tern eggs originally in the nest were in an abandoned state. The quail eggs used for confirmation of abandonment were then collected.

The abandoned eggs were transported to the incubation facility in Chiba City Zoological Park (CCZP) in Styrofoam boxes lined with cushioning material to reduce vibration and kept at about 37 °C during transportation. At the incubation facility, I used an egg candler to determine if the eggs were alive or dead (Daniels, 2019). In the incubators, I kept the eggs at 37.5 °C and 65%–67% humidity (the preferred factors for best hatching; Kasielka, 2009). Eggs were also automatically rotated 90° once per hour (Kasielka, 2009). Embryo development was checked daily using an egg candler.

Artificial brood parasitism of abandoned eggs

In the UK, O'Connell et al. (2015) promptly shifted abandoned tern eggs to other nests. However, owing to the high predation pressure on Little Tern eggs in Japan by the Large-billed Crow *Corvus macrorhynchos* and Carrion Crow *Corvus corone* (Japan Ministry of the Environment, 2014), an extended artificial incubation period was implemented to minimize predation damage. In the Lesser Elaenia *Elaenia chiriquensis*, artificially hatched chicks are returned to the nest to avoid high egg predation rates in the field (Lobo & Marini, 2013). However, given that terns establish a parent-chick relationship through vocal communication before hatching (Saino & Fasola, 2010), pre-hatched eggs must be more carefully placed into selected nests. Given that the average incubation period of the Little Tern is 19–22 days (Brazil, 1991) or 19–21 days (Nakamura & Nakamura, 1995), artificially incubated eggs should be placed in nests at 17–20 days; this is considered 1–2 days before hatching and the stage when unhatched chicks begin to vocalize. Eggs were returned to the colony using the same Styrofoam boxes that were used to transport them to the incubation facility, taking care to avoid vibration and heat retention.

If the developmental stages of the eggs in the nest and the artificially incubated eggs do not coincide, the timing of hatching may be off, and eggs that hatch late may be abandoned. Therefore, I sought nests at the developmental stage just prior to hatching, before placing the foster eggs. The method used to locate pre-hatching eggs in the field involved immersing the eggs in water and determining the developmental stage based on how well they floated (Hays & LeCroy, 1971), in combination with the presence or absence of calls by pre-hatched chicks. The floating egg technique does not affect hatchability (Hansen et al., 2011).

Ground-nesting seabirds are known to readily accept eggs other than their own (Divoky & Harter, 2010; Morales et al., 2010; Silva et al., 2007). To investigate whether Little Terns accepts eggs other than their own, I placed Japanese Quail eggs at different distances from Little Tern nests at another colony in Chiba, Japan, to see if the eggs would be transported to the nest by the next day. Little Terns actively accepted eggs placed within 30 cm of the nest (Table 1). When adding foster eggs for Little Terns, it is necessary to choose nests with one or two eggs to avoid exceeding the full clutch of three eggs, which would compromise incubation. I continued observation to see if the eggs in the nest hatched and if the chicks were cared for by their parents.

The ethical treatment of animals was approved by the Uekusa Gakuen University Animal Research Committee (URAC17-05). Additionally, permission to capture the birds was obtained from the Ministry of the Environment (Kanto Regional Environmental Office 18120712). Moreover, I am qualified to handle wild bird eggs by Yamashina Institute for Ornithology (No. 170779).

RESULTS AND DISCUSSION

Discovery of abandoned eggs and artificial incubation

As mentioned, nests were marked during a preliminary survey on 03 June 2019. At 10h00 on 16 June, fresh car tracks were observed upon my arrival, indicating potential human disturbance (Fig. 1). I examined each nest from outside the colony and decided that those in which parents were absent for more than 30 minutes had been abandoned. After collecting the tern eggs from these nests, quail eggs were substituted and the nests were monitored for five days. I observed no incubation of quail eggs or laying of new eggs, a pattern that indicated abandonment. Four eggs believed to

TABLE 1
Acceptance of Japanese Quail *Coturnix japonica* eggs by Little Tern *Sternula albifrons* in Chiba, Japan, in 2001[a]

Distance from nest (cm)	Quail egg uptake rate (%)	*n*
10	100	5
20	100	5
30	100	5
40	40	5
50	0	5
60	0	3
70	0	3
80	0	1
90	0	1
100	0	1

[a] Japanese Quail eggs were placed at various distances from Little Tern nests, and egg acceptance was evaluated after one day. Data modified from Hayakawa (2001).

have been abandoned were rescued and transported to the CCZP incubation facility by car. The entire process, from egg collection to transportation to the incubation facility, took approximately 70 minutes. Through observation with an egg candler, it was established that one egg (egg A) was ~17 days old. Two eggs (eggs B and C) were considered either alive or dead, and one egg (egg D) was confirmed dead.

