

RESEARCH ARTICLE

Inter-Annual Patterns of Aggression and Pair Bonding in Captive American Flamingos (*Phoenicopterus ruber*)

Nathan B. Frumkin,¹ Tina W. Wey,² Megan Exnicios,¹ Caroline Benham,¹ Mitchell G. Hinton,³ Samantha Lantz,¹ Carolyn Atherton,⁴ Debbie Forde,⁴ and Jordan Karubian^{1*}

¹Department of Ecology and Evolutionary Biology, Tulane University, New Orleans, Louisiana

²Department of Biology, New Mexico State University, Las Cruces, New Mexico

³Department of Wildlife, Fish and Conservation Biology, University of California at Davis, Davis, California

⁴Audubon Zoo, Audubon Nature Institute, New Orleans, Louisiana

Because zoos typically house animals for extended periods of time, longitudinal studies can play an important role in evaluating and optimizing animal care and management. For example, information on patterns of aggression and mating behavior across years can be used to monitor well-being, assess response to changes to group composition, and promote successful reproduction. Here, we report on patterns of aggression and pair bonding by American flamingos (*Phoenicopterus ruber*) at the Audubon Zoo, New Orleans USA across 4 years (2012–2015), a period that included a simultaneous introduction and removal of individuals in 2014. At the population level, overall rates and social network indices of aggressive interactions were relatively stable over the study period, without a strong signal of the 2014 replacement event. At the individual level, flamingos exhibited a high degree of within-individual consistency in levels of aggression initiated ($W = 0.530$, $P < 0.001$), and received ($W = 0.369$, $P = 0.042$). In terms of pair bonds, females re-paired with the same individuals across years more frequently (between 58% and 100% from year to year) than they switched mates, and no bonds were established between pre-existing and introduced individuals. These findings indicate a high degree of stability in aggression and pair bonding behavior in this population of captive flamingos, at both the population and individual level. Longitudinal studies such as this one provide an opportunity to better our understanding of flamingos and other long-lived, group-living animals along with their management needs, especially in terms of maintaining social cohesion in captivity and improving captive breeding programs. Zoo Biol. 35:111–119, 2016. © 2016 Wiley Periodicals, Inc.

Keywords: captive animals; personality; social networks; UCINET; zoo management

INTRODUCTION

A central mission of zoos is to ensure the well-being of captive animals [Van Dyke, 2010; Hosey et al., 2013]. Aggression is a commonly used index of well-being among captive animals. Though some minimum level of aggression is necessary to acquire breeding opportunities or maintain territories [Duckworth, 2006; Alcock, 2009], high levels of aggression can have negative consequences on the physical and psychological health of captive animals and may be indicative of improper enclosure environments, social instability, or chronic stress [Carlstead and Shepherdson, 1994, 2000; Waples and Gales, 2002; McDougall et al., 2006; Morgan and Tromborg, 2007]. Breeding may also reflect well-being of captive animals because lack of

breeding success could be related to stress levels, the social environment, and enclosure design [Carlstead and Shepherdson, 1994; Waples and Gales, 2002]. Observational studies across multiple years (i.e., longitudinal studies) can

*Correspondence to: Jordan Karubian, Department of Ecology and Evolutionary Biology, Tulane University, 400 Lindy Boggs Center, New Orleans, LA 70118. E-mail: jk@tulane.edu

Received 11 August 2015; Revised 24 January 2016; Accepted 04 February 2016

DOI: 10.1002/zoo.21274

Published online 16 February 2016 in Wiley Online Library (wileyonlinelibrary.com).

play an important role in establishing baseline patterns of aggression and breeding behavior needed to effectively monitor well-being in long-lived organisms [Dawkins, 2004; Melfi, 2009; Watters et al., 2009]. For example, longitudinal monitoring can capture subtle, or not so subtle, changes in behavior [Watters et al., 2009] in the context of changes to group composition via the addition and/or removal of individuals.

In some species, changes to group composition can lead to changes in dominance hierarchies that in turn lead to increases in overall aggression (e.g., as some individuals attempt to achieve higher status) and less integration among group members [Monnin and Peeters, 1999; Cleveland et al., 2003; Flack et al., 2006; Maruska and Fernald, 2010]. Changes in group composition can also alter breeding dynamics; indeed, introductions are often used as a strategy to induce breeding among non-reproducing captive animals [Stevens and Pickett, 1994; Farrell et al., 2000]. The impact of an introduction or replacement event can only be quantitatively assessed when pre-introduction data are available for comparison [Watters et al., 2009], further underscoring the importance of longitudinal studies.

Measures of social stability and behavioral consistency relevant to aggression and breeding can occur at several levels: the individual, small groups or pairs, or the whole population. Individuals can demonstrate consistency in behaviors across contexts and/or time, and these behaviors can be correlated; this is often referred to as animal personality or behavioral syndromes [Sih et al., 2004; Krause et al., 2010; Réale et al., 2010; Wilson et al., 2012]. Social relationships between two or more individuals can be measured by tracking pair bonds and small groups through time [Leu et al., 2010; Godfrey et al., 2012]. At the population-level, overall social structure can be quantified through social network analysis [Wey et al., 2008; Krause et al., 2014] or by measuring behaviors at the population level (rates, etc.). Social network analysis provides useful tools for understanding the behavior of captive animals at each of these levels because it allows for direct measurements of social interactions and relationships that can be used to gain insights into social stability and consistency [Wey et al., 2008; Hinton et al., 2013; Krause et al., 2014; Aplin et al., 2015]. However, only a few studies have looked at repeatability and consistency in network traits [Blumstein et al., 2013; Brent et al., 2013; Jacoby et al., 2014; Aplin et al., 2015].

