Breeding success of Little grebe, *Tachybaptus ruficollis*, at a wastewater treatment facility in Khon Kaen University, Thailand: the influence of human activity.

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<td>Keithmaleesatti, Sarun; Khon Kaen University Faculty of Science, Environmental Science Thongcharoen, Kanokporn ; Khon Kaen University Faculty of Science, Environmental Science Doungkomna, Poramad ; Khon Kaen University Faculty of Science, Environmental Science Somjai, Kowit; Khon Kaen University Faculty of Science, Environmental Science Chaianunporn, Thotsapol ; Khon Kaen University Faculty of Science, Environmental Science</td>
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<td>Keyword:</td>
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Breeding success of **Little grebe**, *Tachybaptus ruficollis*, at a wastewater treatment facility at Khon Kaen University, Thailand: the influence of human activity.

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Abstract

Wetlands are crucial nesting areas for **Little grebe**, *Tachybaptus ruficollis*. Our study was conducted at a wastewater treatment facility at Khon Kaen University, northeastern Thailand, to determine whether human disturbance affected the breeding success of birds at wastewater treatment ponds in 2011, 2012 and 2016. We found 14, 8 and 14 active nests in 2011, 2012 and 2016, respectively. The results indicated that human disturbances likely caused the grebes to shift their nest locations. In the 2011 and 2016 seasons, when human disturbance was low, the hatching success was 70%. In 2012, a weed control program at the facility resulted in the dramatic hatching reduction to 9.7%. The results also indicate that some human activities, such as predation and habitat disturbance, could have dramatic effects on the nesting success of *T. ruficollis*.
Keywords: Breeding success, Human disturbance, *Tachybaptus ruficollis*, Wastewater treatment facility, Khon Kaen University.

1. Introduction

The number and diversity of wetland birds can serve as important bioindicators of ecosystem changes (Yuan et al., 2014). Wetlands provide resources such as food, water, and shelter that are necessary for the successful reproduction of many wildlife species (Bilal et al., 2013), yet human activities can produce changes to habitat structure and function that negatively impact the reproduction of many wildlife species (Carney & Sydeman, 1999; Rais, Kabeer, Anwar, & Mahmood, 2010). Previous studies have shown a variety of responses of the impacts of human disturbance on water birds (Weller, 1999), some of which impact the breeding success of seabirds, such as double-crested cormorants, *Phalacrocorax auritus* (Ellison & Cleary, 1978); least auklets, *Aethia pusilla*; and crested auklets, *A. cristatella* (Piatt, Roberts, Lidster, Well, & Hatch, 1990), and other waterfowl species (Batt et al., 1992). Black skimmers, *Rynchops niger*, are also sensitive to human activities during the egg-laying and incubation periods (Safina & Burger, 1983). Additionally, human predation was a cause of nest failure of horned grebes, *Podiceps auritus*, in Scotland (Summers, Mavor, & Hancock, 2009).

Hockin et al. (1992) suggested that the main reasons for lower breeding success are nest desertion and increased predation on eggs. Studies have shown that artificially constructed habitats are used as breeding sites by many bird species, including waterfowl (family Anseriformes) (Flake & Cieminski, 1996).

Artificial wetlands, such as water treatment facilities and large water reservoirs, have the potential to support bird species, such as white-headed duck, *Oxyura*. 
leucocephala, and marbled duck, Marmaronetta angustirostris (Giosa, Mammides, & Zotos, 2018). In Thailand, wastewater treatment plants often contain stabilization or oxidation ponds. In 2001, a wastewater treatment plant was built at Khon Kaen University. The wastewater treatment facility at Khon Kaen University has yielded sightings of several bird species, including the eastern cattle egret, Bubulcus coromandus, and the Chinese pond heron, Ardeola bacchus. Some birds have used this location as a breeding site, such as the little grebe, Tachybaptus ruficollis, and the black-winged stilt, Himantopus himantopus [Pers.Obs.].

*Tachybaptus ruficollis* (family Podicipedidae) is found across Europe, Asia, and Africa (Hockey, Dean, & Ryan 2005; Bilal et al., 2013; Athamnia et al., 2015).

