

## **Grazing of Herbivories Affecting Plant Growth: A Case Study of Water Buffaloes Grazing on Red Indian Water Lily at Thale Noi Wildlife Sanctuary, Southern Thailand**

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### **Abstract**

In this study, we investigated how buffaloes grazing on water lily influenced water lily leaf at Thale Noi Wildlife Sanctuary, southern Thailand. We estimated water buffalo population size, and measured water quality parameters and water lily leave sizes with present and absent of water buffaloes. Water buffalo population size were estimated by using aerial photographs from multi-rotor drones at five study sites. Photographs were taken at 50 m above ground. We measured water quality parameters (i.e. water temperature, pH, dissolved oxygen (DO), total dissolved solid (TDS) and electric conductivity (EC)) at two sites: site with buffaloes present and site with buffaloes absent. We collected 30 water lily leaves per study site at three study sites based on distances further away from water buffalo herds: short distance (100 m), intermediate distance (200 m) and long distance (300 m) from a buffalo herd of 200 buffaloes. We took photographs of water lily leaf with scale, measured leaf radius and calculated the leaf area with the Adobe Photoshop CS6 Extended software. Our results showed that there were five water buffalo herds in Thale Noi Water Sanctuary with a total of 176 males, 243 females and 105 juveniles. There were two herds that female biased and three herds with equal sex ratios. In areas with buffaloes present, water temperature, pH, DO, water lily radius and leaf area were higher but TDS and EC measurements were lower than areas without buffaloes. This indicates that water lilies and water quality parameters were influenced by water buffaloes.

**Keywords:** Water lily leaf, Buffalo, Thale Noi, Phatthalung

### **Introduction**

Large herbivores regulate the nutrient cycling and primary production in grassland ecosystems. These herbivores can mediate changes in plant community composition, production and the deposition of feces [1-3]. Feces and urine from large herbivores provide a fast decomposition pathway in grasslands by returning highly decomposable resources that are rich in plant-available nutrients [4]. Herbivores consumed 30 ~ 50% of aboveground plant biomass annually [5-6]. The highest excretion of  $\text{Na}^+$  was observed through feces of buffaloes and was significantly higher during summer than in winter [7]. Buffaloes are used for conservation grazing in the Black Sea area to maintain optimal conditions for bird life in a nature reserve [8].

Thale Noi Wildlife Sanctuary is the largest freshwater lake in the south of Thailand (about 285,625 Rai) covering three provinces: Phatthalung, Songkhla and Nakhon Si Thammarat provinces. The average temperature throughout the year is  $28.1^\circ\text{C}$  with a range of  $18.0$ -  $39.4^\circ\text{C}$  [9]. Thale Noi Wildlife Sanctuary is consisting of paddy, swamp and grassland. Biodiversity of Thale Noi Wildlife Sanctuary pronounced as a wetland of the world or first Ramsar Site of Thailand covering with water plants such as water hyacinth, various species of water lilies and floating plants with an average depth of 1.25 m [10].

Now the ecology of Thale Noi Wildlife Sanctuary is changed due to several anthropogenic factors such as water pollution, heavy metal contamination, land loss from invasion for agriculture, and rapid spread of weeds from excessive fertilizers used in agriculture. This anthropogenic effects reduce diversity and abundance of aquatic plants, and animals in many aspects. Red Water lily, the symbol of Thale Noi, is greatly reduce in numbers and area covered [11-13]. Red water lily serves as one of the major food source for water buffaloes. In this study, we conducted three field experiments to investigate the effect of large herbivore (water buffaloes) grazing on water lilies at Thale Noi Wildlife Sanctuary in southern Thailand. We tested the following hypotheses: (1) if water buffaloes affect the water lilies, then water lily leaf radius and area should be negative association with increasing distances from water buffaloes, (2). The quality of water are differ between buffaloes area and without buffaloes.