Because they are highly gregarious and among the most common birds in zoos, flamingos are well suited for monitoring how aggression and social relationships change over time, with a focus on social stability and behavioral consistency [Rose et al., 2014]. Relatively high levels of aggressive interactions have been documented for captive flamingos at the group [Perdue et al., 2011; Hinton et al., 2013; Hughes and Driscoll, 2014] and individual levels [Hughes, 2015]. Hughes [2015] assessed dominance over multiple years, finding that dominance interactions were

stable across seasons; however, little other information exists on inter-annual variation in other aspects of flamingo behavior [Rose et al., 2014]. Although pair bonds among wild flamingos do not typically persist beyond one breeding season and the mate-switching rate between seasons can be greater than 98% [Cezilly and Johnson, 1995; Cezilly et al., 1997], captive populations exhibit relatively high mate fidelity potentially due to smaller population sizes [Pickering, 1992; Farrell et al., 2000; Shannon, 2000; Studer-Thiersch, 2000]. The relative lack of information on long-term behavioral patterns in flamingos have led Rose et al. [2014] and others to call for further study of captive flamingo behavior to enhance management techniques and improve captive flamingo welfare.

Here, we report on patterns of aggression and breeding behavior among captive American flamingos *Phoenicopterus ruber* at the Audubon Zoo in New Orleans, LA, across 4 years (2012–2015), a period that included an introduction and removal (hereafter “replacement”) event in 2014. At the population-level, we predicted that patterns of aggression (as measured by social network structure and rate data) would be relatively consistent across years, but that the replacement event would be associated with a detectable increase in aggression as social instability increases post-introduction. At the individual-level, we similarly predicted that individuals would be consistent in their levels of aggression across years, but that the year of the replacement event would be an outlier as overall aggressions spikes due to instability. Given previous reports of high mate fidelity [Pickering, 1992; Shannon, 2000; Studer-Thiersch, 2000], we predicted high pair bond persistence overall, but considered it likely that the replacement event might lead to new pair bonds and an increase in overall breeding activity [Stevens and Pickett, 1994; Farrell et al., 2000]. Our findings supported the idea that aggression and pair bonding dynamics were highly stable over time, both within and among individuals, but did not support any detectable signal of the replacement event on aggression, pair bonding, or breeding activity.

METHODS

Study Species and Study Site

We studied a group of individually marked American flamingos (*Phoenicopterus ruber*) at the Audubon Zoo in New Orleans, LA between 2012 and 2015. The flamingo exhibit is located at the central entrance of the zoo and contains a main area of sand surrounded by a moat and a central pool where the flamingos typically feed. Between 2012 and 2015, group size fluctuated only slightly (34–37 individuals, consisting of 15–17 males and 18–20 females). However, in February 2014, eight new individuals (five males, three females) were added and five existing individuals (three males, two females) removed. This replacement was motivated in part to promote formation of new pairs and promote breeding, as there had been no

successful breeding attempts associated with this exhibit since before the project began. The new individuals were introduced to the exhibit from a secondary flamingo exhibit located at the rear of the zoo with approximately twice the population size; individuals from this exhibit also are not known to have bred in recent years. The capture of the flamingos for the transfer was done opportunistically, although certain individuals were kept in the front exhibit for management reasons (e.g., easier access by keepers for veterinary assistance). Age and sex of all individuals were obtained from Audubon Zoo records.

Data Collection

We observed flamingo interactions in the late winter and spring (Feb–May) each year from 2012 to 2015. We followed the same data collection protocol established in Hinton et al. [2013], using a standardized ethogram found therein. Briefly, we used a combination of focal-animal sampling and opportunistic observations to record aggressive interactions [Altmann, 1974]. We randomly selected a focal individual to observe for a 5-min period during which we recorded its behavior each minute. In this time, we recorded all aggressive interactions involving the focal as well as any extra-focal aggressive interactions that we observed. Due to a small population size and small enclosure size, most extra-focal interactions could be observed regardless of their proximity to the focal individual. Aggressive interactions included pecks, stand offs (two or more individuals exchanged vocalizations and waved their heads in a display), and chases [Hinton et al., 2013]. In that 5-min period, we also opportunistically recorded the number of courtship displays that occurred in the group. Since close proximity is one of the ways to identify pair bonds, especially after courtship has ended [Shannon, 2000; Studer-Thiersch, 2000], we recorded a potential pair bond if the focal individual spent the majority of the 5-min observation period <0.5 m from another individual. Other behaviors used to identify pair bond behavior were paired walking (often associated with foraging) where two individuals would walk

in close proximity sometimes with one following the other, paired aggression where two individuals would exhibit aggression towards a third individual or another pair, and paired defense of a nest mound [Shannon, 2000; Studer-Thiersch, 2000; Perdue et al., 2011]. When we observed a potential pair bond for two or more observation days, we listed it as a confirmed pair bond in order to ensure that pair bonds were established across multiple days.

Statistical Analyses

We used UCINET software [Borgatti et al., 2002] to construct and analyze social networks derived from the aggression data acquired from 2012 to 2015. In order to compare 4 years’ worth of data that varied in data collection efforts, we analyzed data from the same 3-week time period (Feb 19–Mar 13) each year and used it to construct yearly social networks (one per year from 2012 to 2015). We selected this period, which corresponded to the “courtship” phase [as defined in Hinton et al., 2013] across all 4 years, to control for any potential sources of variation associated with different stages of breeding. We constructed social networks using all aggressive interactions for which both initiator and recipient were identified. We calculated individual level (i.e., degree centrality, the number of connections an individual has) and population level (i.e., degree centralization, a measure of variance in the number of aggressive connections among individuals) social network metrics for each of these four networks (see Table 1 for definitions and application of these metrics). To determine if there were similarities in network structure across years, we used a multiple regression quadratic assignment procedure (MRQAP) in UCINET [Borgatti et al., 2002] to determine if there were significant correlations between pairs of networks (years) containing the 27 individuals present for the entire study, while controlling for sex by restricting permutations within sex. We estimated significance of network measures by permutations because network data violates assumptions about the independence of dependent variables [Hanneman and Riddle, 2005; Croft et al., 2011].