According to Birdlife International (2018), its global conservation status is ‘Least Concern’ (Lc). The grebe is a resident and common bird in Thailand. It usually lives in freshwater wetland habitats, such as marshes, ponds, lakes and canals (Lekagul & Round, 1991). The grebe is an omnivorous bird, and its food consists of damselflies, small insects and freshwater algae (Khobkhet, 2000). In northeastern Thailand, the species is found in natural lake habitats, such as Nong Han, Udon Thani Province and Nong La Leng Keng, Khon Kaen Province [Pers. Obs]. The grebe was first observed at the wastewater treatment pond on the Khon Kaen University campus in 2007. Chicks were first observed at the wastewater facility in 2009 (Thongcharoen, Robson, & Keithmaleesatti, 2018). Nesting success data of *T. ruficollis* at wastewater treatment ponds are nonexistent in Southeast Asia. The two-fold purpose of this study was 1) to analyze the breeding success of *T. ruficollis* at the wastewater treatment ponds in 2011, 2012 and 2016 and 2) to determine the possible effects of human disturbance on the
breeding success of *T. ruficollis* and highlight the influences of human disturbance on the breeding success of a water bird that lives in an anthropogenic habitat.

2. Materials and Methods

Study area and study species

Khon Kaen University, located in northeastern Thailand, occupies approximately 8.8 km² (880 ha) of land that includes education buildings, commercial zones, dormitories, agricultural and aquaculture farms, natural ponds, a dry dipterocarp forest, and recreational parks. In 2001, a wastewater treatment facility was built on a 21.44 ha area on the west side of the campus (near 16° 46’ E, 102° 80’ N) and has been operational since 2003. There are two wastewater systems and ten oxidation ponds in the area. The first system is for hospital discharge. It includes 2 facultative ponds, ponds 1 and 2 (each 3 ha in size), and 3 maturation ponds, ponds 3, 4, and 5 (each 1.36 ha in size). The second system is for the treatment of municipal wastewater. It consists of 2 facultative ponds, ponds 6 and 7 (each 3.75 ha in size), and 3 maturation ponds, ponds 8, 9, and 10 (each 1.20 ha in size) (Figure 1). *Typha* spp. is the dominant emergent vegetation along the edge of each pond. The water surfaces of ponds 1, 2, 3, 6 and 7 are covered by *Pistia* spp. during some months of the year. The depths of the facultative ponds and maturation ponds are 1.90-2.50 meters and 1.00-2.00 meters, respectively.

Human activity within the area includes weed control (cutting of weed and grass), which is performed in the area approximately five times per year. Furthermore, sediment was dug from all the ponds by a tracked excavator from August 2011 to August 2012.

Study design and data collection
The breeding success of little grebe was observed during three breeding seasons in 2011, 2012 and 2016. A small population of grebes foraged and inhabited ponds 1, 2, 3, 6 and 7. During both the 2011 and 2016 breeding seasons, ponds 1, 2, 3, 6, 7, and 8 were free of human activity, such as weed management and sediment removal. After August 2011, ponds 1, 2, 6 and 7 were altered due to dredging, weed control, and construction projects. However, the facultative ponds (ponds 1, 2, 6 and 7) were not used for fishing because of high levels of organic pollution. Ponds 4, 5, 9 and 10 experienced disturbances during the three seasons as a result of fishing pressure from local people. No nests were found in the study area between August 2011 and April 2012.

We measured and compared the breeding period, nest size, nest location, egg size, egg weight, clutch size, and hatching success of T. ruficollis over three breeding seasons. Nest searches were conducted by boat 3 times per week using a telescope during the breeding season. Each survey lasted three hours (from 8:00 to 11:00). The nests were marked with small plastic ribbons, and their locations were recorded by a global positioning system (GPS). The length and breadth of nests were measured using a measuring type, and the eggs were measured using a Vernier caliper (±0.01 mm), and each was weighed on a digital scale in the field. The observers were different among the years.

Data and statistical analyses

An active nest was defined as a nest where incubation was observed. An inactive nest was a nest in which a little grebe built but did not lay its eggs. Nesting success was defined as the point when at least one egg per clutch hatched, and hatching was assumed
to take place at a point in time when the first young bird broke free of its shell (Mayfield, 1975). Hatching success was calculated by summing all the eggs hatched out of the total number of eggs. Nesting success was estimated based on the proportion of the active nests where at least one egg hatched. Furthermore, nest success was calculated using the Mayfield method (1975), where daily nest survival = (DSR)^d, such that

\[ DSR = 1 - \frac{\text{No. of failed nests}}{\text{No. of exposure day}}. \]

We estimated the breeding period (d) of fifteen successful nests as 22.4 days.

The egg data from the three seasons were analyzed using descriptive statistics of the mean and the standard deviation (SD). In addition, one-way ANOVA at \( \alpha = 0.05 \) was used to detect the differences in egg width, egg length and egg weight among the three seasons. If there were any significant difference among the seasons, Tukey’s multiple comparisons of the means were further used for the pairwise analysis.