## Materials and Methods

### Study site



**Figure 1** (a) map of Patthalung province, Thailand, (b) map of Thale Noi Wildlife Sanctuary, (c) water lily study sites and (d) buffaloes study site



Figure 2 Buffaloes study sites

### Buffalo Population Estimation

The water buffalo (*Bubalus bubalis*) has a body length of 240 to 300 cm and a tail length of 60 to 100 cm. Adult buffalo skin color varies from light to dark brown. In this study, we used the DJI Drone Spark with a camera resolution of 12 MP [14-15] to take top-viewed photos of buffalo herds from 50 meters high above ground (Figure 3a). We counted the number of males, females and juveniles from the photographs [16]. The areas were calculated by using an Adobe Photoshop CS6 Extended program [17] (Figure 3b) and used to estimate a water buffalo density.

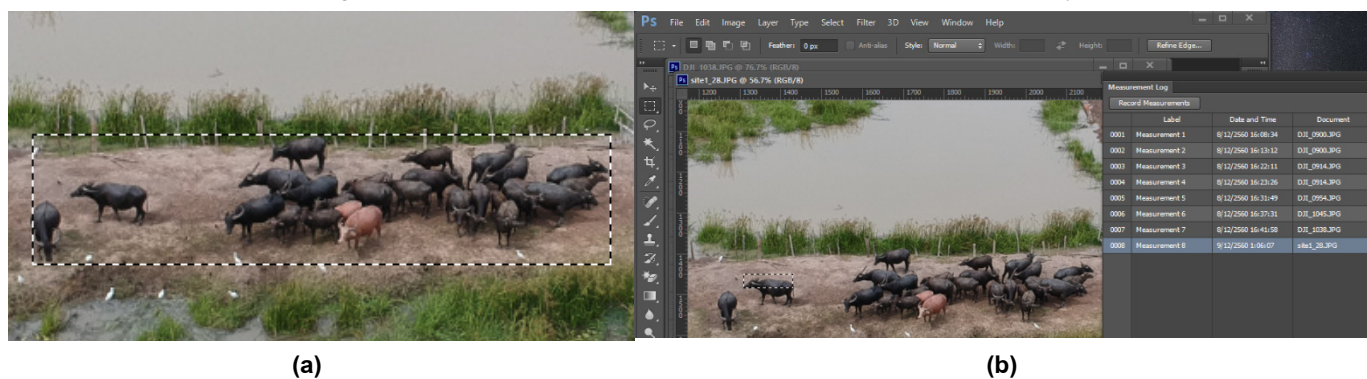


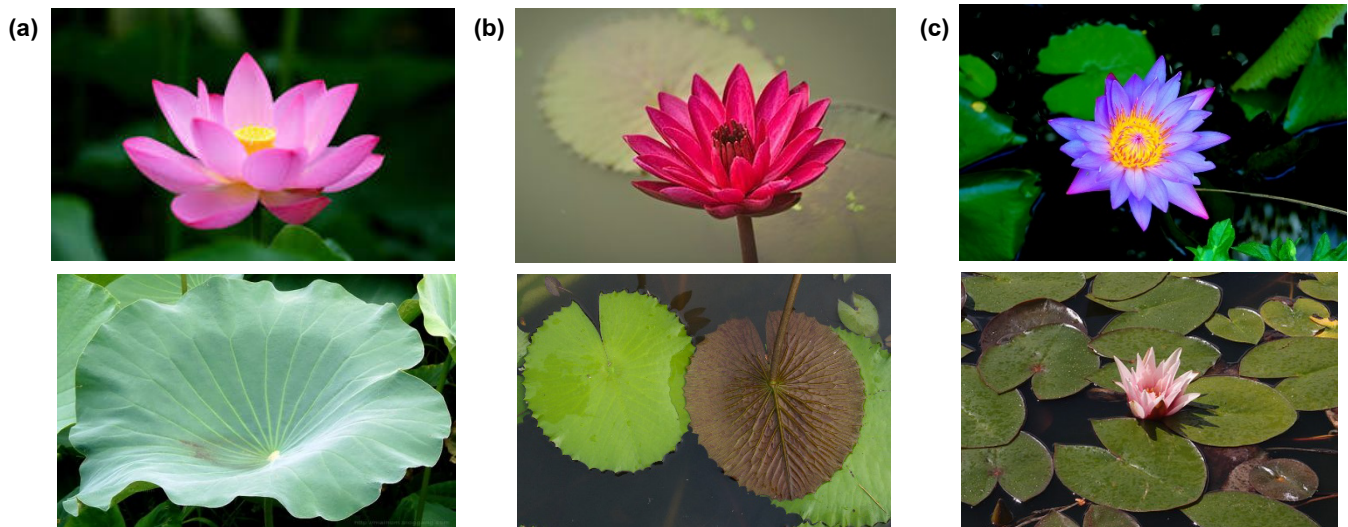
Figure 3 Top view drone photography. (a) buffalo herd and (b) area measurements in the Photoshop CS6 Extended software