TABLE 1. Social network metrics used in this study of aggression in captive American flamingos at the Audubon Zoo, New Orleans LA (adapted from Table 1 in Hinton et al., 2013 which drew its definitions from Wey et al., 2008 and Hanneman and Riddle, 2005)

Metric	Definition	Application
Individual-level metrics		
Out-degree centrality	Number of ties originating from an individual	Metric used to determine which individuals initiate aggressive interactions
In-degree centrality	Number of ties directed towards an individual	Metric used to determine which individuals receive aggression
Population-level metrics		
Degree Centralization	An index of variance in the number of ties among individuals	Measure of how evenly among individuals aggressive interactions are initiated and received (lower measures indicate more equal distribution; higher values indicate similarity to a star-network with one or few especially well-connected individuals)

Mean outdegree is a useful measure of overall aggression initiated (see Table 1), but differences in sampling across years complicate inter-annual comparisons, so instead we used measures of the rate of aggression to identify changes over time. We used a one-way ANOVA to compare mean rates of aggression (number of interactions per observation session) of the group across years. For this measure of rate, we only compared rates from 2013 to 2015 because the first 2 weeks of 2012 had observation sessions of a different length, and extra-focal interactions were not recorded. We also measured rate of aggression of the group across all four years as the number of recorded interactions from focal observations per minute of observation and used a Kruskal–Wallis test to compare them. To determine which years specifically had higher rates than other years, we used a mixture of Mann–Whitney and Kruskal–Wallis tests. We used R (Version 3.0.1) to conduct ANOVAs, Mann–Whitney tests, and Kruskal–Wallis tests [R Development Core Team, 2013].

We used an E–I Index permutation test in UCINET to determine if the individuals already present in the exhibit formed a distinct “group” relative to introduced individuals, as measured by directed aggression. To do so, we divided these individuals into two groups (i.e., pre-existing vs. introduced) and tested for non-random patterns of aggression either within or among groups. The E–I (External–Internal) Index ranges from -1 (all ties internal, meaning aggression was directed entirely within their group) to $+1$ (all ties external, meaning aggression was directed entirely outside their group) [Hanneman and Riddle, 2005]. However, the range of possible values sometimes can be restricted by the group sizes and density, so UCINET provides a rescaled index value that ranges between -1 and $+1$ [Hanneman and Riddle, 2005]. The E–I Index is calculated for the entire population and for individuals, but the permutation test is conducted at the population-level and determines if a given E–I Index value is significantly different from random mixing [Hanneman and Riddle, 2005]. Since previous E–I Index analyses did not find effects of sex [Hinton et al., 2013; Frumkin et al., unpublished data], we did not include this factor in the current analysis.

To determine consistency in outdegree and indegree across years at the individual-level, we adapted a permutation analysis used by Jacoby et al. [2014; see also Wilson et al., 2012] including only individuals that were present all 4 years. Using R [R Development Core Team, 2013], we first calculated Kendall’s coefficient of concordance (W) to analyze consistency of ranks across years in the original networks. We analyzed relative ranks rather than actual values of outdegree and indegree because these values are likely to be influenced by social dynamics beyond the individual, so rankings allowed us to measure consistency in relative aggression without making assumptions about what generated absolute levels of aggression. To assess statistical significance, we then compared the real values of W to a frequency distribution of W values generated from 20,000

node permutations, where the P -value was the proportion of W values from permutations to which the W of the original network was equal or more extreme. Data permutations are preferred for assessing statistical significance of factors influencing network measures because the network measures are non-independent, violating assumptions of many standard statistical tests (such as linear regression), and thus P -values inferred from standard tests are often anti-conservative [Croft et al., 2011]. Because males and females differ in their level of aggression in this system, we used restricted node permutations, where values were only randomized among individuals of the same sex, implemented with the “permute” package [Simpson, 2015]. We also considered that pair bonds could influence patterns of individual consistency in aggression, so we reran the analyses on the subset of data excluding within-pair aggression. However, as there were very few within-pair aggressive interactions and the results were virtually identical, we only report results from analyses of the full dataset.

To assess year to year changes in male–female pair-bond formation and persistence, we compared the proportions of paired females and unpaired females from year to year using a chi-squared test. We also compared the proportions of females that retained and switched their pair bond from year to year using a Fisher’s exact test in R to determine if patterns of pair bond persistence and switching significantly varied across years.

RESULTS

Population-Level Aggression

We observed relatively little inter-annual variation in population-level patterns of aggression across the 4-year study period. Although annual rates of aggression in the group differed across years over the entire study ($H = 9.837$, d.f. = 3, $P = 0.020$, Fig. 1a), the only statistically significant pairwise difference between years was 2012 versus 2015 (Mann–Whitney U -test, $U = 49$, $P = 0.004$); there were no significant differences between the other years (2012–2014: $H = 4.094$, d.f. = 2, P -value = 0.129; 2013–2015: $H = 3.637$, d.f. = 2, P -value = 0.162). Outdegree (aggression initiated) networks were significantly centralized all 4 years indicating one or a few individuals were initiating a majority of the aggression (Table 2). Indegree (aggression received) networks were more centralized following the introduction indicating one or a few individuals became more targeted after the introduction (Table 2). Overall, there was moderate change in network structure across years for the individuals present the entire study, with three sets of networks (2013/2014, 2012/2015, 2013/2015) showing a lack of significant similarity in structure based on the multiple regression quadratic assignment procedure (MRQAP) correlation (Table 3). Following the replacement, there was no difference in overall rates of aggression in the