**Ethics statement**

This study was performed in strict accordance with the recommendations of the Wildlife Preservation and Protection Act, B.E. 2535, and the Animals for Scientific Purposes Act, B.E. 2558. In the 2016 season, the study was permitted (DNP no. 0909.204/9787) by the Department of National Parks, Wildlife and Plant Conservation, Ministry of Natural Resources and Environment and the Khon Kaen University Committee on the Ethics of Animal Experiments, permit number ACUC-KKU-36/60.

3. Results

**Nest location and breeding season**
The three years of data indicated that *Tachybaptus ruficollis* breeding occurred from the rainy season to the early winter season in northeastern Thailand, from late May to late November, at this wastewater site. In 2011, fifteen nests (fourteen active and one inactive) were observed during the breeding season. In 2012, twelve nests (eight active and four inactive) were observed in the treatment area; however, some grebes were disturbed by human activities before egg laying occurred. In 2016, twenty active nests were found in the breeding season. The nest locations were likely different in the three seasons. In 2011, 14 nests (93.3\%) were located in ponds 6 and 7, and only one nest was located in pond 2. However, eight nests (66.7\%) were found in ponds 1 and 2 in 2012 (May-July) and 4 nests were found in ponds 6 and 7 (August). In 2012, the nest locations changed after a weed control program was performed. The weeds around ponds 6 and 7 disappeared completely between May and June 2012. In July, the weed control program stopped, and vegetation re growth was rapid during July and August. Four grebe nests were found on ponds 6 and 7 in August. In 2016, all the nests were (100\%) again found on ponds 6 and 7 when there was no human disturbance at these ponds (Figure 2).

**Breeding ecology**

The *T. ruficollis* nests were built as floating platforms near the shoreline or in shallow water. The adult grebes used submerged or emergent vegetation such as *Typha* spp. or *Ipomoea aquatic* growing nearby for nest construction purposes. The nest dimensions were between 24.0 and 42.0 cm (n =47), and the height above the water level was 3.0-14.0 cm (n = 47). The clutch size of *T. ruficollis* in the three seasons was 1-6 eggs per nest (n= 42). The most frequent clutch size was 4 eggs (38.10\%). The mean clutch size was $3.55 \pm 1.19$ eggs (n= 42 active nests) in the three seasons, and the
average clutch size per nest was 3.86 ± 0.77 eggs (n=14) in 2011, 3.88 ± 0.69 eggs (n=8) in 2012 and 3.20 ± 1.51 eggs (n=20) in 2016. The eggs were elliptical-shaped and white in color. The average width, length and weight of the eggs in the three seasons were 25.32 ± 1.17 mm, 35.35 ± 1.56 mm, and 12.23 ± 1.23 g, respectively (n=148) (Table 1). One-way ANOVA showed that the mean egg length among the three seasons was not significantly different; however, the mean egg weight and width were significantly different (p<0.05) (Table 2). The mean egg weight and width in 2016 were less than in 2011 and 2012. The incubation period of *T. ruficollis* (n=15) was approximately 20-26 days in 2011 and 2016. In 2012, we were not able to measure the incubation period because the nests were only found after clutch completion. The number of successful nests in 2011, 2012 and 2016 were fourteen (100.0% apparent success), one (12.5%) and fourteen (70.0%), respectively (Table 3). The hatching success of the eggs was 74.1% in 2011 (n=53 eggs), 81.0% in 2016 (n =63 eggs) and only 9.70% in 2012 (n=31 eggs). The nest successes by the Mayfield method in 2011, 2012 and 2016 were 100.0%, 23.8% and 68.6%, respectively.

4. Discussion

Regional comparison of the breeding season

Previous studies have indicated variability in the breeding season of *Tachybaptus ruficollis*, but we found that it was between May and November during the three years of study at this particular site. In India, it was reported that the breeding season of grebes is also from May to August (Bates & Lowther, 1952; Ali & Ripley, 1983; Fazili, Shah, Jan, & Bilal, 2008). In Europe, it was observed that *T. ruficollis* breed from February to September, with the peak occurring in May (Moss & Moss, 1993; Harrison
& Castell, 2002). In Thailand, Khobkhet (2000) reported that *T. ruficollis* breed during the rainy season, from June to September. Data from a large freshwater lake in the Bung Bora Phet non-hunting area, Nakorn Sawan Province, Thailand, indicated that the breeding season of *T. ruficollis* was also between May and November, with a peak (the highest number of active nests) from July to September (Eiamampai, 2006). Food abundance might determine the breeding season of grebes at Khon Kaen University because the main food sources of *T. ruficollis* are aquatic invertebrates such as damselflies, shrimp, small aquatic animals and freshwater algae (Khobkhet, 2000), which may be more abundant during the rainy season (Leigh, Rand & Windsor, 1996).