### Water Lily Data Collection

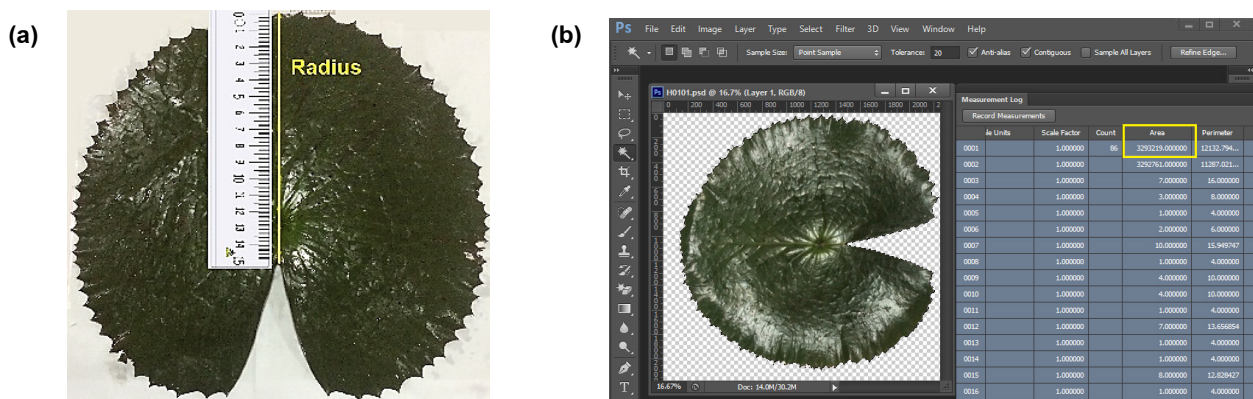
There were three water lily species present in Thale Noi Wildlife Sanctuary: Sacred lotus (*Nelumbo nucifera* Gaertn), Red Indian water lily (*Nymphaea pubescens* Willd) and water lily (*Nymphaea stellata* Wild) (Figure 4). The dominant species of this area and used for commercial purposes was the Red Indian water lily. In this study, we collected



Red Indian water lily leaves at three sites in October 2017: 100, 200 and 300 m further away from water buffalo herds with 30 leaves per site. We measured leaf radius and area with an Adobe Photoshop CS6 Extended program (**Figure 5**).



**Figure 4** Water lily species present at Thale Noi Wildlife Sanctuary. (a) Sacred lotus (b) Red Indian water lily and (c) Water lily



**Figure 5** Water lily leaf measurement (a) leaf radius (b) leaf area by an Adobe Photoshop CS6 Extended program

### Water Quality Measurement

We measured water quality parameters three times per site at two study sites: (1) site with buffaloes present and (2) site with buffaloes absent. Water quality parameters were composed of water temperature, pH, dissolved oxygen, total dissolved solid and electric conductivity [18-19].