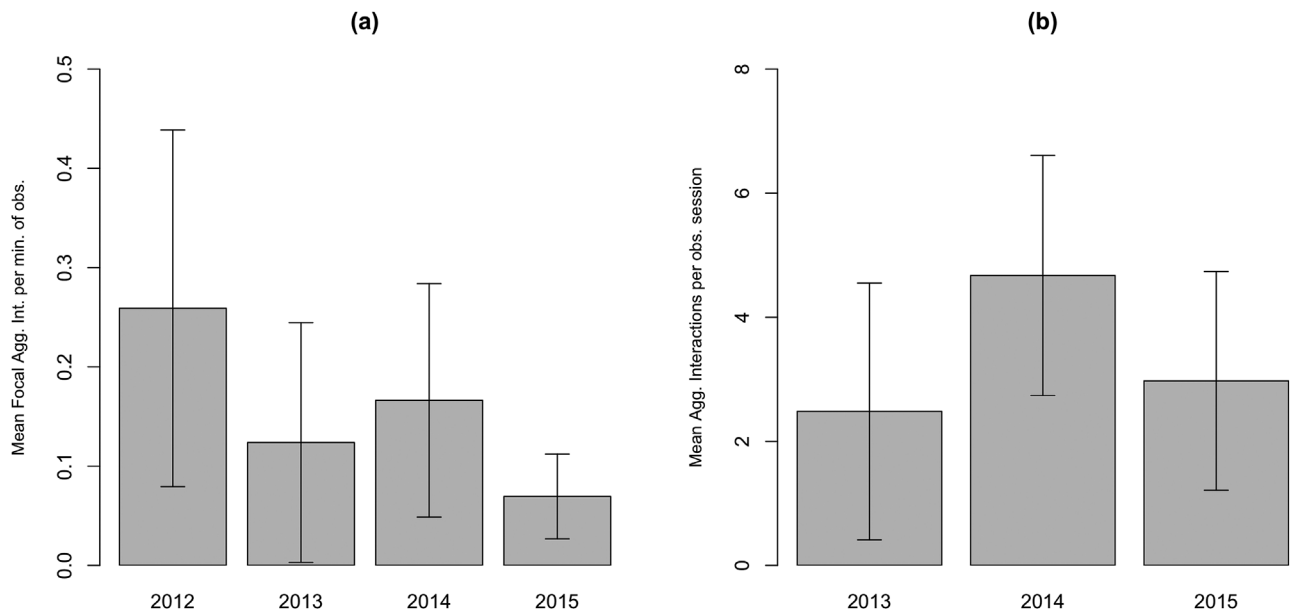


Fig. 1. Population-level patterns of aggression among captive American flamingos at Audubon Zoo, New Orleans, LA. Panel (a) depicts mean rates of focal aggression (\pm standard deviation) measured at the level of the group as the number of recorded aggressive interactions from focal observations per minute of observation. The rate in 2012 was significantly higher than 2015 (Mann–Whitney U -test, $Z = 2.88$, $P = 0.004$), but the other rates did not significantly differ. Panel (b) depicts mean rates of overall aggression (\pm standard deviation) measured at the level of the group as the number of aggressive interactions per observation session. There was no significant difference across years ($F = 1.525$, d.f. = 2, $P = 0.275$).

group ($F = 1.525$, d.f. = 2, $P = 0.275$; Fig. 1b) and no evidence that aggression was non-randomly clustered among already-present versus introduced individuals (E–I Index—2014: -0.153 , 2015: -0.279 ; Rescaled E–I Index—2014: -0.153 , 2015: -0.214 ; E–I Index Test—2014: $P = 0.192$, 2015: $P = 0.472$).

Individual-Level Aggression

Substantial variation existed between individuals in terms of aggressive encounters initiated (i.e., outdegree), with some individuals consistently exhibiting higher levels of aggression than others (Fig. 2a, $W = 0.530$, $P < 0.001$). Receipt of aggression (i.e., indegree) was less differentiated among individuals (Fig. 2b), but there was significant consistency within individuals for indegree across years

based on restricted permutations controlling for sex ($W = 0.369$, $P = 0.042$).

Pair-Bonding and Breeding

The proportion of pair-bonded females in the study population did not significantly vary across years (Fig. 3a; $\chi^2 = 2.486$, d.f. = 3, $P = 0.478$). Females retained the same male partner from previous years more often than they switched to a different male, with the highest rates of pair bond switching (different males) in the year preceding the replacement (Fig. 3b). Though there was some increase over time (Fig. 3b), the proportion of females retaining their partners did not significantly vary across years (Fisher's Exact Test— $P = 0.102$). Following the replacement, no pair bonds were formed between already-present and introduced

TABLE 2. Population Characteristics and Degree Centralization of the study population of American flamingos at Audubon Zoo, New Orleans LA across the years (2012–2015)

Year	2012	2013	2014	2015
Population Size	34	35	37	36
Males	16	16	17	16
Females	18	19	20	20
Sex Ratio (F:M)	1.125	1.188	1.176	1.250
# interactions	75	183	241	434
Degree Centralization	43.07%	53.81%	35.65%	70.37%
Out Degree	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$
Degree Centralization	14.97%	23.53%	32.79%	61.55%
In Degree	$P = 0.1521$	$P = 0.0267$	$P = 0.0003$	$P < 0.0001$

2012 only includes focal interactions during our designated time period (Feb 19–Mar 13) because extra-focal interactions were not recorded until March 16, 2012.

TABLE 3. Measures of the overall similarity of social network structures for American flamingos in Audubon Zoo, New Orleans LA across the 4-year study period using multiple regression quadratic assignment procedure (MRQAP) correlation controlling for sex by restricting permutations within sex

A	2012	2013	2014	2015
2012	1.000	0.145	0.115	0.058
2013	–	1.000	–0.016	0.012
2014	–	–	1.000	0.182
2015	–	–	–	1.000

B	2012	2013	2014	2015
2012	0	0.006	0.022	0.125
2013	–	0	0.683	0.744
2014	–	–	0	0.002
2015	–	–	–	0

A significant correlation ($P < 0.05$) indicates that the network structures of the 2 years being compared are similar. Shown are (A) MRQAP Correlations (B) MRQAP Significance between years (2012–2015) (2-tailed P -values testing the hypothesis that correlations are higher than by chance alone).

individuals. Overall, there was no successful breeding recorded during the course of the study. The presence of eggs was only noted in 1 year (2013) when two eggs of unknown fertility were laid but were not incubated.