Eggs A, B, and C were heated in an incubator. Given that Egg A was collected 3–4 days before hatching on 16 June, it was incubated for only 2 days. Egg B was found to be in a pre-warming state at the time of collection. Therefore, it was kept warm until the nineteenth day. Egg C was found to be dead.

Artificial brood parasitism of abandoned eggs

Egg A was transported to the colony on 18 June. Among the nests in which parents were incubating eggs, we found a nest (Nest 1) containing one egg that was determined to be close to hatching and inserted Egg A. When observed on 20 June, both chicks had hatched (Fig. 2). On 21 June, the parent was observed attending two chicks in the nest, indicating that the foster egg had been accepted. No chicks were sighted in the nest after 22 June. However, given that Little Terns are nidifugous and no dead chicks were observed, it appears likely that the parent bird relocated with the chicks.

Fig. 1. Tracks of a car that likely disturbed the Little Tern *Sternula albifrons* colony in Oamishirasato City, Chiba Prefecture, Japan. No nests are visible in this image. Photo credit: Masaharu Hayakawa, 10h00 on 16 June 2019

Fig. 2. Little Tern *Sternula albifrons* chicks hatched from Egg A and a host egg at a colony in Oamishirasato City, Chiba Prefecture, Japan. Photo credit: Masaharu Hayakawa, 20 June 2019

Egg B was transported to the colony on 05 July, and efforts were made to locate a nest containing eggs just before hatching. However, owing to the limited number of nests present during the late breeding stage, it was not possible to find nests with eggs on the verge of hatching. Therefore, Egg B was placed in a nest containing two eggs aged ~16 d (Nest 2). By 08 July, only the parasitized egg had hatched, resulting in two eggs and one chick. On 09 July, three chicks were observed, and the parent bird was keeping them warm. No chicks were observed on 10 July. Similar to Nest 1, no dead chicks were found, suggesting that the parent bird likely moved the chicks to a grassy area with plenty of places to hide. In Nest 2, the hatching dates were 1–2 days apart, but both the parasitized egg and the original eggs were successfully hatched and raised (Table 2).

It is known that ground-nesting birds generally lay one egg per day and incubate them after a full clutch is formed, so that they hatch at the same time (Ehrlich et al., 1994). However, Hayakawa (2001) surveyed entire active nests in a Little Tern colony and reported that only 13.4% of nests had all eggs hatch in a single day (Table 3). I have observed cases in which incubation begins before a full clutch is formed, and the surface temperature of the ground at the nesting site is high, so it is possible that effective incubation begins even before eggs are actively incubated. Since there is a difference in the hatching date even in the natural state, this suggests that it is acceptable for the hatching date of an artificially parasitized egg to differ by one or two days from that of the host eggs. Since fledging success is possible even if the expected hatching dates do not exactly match, artificial nest parasitism appears to be a viable method, even in small colonies or in late breeding stages with a small number of nests. Use of the described technique may be an effective technique for restoring the Little Tern population, a colonial ground-nesting bird that is threatened with extinction in Japan.

TABLE 2
Observations of the brood parasitism method employed in Little Tern *Sternula albifrons* nests near Oamishirasato City, Chiba Prefecture, Japan, in June–July 2019

	Time								
Egg	16 June	18 June	20 June	21 June	22 June	05 July	08 July	09 July	10 July
A	Collected the 17-day egg	Placed one egg in Nest 1	Two chicks hatched	Parental bird incubated two chicks	Lost track of two chicks				
B	Collected the 0-day egg	Placed egg in the artificial incubator	→	→	→	Placed two eggs in Nest 2	Observed one chick and two eggs	Observed three chicks	Lost track of three chicks

TABLE 3
Synchrony of hatching in Little Tern *Sternula albifrons* nests in Chiba, Japan, in 2001[a]

Clutch size	2 eggs	3 eggs
Hatch by one day	7 nests	6 nests
Hatch by two days more	25 nests	59 nests

[a] The percentage of eggs hatching in a single day was low (13.4%). Whether the clutch size was two eggs or three eggs, the last eggs laid were always the last to hatch. Data modified from Hayakawa (2001).

CONFLICT OF INTEREST

The author declares no conflict of interest.

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