## Data Analysis

We used crosstab chi-square tests to test the sex ratio of male and female at each study site. We used t-tests to test the differences in water quality measurements and water lily leave size between areas with buffaloes and no buffaloes. Regression analysis showed the relationship between distances from buffaloes and the size of water lily leaves (radius and area of leaf). All significant tests were two tailed at a significant level of 0.05.

## Results and Discussion

There are two sites that female biased sex ratios and three sites that the number of juveniles was closed to the number of male buffaloes (**Table 1**). Water buffalo density was ranging from 0.03-0.30 individuals/m<sup>2</sup> (**Table 2**). At first study site, we take a photo while the buffaloes were in the barracks and the second site was the widest feeding area. In areas with buffalo present, water temperature, pH, DO were higher but TDS and EC were lower than area without buffaloes (**Table 3**). Water lily leave radius and areas at distances at 100 m away from buffalo herds were larger than at 200 and 300 m away from the buffalo herds (**Table 4**). Water lily leave radius and areas were negative associated with distances away from buffalo herds (**Figure 6**).

**Table 1** The number of male, female and juvenile buffaloes in five study sites at Thale Noi Wildlife Sanctuary, southern Thailand (\* $P < 0.05$ ).

Study Sites	Males (M)	Females (F)	Juveniles (J)	Statistical test (M:F)
1	7	12	9	$\chi^2_1 = 1.32$
2	18	21	10	$\chi^2_1 = 0.23$
3	35	55	32	$\chi^2_1 = 4.44^*$
4	44	69	27	$\chi^2_1 = 5.53^*$
5	72	86	27	$\chi^2_1 = 1.24$
Total	176	243	105	$\chi^2_1 = 10.71^*$

**Table 2** Water buffalo densities at Thale Noi Wildlife Sanctuary.

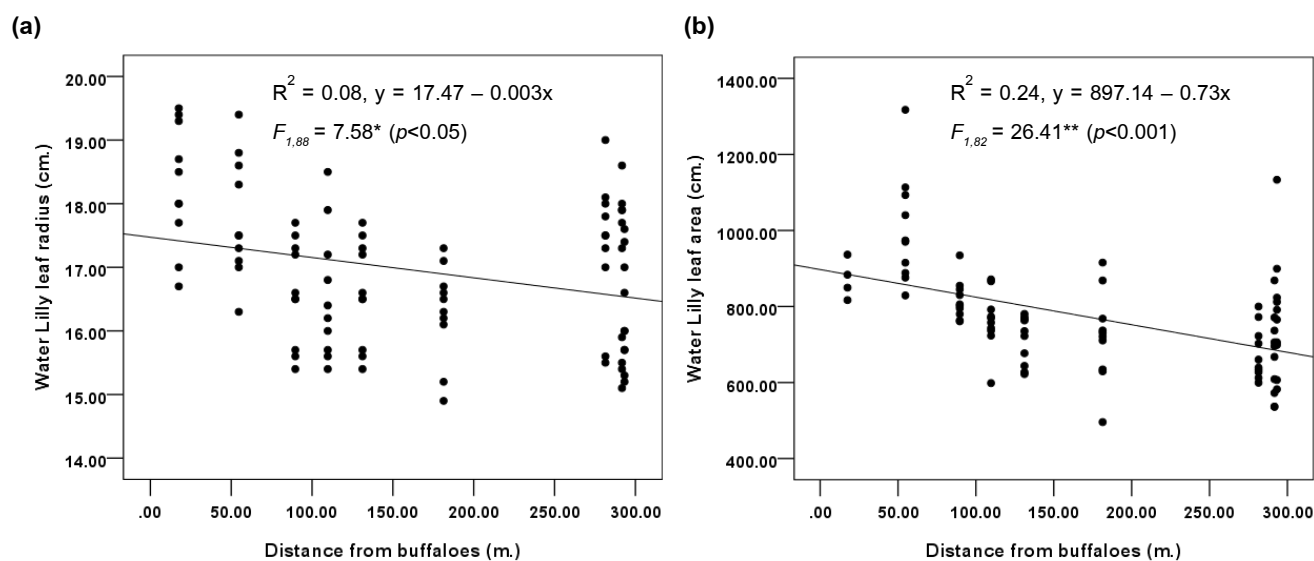
Study Sites	The number of buffaloes	Area (m <sup>2</sup> )	Buffalo density (individuals/m <sup>2</sup> )
1	28	92.37	0.30
2	49	1,678.80	0.03
3	122	3,989.07	0.03
4	140	3,641.08	0.04
5	185	2,002.74	0.09
Total	524	11,404.06	0.05