DISCUSSION

Aggression

Our study revealed relatively little variation in population-level aggression by captive flamingos across years, even in the context of a replacement. The lack of major changes in rates of aggression is consistent with our prediction that population-level aggression would remain relatively stable over time. However, counter to a second prediction, we did not see significant population-level changes in response to a replacement event. Outdegree networks were significantly centralized all 4 years, suggesting that relatively few individuals were initiating most of the aggression. Indegree networks became more centralized over time, suggesting that some individuals became increasingly

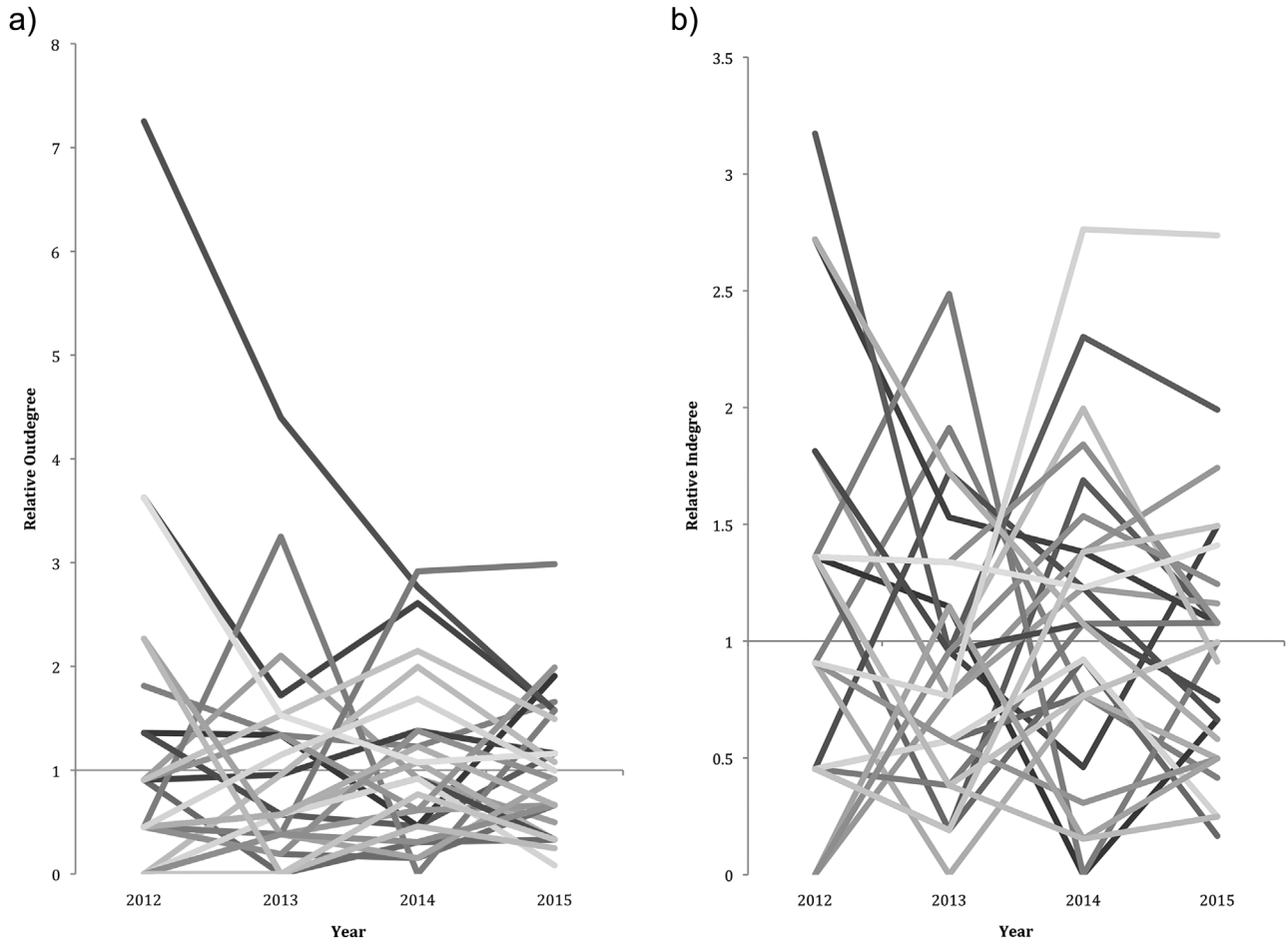


Fig. 2. Individual captive American flamingos were significantly consistent across years (2012–2015) in terms of aggression initiated (i.e., outdegree) and aggression received (i.e., indegree). (a) Individual flamingos (represented in the figure by individual lines) exhibited marked differences in annual standardized individual outdegree (individual outdegree divided by mean outdegree), and these differences were consistent over time. (b) Standardized individual indegree was less differentiated among individuals, but there was still significant individual-level consistency across years.