**Breeding ecology and human disturbance**

The clutch size of *T. ruficollis* at the wastewater treatment facility during the three seasons ranged from 1 - 6 eggs with an average of 3.6. Other data from India indicated a range of 3-6 eggs with an average of 4.7; and in Algeria, the clutch size varied from 2-7 eggs with an average of 4.7; however, in Britain and Ireland, 1-8 eggs and a modal clutch size of 4 were reported (Athamnia et al., 2015; Fazili et al., 2008; Moss & Moss, 1993). The average dimensions of 148 eggs in the three seasons (35.35 ± 1.56 mm x 25.32 ± 1.17 mm) were similar to those reported by Bates and Lowther (1952); Ali & Ripley (1983); Khobkhet (2000); Fazili et al. (2008); and Athamnia et al. (2015) (36.6 mm x 25.1 mm, 36 mm x 25 mm, 35.6 mm x 25.51 mm, 36.5 ± 1.18 mm x 25.2 ± 0.48 mm and 36.3 ± 1.2 mm x 25.8 ± 0.7 mm, respectively). Furthermore, our data indicated that the mean egg weight and width were significantly different in 2016 than those in 2011 and 2012. However, Eeva and Lehikoinen (1995) suggested that the intraspecific variation in egg size of birds depends on the size and age of the females. Therefore, the
breeding females in 2016 might have been younger and smaller (e.g., first breeding birds) in size than the females in 2011 and 2012.

This study showed that the hatching success and nest success were high in 2011 (74.1% and 100.0%, respectively) and 2016 (81.0% and 68.6%, respectively), but it sharply declined in 2012 (9.7% and 73.8%, respectively). In 2012, three nests (42.9%) were destroyed by mowing, and the eggs in four nests (57.1%) were stolen by workers at the site since they were highly visible after mowing occurred (Pers. Obs.). In comparison, Fazili et al. (2008) indicated that the hatching success at a site in Kashmir, India, was 79.8%. Furthermore, Moss and Moss (1993) reported a 53.0% hatching success in Britain and Ireland. Although the hatching success rates in 2011 and 2016 were not very different from previous studies, the success rate declined dramatically in 2012 as a result of human activity. Weed control at the water treatment facility during the breeding season actually changed the nesting behavior of the grebes. It led to a lack of nesting materials, which is important for little grebe because weeds are used for maintaining temperature and humidity and deterring visual predators (Prokop & Trnka, 2011). Human activity might also affect the nest site selection and hatching success of grebes since changes occurred between the three observation periods. In 2012, ponds 1 and 2 were first selected as nest sites due to the weed control measures on ponds 6 and 7, and after the weed control program finished in August 2012, active nests were found again on ponds 6 and 7. As mentioned previously, all of the eggs in these four nests were stolen by workers at the site. Additionally, all the nests were found on ponds 6 and 7 after the human impacts declined in 2016. We believe that the two facultative ponds had lower disturbances than the other ponds because their edge slopes are very steep and are 0.5-1 meters deeper than the other ponds. For this reason, the grebes foraged in
these ponds. The shift of nesting sites between seasons due to human activities and
human predation is consistent with another study on closely related species, e.g.,
Athamnia et al. (2015) found that predation and adverse weather are important causes of
little grebe nest failure. In our study, there was adverse weather, such as heavy rain
during the rainy season. The precipitation increased the water level in the ponds and
flooded four nests in the 2016 season. Furthermore, Fournier and Hines (1999) reported
that two main factors, including the risk of predation by common raven and raccoons
and the distance of the nest from the shore and the distance from the nest to open water,
determined the position of horned grebe Podiceps auritus nests.

In this study, we showed that humans can affect bird breeding and nesting activity
both positively and negatively. The wastewater treatment ponds at Khon Kaen
University provided a suitable habitat to attract the grebes, as measured by nest
construction and breeding success. Similarly, Tucakov and Puzovic (2006) reported that
waterbirds in the family Anatidae, Rallidae, and shorebirds in the family Charadriidae,
Scopacidae, used the wastewater pools of sugar refineries as breeding areas. Orlowski
(2013) also reported that wastewater treatment wetlands in the southern United States,
Australia and Africa are important for breeding, foraging and stop-over areas for many
bird species. Therefore, the wastewater treatment pond at Khon Kaen University could
serve as an attractive breeding site for other waterbirds as well. However, this study also
noted that human disturbances near the pond areas, such as weed control or egg
predation, have a negative influence on breeding success. Such activities may have led
to the overall reduction of hatching success (from over 70% to less than 10%).