**Table 3** Mean ( $\pm$  SD) of water quality measurements in area with buffaloes and without buffaloes (\* $P < 0.05$ ).

Parameter	Buffalo present	Buffalo absent	t - test
Temperature ( $^{\circ}\text{C}$ )	29.94 $\pm$ 0.53	29.07 $\pm$ 0.66	$t_{16} = 3.13^*$
pH	7.21 $\pm$ 0.0	6.53 $\pm$ 0.11	$t_{16} = 14.91^*$
DO (mg/L)	4.61 $\pm$ 0.93	2.83 $\pm$ 0.87	$t_{16} = 4.20^*$
TDS (ppm)	25.32 $\pm$ 0.26	33.77 $\pm$ 0.37	$t_{16} = -55.35^*$
EC ( $\mu\text{S}/\text{cm}$ )	501.67 $\pm$ 2.00	637.78 $\pm$ 3.67	$t_{16} = -97.77^*$

**Table 4** Mean ( $\pm$  SD) of water lily leaf radius and area at three distances away from buffaloes (\* $P < 0.05$ ).

Water lily leave	Distances from buffaloes (m)			Statistical test
	100 m	200 m	300 m	
Radius (cm)	17.55 $\pm$ 1.14 <sup>a</sup>	16.49 $\pm$ 0.86 <sup>b</sup>	16.84 $\pm$ 1.15 <sup>b</sup>	$F_{2,87} = 7.897^*$
Area ( $\text{cm}^2$ )	903.07 $\pm$ 131.38 <sup>a</sup>	731.75 $\pm$ 89.61 <sup>b</sup>	709.56 $\pm$ 124.59 <sup>b</sup>	$F_{2,81} = 21.67^*$

**Figure 6** Water lily leave size and the distances from buffaloes. (a) water lily leave radius and (b) water lily leave area

## Conclusions

At Thale Noi Wildlife Sanctuary, southern Thailand, we investigated five study sites and found that the ratio of male, female and juvenile were significant differ. When considering each distance, found that only two sites that male different from female buffaloes. Three sites where the number of juvenile were close to the number of male. Water impurities affect the growth of aquatic plants. Use of irrigation water salinized by NaCl with an electrical conductivity (EC) more than twice as high as that recommended for optimal calla growth [20]. Water Lily *Nymphaea* has potential as an optimal, highly effective phytoremediation tool for the removal of Cd from polluted waste sources [21-22]. Temperature, pH and DO in buffalo area were higher than without buffalo while TDS and EC were lower. Research related to leaf changes, for example Herbivory drastically increased leaf senescence, reducing leaf density [23]. Distances affecting the size of water lily leaves. At distances from the group of buffaloes less than 100 m., the radius and the area of leaves were bigger than 100 m. and further. The distance from the buffalo can be explained by the size of water leaves and negative relationship between all. Biomass affects the growth of aquatic plants. Just like the results of the past, biomass allocation in response to water depth. Plants in 30 cm. water produced more but smaller and shorter-lived leaves than plants at 60 cm. and 90 cm. water levels [24]. After one month, seedlings in 90 cm. water had less biomass than those in 30 cm. [25]. Commercial expansion has affected the natural stocks of this plant, in turn, affecting the nursery function of these habitats [26]. So, to solve the water lily problems were not to focus on it was the food of the buffaloes [27] or feces was the cause of waste water. Other factors, such as household waste water discharge or to collect water lily for trade, should be considered.

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