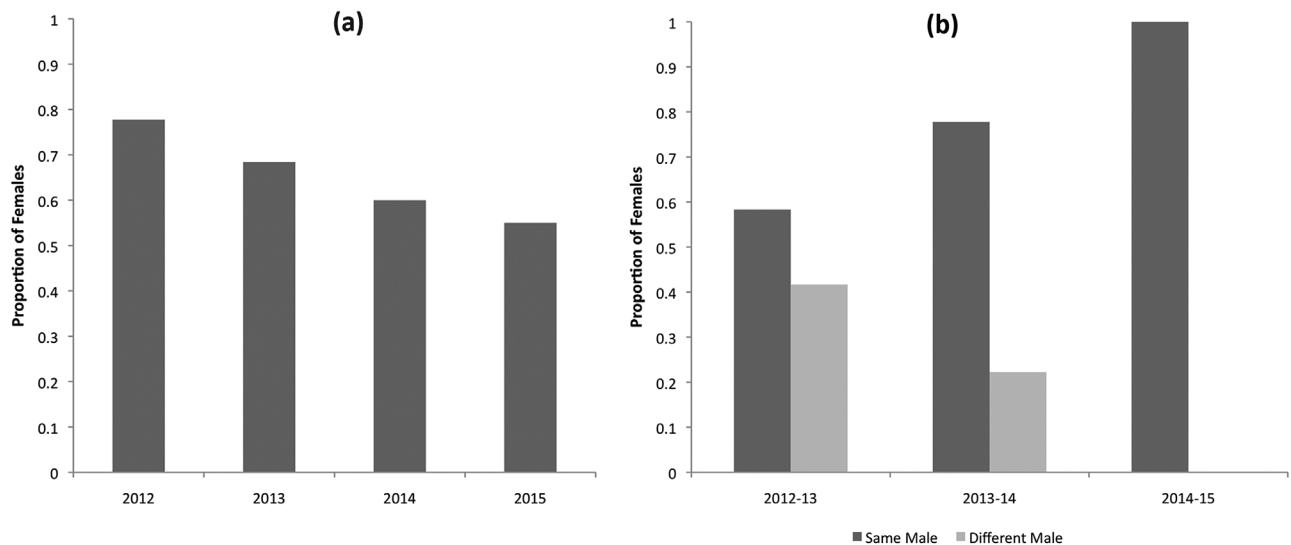


Fig. 3. Patterns of pair-bonding in a group of captive American flamingos at Audubon Zoo, New Orleans, LA. Panel (a) depicts the proportion of American flamingo females that stayed in a male–female pair bond across years (2012–2015) which did not significantly vary. Panel (b) depicts the proportion that switched pair bonds versus kept the same bonds. Females retained pair bonds more frequently than they switched. The proportion of females that retained pair bonds appears to increase over time, but this variation across years was not significant.

targeted. Besides 2013 and 2014, the only years in which network structures were not significantly correlated were non-consecutive years (2012/2015 and 2013/2015), suggesting that there was little year-to-year change in network structure (between those individuals present all years). Instead, differences in structure across non-consecutive years may occur as subtle changes that accumulate over time. However, there does not appear to be any directionality in the nature of these changes, as evidenced by the correlation between network structure in 2 non-consecutive years, 2012 and 2014. These results suggest that aggression in the population was relatively consistent across consecutive years, in support of our prediction of consistency over time.

Our prediction that individual-level aggression would be consistent across years was also well supported, as individuals exhibited consistent levels of both initiated and received aggression across years. Many animals exhibit the consistency and repeatability of behaviors indicative of personalities [Sih et al., 2004; van Oers et al., 2004; Réale et al., 2007; Conrad et al., 2011; Jacoby et al., 2014]. The existence and patterns of social personality types, in particular, is an area of recent interest, and our study adds to a small but growing literature considering the between-individual consistency of social network position in animals [Wilson et al., 2012; Blumstein et al., 2013; Jacoby et al., 2014; Aplin et al., 2015]. Also, to our knowledge there has been only one other study of personality in flamingos prior to the current study: McCully et al. [2014] assigned personality scores based on aggression and boldness and suggest that American flamingos associate with individuals of similar personality while Chilean flamingos (*P. chilensis*) do not associate by personality. Also, Hughes [2015] found that individual dominance status was stable across years and

seasons. We [along with Hughes, 2015 and McCully et al., 2014] have found a clear pattern of individual consistency in individual flamingo behaviors and suggest that individual flamingos exhibit aggressive personalities. This has management implications in the context of captive breeding programs because some personality traits may relate to breeding success and a population's variation in personality may change over time in captivity, though more research is needed to understand the impacts of captivity on personality structure and consistency [Dingemanse and Réale, 2005; McDougall et al., 2006; Tetley and O'Hara, 2012]. Understanding individual personalities also enables managers to key in on individual tendencies and make attempts to maintain group cohesion by preventing incompatible individuals from being housed together, thereby avoiding chronic stress [Tetley and O'Hara, 2012]. Though, since the American flamingo's patterns of association are related to their personality [McCully et al., 2014] and research on American flamingo group organization is limited, managers may find maintaining cohesion in their groups more difficult than other species in which the personalities are known.

Contrary to our predictions, our findings suggest that a replacement event (introduction of eight new individuals and concurrent removal of five individuals) in 2014 had little impact on patterns of aggression. There was no significant increase in the rate of aggression in 2014 following the replacement event, and comparisons of network structure and outdegree centralization across years showed little signal of the replacement event. The results also suggest that the replacement did not affect social interactions because original individuals did not significantly direct aggression towards introduced individuals, nor did introduced individuals direct more aggression towards original individuals. The

network structure of 2014 was not significantly correlated with 2013, which is consistent with a change associated with the replacement event. However, 2014 network structure is also significantly correlated with 2012, making the trend less clear. The fact that social structure in both 2012 and 2013 were not significantly correlated with 2015 could suggest a delayed signature of the replacement event. Because there were significant correlations between consecutive years, there could be carryover across years in subtle changes to social structure [Sih et al., 2009]. Thus, even though social interactions in the year of the replacement (2014) were not distinguishable from 2012 or 2015, changes to social interactions that carried over from 2014 may have been slightly different due to the replacement event resulting in a delayed effect on network structure, indicated by a different structure in 2015 relative to 2012/2013. However, this pattern may also be a result of accumulated changes as was mentioned earlier. Also, some individuals did become more targeted after 2014 according to indegree centralization, which could conceivably be attributable to the replacement event. However, though there were some small changes after replacement, we conclude that the available data do not provide any indication that the replacement event had a major impact on aggression.

Pair Bonding

We found support for our prediction that individuals are more likely to retain pair bonds from the previous year instead of forming new ones. This result is consistent with previous research that has found that mate fidelity is high in captive populations, in strong contrast to high rates of mate switching seen in the wild [Cezilly and Johnson, 1995; Cezilly et al., 1997; Shannon, 2000]. This consistency in pair identities could be due to the fact that zoo populations have small, closed populations that limit the ability to form new pair bonds [Studer-Thiersch, 2000]. Our results also show that patterns of pair bonding did not radically vary across years suggesting that flamingos can display a high-degree of consistency in multiple patterns of behavior, aggression, and pair-bonding.