5. Conclusions
This study is the first report of the nesting activity of *T. ruficollis* at a wastewater treatment facility in Southeast Asia. The data showed that the little grebe adapted and established a new breeding colony in an anthropogenic habitat, as measured by nesting success. The results also indicate that some human activities, such as weed control, can have dramatic effects on the behavior of *T. ruficollis*. Our study showed the importance of artificially created habitats and proper management schemes to benefit some waterbirds and ensure their survival. However, the management of such habitats without ecological knowledge might have disastrous effects on these same species. Policies should reflect both environmental and social concerns.

**Acknowledgments**

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**References**


Figure 1. Overview of the wastewater treatment facility at Khon Kaen University and the location of sample collection site (Thongcharoen, Robson & Keithmaleesatti, 2018).

Figure 2. Nest location of little grebe in three seasons (2011, 2012 and 2016). Yellow indicates successful nests (hatching), and orange indicates active but failed nests (no hatching). Purple represents inactive nests (no eggs). The number indicates the order in which the nests were established in a given season.
Table 1. Little grebe *T. ruficollis* egg characteristics during seasons 2011, 2012 and 2016 at Khon Kaen University wastewater treatment facility.

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<tr>
<th>Egg characteristic</th>
<th>2011 season (n=54) mean ± S.D.</th>
<th>2012 season (n=31) mean ± S.D.</th>
<th>2016 season (n=63) mean ± S.D.</th>
<th>Total (n =148) mean ± S.D.</th>
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<tr>
<td>Egg width (mm.)</td>
<td>25.61 ± 1.36</td>
<td>25.72 ± 0.66</td>
<td>24.89 ± 1.04*</td>
<td>25.32 ± 1.17</td>
</tr>
<tr>
<td>Egg length (mm.)</td>
<td>35.64 ± 1.65</td>
<td>35.06 ± 1.78</td>
<td>35.22 ± 1.33</td>
<td>35.35 ± 1.56</td>
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<tr>
<td>Egg weight (g)</td>
<td>12.76 ± 1.13</td>
<td>12.39 ± 1.33</td>
<td>11.68 ± 0.98*</td>
<td>12.22 ± 1.22</td>
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* significantly different by one-way ANOVA (p<0.05)

Table 2. Analysis of variance for the (A) egg width, (B) egg length and (C) egg weight among years.

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<td>Year</td>
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<td>Year</td>
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<td>217.25</td>
<td>18.593</td>
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Table 3. Comparison of the clutch size, success nests and failed nests of active nest of the Little Grebe *T. ruficollis* in season 2011, 2012 and 2016 at the Khon Kaen University wastewater treatment facility.

<table>
<thead>
<tr>
<th>Clutch size</th>
<th>Number of nest season 2011 (Success : Failed)</th>
<th>Number of nest season 2012 (Success : Failed)</th>
<th>Number of nest season 2016 (Success : Failed)</th>
<th>Total nest (%)</th>
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<tr>
<td>1</td>
<td>0 (0:3)</td>
<td>0 (0:3)</td>
<td>3 (0:3)</td>
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<tr>
<td>2</td>
<td>0 (0:3)</td>
<td>0 (0:3)</td>
<td>3 (0:3)</td>
<td>3 (7.1%)</td>
</tr>
<tr>
<td>3</td>
<td>5 (5:0)</td>
<td>2 (0:2)</td>
<td>6 (6:0)</td>
<td>13 (31.0%)</td>
</tr>
<tr>
<td>4</td>
<td>6 (6:0)</td>
<td>5 (1:4)</td>
<td>5 (5:0)</td>
<td>16 (38.1%)</td>
</tr>
<tr>
<td>5</td>
<td>3 (3:0)</td>
<td>1 (0:1)</td>
<td>2 (2:0)</td>
<td>6 (14.3%)</td>
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<tr>
<td>6</td>
<td>0 (1:0)</td>
<td>0 (1:0)</td>
<td>1 (1:0)</td>
<td>1 (2.4%)</td>
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<tr>
<td><strong>Total</strong></td>
<td><strong>14 (14:0) 100.0%</strong></td>
<td><strong>8 (1:7) 12.5%</strong></td>
<td><strong>20 (14:6) 70.0%</strong></td>
<td><strong>42 (100.0%)</strong></td>
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