The introduction of new individuals at this zoo took place in part to promote breeding within this group, which had not successfully produced young in several years. However, no pair bonds included new individuals after the replacement, and no breeding occurred in either 2014 or 2015. There is evidence in other flamingo populations that the addition of new individuals can promote breeding success [Stevens and Pickett, 1994] sometimes in the subsequent year [Farrell et al., 2000]. This suggests that there is still an opportunity for the replacement event to be successful in the future. However, high pair bond persistence likely limits the effect that the new individuals could have on changes in breeding behavior. Despite the high levels of repetition, the presence of new individuals could stimulate breeding in an alternative pathway to pairing such as extra-pair copulations which

have been documented in both captive and wild flamingos [Shannon, 2000; Studer-Thiersch, 2000]. Since this population at Audubon Zoo has bred before even with some individuals exhibiting a relatively high degree of mate fidelity [Shannon, 2000], it is difficult to directly connect the high levels of repeated pair bonds to this lack of breeding. It is likely that other factors (such as age or colony size) are also playing a role in this population's breeding dynamics [Rose et al., 2014], and since it appears that flamingos form preferential relationships, more detailed research will be needed to understand long-term relationships between individuals [Rose and Croft, 2015].

CONCLUSION

This study has shown that a captive group of American flamingos exhibit high levels of consistency in behavior (aggression and pair bond formation) at both a population-level and individual-level. This individual consistency in levels of aggression could very well be related to dominance in these birds with hyper-aggressive individuals being more dominant. Other studies have found evidence of a semi-linear or non-linear dominance hierarchy in American flamingos [Hughes and Driscoll, 2014; Royer and Anderson, 2014], so it would be interesting to investigate dominance in the Audubon population in order to better grasp how individuals fit within the group and the role of their individual levels of aggression. Also, we have only used aggressive interactions to construct social networks, but other social data could be used to construct social networks like social proximity [as shown by McCully et al., 2014]. In the future, it would be worthwhile to consider these other aspects of flamingo sociality along with other recently described social behaviors [see Rose and Croft, 2015] to gain a better grasp of flamingo social organization. Continued study of individual and group stability in flamingos and other long-lived, group-living animals, is needed to refine management and care, as well as to gain a better insight into broad patterns of social behavior in captive animals relative to their free-living counterparts.

ACKNOWLEDGMENTS

We would like to thank the two anonymous reviewers who provided helpful feedback on an earlier version of this article. We would like to thank the Audubon Zoo and the Audubon Nature Institute for allowing us to conduct this research, the Tulane students that have assisted with data collection, and Erik Enbody and the students of Tulane's Experimental Animal Behavior course (EBIO 3690) for their feedback throughout the process of this study.

REFERENCES

- Alcock J. 2009. *Animal behavior*. 9th edition. Sunderland, MA: Sinauer Associates, Inc. p 606.
- Altmann J. 1974. Observational study of behavior: sampling methods. *Behav* 49:227–266.

- Aplin LM, Firth JA, Farine DR, et al. 2015. Consistent individual differences in the social phenotypes of wild great tits, *Parus major*. *Anim Behav* 108:117–127.
- Blumstein DT, Petelle MB, Wey TW. 2013. Defensive and social aggression: repeatable but independent. *Behav Ecol* 24:457–461.
- Borgatti SP, Everett MG, Freeman LC. 2002. Ucinet for windows: software for social network analysis. Harvard, MA: Analytic Technologies.
- Brent LNJ, Heilbronner SR, Horvath JE, et al. 2013. Genetic origins of social networks in rhesus macaques. *Sci Rep* 3:1–8.
- Carlstead K, Shepherdson D. 1994. Effects of environmental enrichment on reproduction. *Zoo Biol* 13:447–458.
- Carlstead K, Shepherdson D. 2000. Alleviating stress in zoo animals with environmental enrichment. In: Moberg GP, Mench JA, editors. *The biology of animal stress: basic principles and implications for animal welfare*. New York: CABI Publishing. p 337–354.
- Cezilly F, Boy V, Tourenq CJ, Johnson AR. 1997. Age-assortative pairing in the greater flamingo *Phoenicopterus ruber roseus*. *Ibis* 139:331–336.
- Cezilly F, Johnson AR. 1995. Re-mating between and within breeding seasons in the greater flamingo *Phoenicopterus ruber roseus*. *Ibis* 137:543–546.
- Cleveland AL, Ludlow AM, Itzkowitz M, Draud M, Haley M. 2003. Dominance relationships between male territorial neighbors in the beaugregory damselfish (*Stegastes leucostictus*). *Behav* 140:1021–1037.
- Conrad JL, Weinersmith KL, Brodin T, Saltz JB, Sih A. 2011. Behavioural syndromes in fishes: a review with implications for ecology and fisheries management. *J Fish Biol* 78:395–435.
- Croft DP, Madden JR, Franks DW, James R. 2011. Hypothesis testing in animal social networks. *Trends Ecol Evol* 26:502–507.
- Dawkins MS. 2004. Using behaviour to assess animal welfare. *Animal Welfare* 13:3–7.
- Dingemanse NJ, Réale D. 2005. Natural selection and animal personality. *Behav* 142:1165–1190.
- Duckworth RA. 2006. Behavioral correlations across breeding contexts provide a mechanism for a cost of aggression. *Behav Ecol* 17:1011–1019.
- Farrell MA, Berry E, Marples N. 2000. Breeding behavior in a flock of Chilean flamingos (*Phoenicopterus chilensis*) at Dublin Zoo. *Zoo Biol* 19:227–237.
- Flack JC, Girvan M, de Waal FBM, Krakauer DC. 2006. Policing stabilizes construction of social niches in primates. *Nature* 439:426–429.
- Godfrey SS, Bradley JK, Sih A, Bull CM. 2012. Lovers and fighters in sleepy lizard land: where do aggressive males fit in a social network? *Anim Behav* 83:209–215.
- Hanneman RA, Riddle M. 2005. Introduction to social network methods. Riverside, CA: University of California, Riverside. <http://faculty.ucr.edu/hanneman/>
- Hinton MG, Bendelow A, Lantz S, et al. 2013. Patterns of aggression among captive American flamingos (*Phoenicopterus ruber*). *Zoo Biol* 32:445–453.
- Hosey G, Melfi V, Pankhurst S. 2013. Zoo animals: behaviour, management, and welfare. 2nd edition. Oxford, UK: Oxford University Press. p 643.
- Hughes AL. 2015. Stability of social behavior in captive American flamingos (*Phoenicopterus ruber*): a quantitative case study. *Zoo Biol* 34:305–313.
- Hughes AL, Driscoll C. 2014. Being in the thick of things: context-dependent network centrality in a captive flock of American flamingos. *J Ethol* 32:83–90.
- Jacoby DMP, Fear LN, Sims DW, Croft DP. 2014. Shark personalities? Repeatability of social network traits in a widely distributed predatory fish. *Behav Ecol Sociobiol* 68:1995–2003.
- Krause J, James R, Croft DP. 2010. Personality in the context of social networks. *Phil Trans R Soc B* 365:4099–4106.
- Krause J, James R, Franks DW, Croft DP, editors. 2014. *Animal social networks*. New York: Oxford University Press. p 288.
- Leu ST, Bashford J, Kappeler PM, Bull CM. 2010. Association networks reveal social organization in the sleepy lizard. *Anim Behav* 79:217–225.
- Maruska KP, Fernald RD. 2010. Behavioral and physiological plasticity: rapid changes during social ascent in an African cichlid fish. *Horm Behav* 58:230–240.
- McCully F, Croft D, Lee R, et al. 2014. Personality as a characteristic of social position in captive flamingo flocks. Association for the Study of Animal Behaviour Winter Conference 2014; 2014 Dec 4–5; London, UK.
- McDougall PT, Réale D, Sol D, Reader SM. 2006. Wildlife conservation and animal temperament: causes and consequences of evolutionary change for captive, reintroduced, and wild populations. *Anim Conserv* 9:39–48.
- Melfi VA. 2009. There are big gaps in our knowledge, and thus approach, to zoo animal welfare: a case for evidence-based zoo animal management. *Zoo Biol* 28:574–588.
- Monnin T, Peeters C. 1999. Dominance hierarchy and reproductive conflicts among subordinates in a monogynous queenless ant. *Behav Ecol* 10:323–332.
- Morgan KN, Tromborg CT. 2007. Sources of stress in captivity. *Appl Anim Behav Sci* 102:262–302.
- Perdue BM, Gaalema DE, Martin AL, Dampier SM, Maple TL. 2011. Factors affecting aggression in a captive flock of Chilean flamingos (*Phoenicopterus chilensis*). *Zoo Biol* 30:59–64.
- Pickering S. 1992. A short report on the 1990 Slimbridge (UK) flamingo breeding season. Flamingo Research Group Newsletter 6:33–36.
- R Development Core Team. 2013. R: a language and environment for statistical computing. Vienna, Austria: R Foundation for Statistical Computing.
- Réale D, Dingemanse NJ, Kazem AJN, Wright J. 2010. Evolutionary and ecological approaches to the study of personality. *Phil Trans R Soc B* 365:3937–3946.
- Réale D, Reader SM, Sol D, McDougall PT, Dingemanse NJ. 2007. Integrating animal temperament within ecology and evolution. *Biol Rev* 82:291–318.
- Rose PE, Croft DP. 2015. Evidence of directed interactions between individuals in captive flamingo flocks. *Wildfowl* 65:121–132.
- Rose PE, Croft DP, Lee R. 2014. A review of captive flamingo (*Phoenicopteridae*) welfare: a synthesis of current knowledge and future directions. *Int Zoo Yb* 48:139–155.
- Royer EA, Anderson MJ. 2014. Evidence of a dominance hierarchy in captive Caribbean flamingos and its relation to pair bonding and physiological measures of health. *Behav Process* 105:60–70.
- Shannon PW. 2000. Social and reproductive relationships of captive Caribbean flamingos. *Waterbirds* 23:173–178.
- Sih A, Bell A, Johnson JC. 2004. Behavioral syndrome: an ecological and evolutionary overview. *Trends Ecol Evol* 19:372–379.
- Sih A, Hanser SF, McHugh KA. 2009. Social network theory: new insights and issues for behavioral ecologists. *Behav Ecol Sociobiol* 63:975–988.
- Simpson GL. 2015. Permute: functions for generating restricted permutations of data. R package version 0.8-4. <http://CRAN.R-project.org/package=permut>
- Stevens EF, Pickett C. 1994. Managing the social environments of flamingos for reproductive success. *Zoo Biol* 13:501–507.
- Studer-Thiersch A. 2000. What 19 years of observation on captive greater flamingo suggests about adaptations to breeding under irregular conditions. *Waterbirds* 23:150–159.
- Tetley CL, O'Hara SJ. 2012. Ratings of animal personality as a tool for improving the breeding, management and welfare of zoo mammals. *Anim Welfare* 21:463–476.
- Van Dyke F. 2010. Conservation biology: foundations, concepts, applications. 2nd edition. New York: Springer. p 477.
- van Oers K, Drent PJ, de Goede P, van Noordwijk AJ. 2004. Realized heritability and repeatability of risk-taking behaviour in relation to avian personalities. *Proc R Soc Lond B* 271:65–73.
- Waples KA, Gales NJ. 2002. Evaluating and minimising social stress in the care of captive bottlenose dolphins (*Tursiops aduncus*). *Zoo Biol* 21:5–26.
- Watters JV, Margulis SW, Atsalis S. 2009. Behavioral monitoring in zoos and aquariums: a tool for guiding husbandry and directed research. *Zoo Biol* 28:35–48.
- Wey T, Blumstein DT, Shen W, Jordan F. 2008. Social network analysis of animal behaviour: a promising tool for the study of sociality. *Anim Behav* 75:333–344.
- Wilson ADM, Krause S, Dingemanse NJ, Krause J. 2012. Network position: a key component in the characterization of social personality types. *Behav Ecol Sociobiol* 67:163–173.