

Five breeding attempts of male Common Kingfisher (*Alcedo atthis*) during the season consisted of pairing with two females

Päť hniezdení samca rybárika riečneho (Alcedo atthis) za sezónu pozostávalo z párenia s dvomi samicami

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Abstract. In 2018, the unique breeding effort of Common Kingfisher was documented in the Danube River system, southwestern Slovakia. Preliminary observations indicated that one breeding pair managed to breed five times of which three attempts were successful. The aim of the present work was to determine whether the observed breeding pair exhibiting remarkable reproductive parameters did not undergo extra-pair interactions or partner exchange during a long breeding season using molecular techniques. Our results show, that male bred not with one, but with two females in sequence during the season. Male found a new mate during the incubation of the third brood. Due to the relatively short distance between nesting burrows and raising many offspring from both broods, we assume that the possibility of simultaneous bigamy is relatively low. Since the females very rarely leave males during breeding, we suggest that exchange of the female partner was due to a loss of the partner rather than a consequence of an abandonment by the female.

Key words: Common Kingfisher, multiple broods, partner exchange, polygamy

Based on the amount of behavioral work, monogamy with a strong social bond between the partners and a high level of parental care has long been considered the most common bird breeding system (Lack 1968). By using molecular genetic techniques, it has been shown that only about 14% of species are truly monogamous. Species breeding once a year exhibit often mixed paternity (Griffith et al. 2002, Westneat and Stewart 2003). Species laying multiple broods can form pair bonds for the entire breeding season (Haftorn 1978, Wallander and Andersson 2003, Stepniowski & Halupka 2018) or switch the partners between breeding attempts (Emlen & Oring 1977, Warriner et al. 1986, Pogány et al. 2008, Jamieson 2011).

We study breeding strategy in the Common Kingfisher (*Alcedo atthis*), which is considered

to be socially monogamous within the season, rarely between seasons. Social polygyny or polyandry is a minor strategy, but it occurs occasionally in the population (Cramp 1992, Čech 2006, 2007, 2009, 2017, Libois 2018). No one has genetically confirmed it yet. Kingfishers breed from March to September and lay multiple broods (up to four), which usually overlap (Morgan & Glue 1977; Cramp 1992; Novotný 1994, Woodall 2001, Čech 2006, 2010, Turčoková et al. 2016, Rubáčová et al. 2020). During the overlapping, the female leaves the young at an early age and establishes another clutch, while the male remains and takes care of the offspring, but often the female helps (Cramp 1992, Čech 2006).

In 2018, one of the longest breeding seasons of the Common Kingfisher in Central Europe

was documented in the Danube River system. On the basis of the identification of individuals at the beginning of the season and their subsequent observation, it was assumed that one breeding pair managed to breed five times. The first two breeding attempts were unsuccessful and in the remaining three attempts parents managed to successfully raise a total of 19 young (Rubáčová & Melišková 2020).

The aim of this work was to determine whether the observed breeding pair exhibiting potentially five breeding attempts did not undergo extra-pair interactions or partner exchange during a long breeding season using molecular techniques.

From March to September in 2018, the breeding of a kingfisher pair was checked at the locality of Fodráska (southwestern Slovakia). The breeding burrows were located in the banks of artificially created water channel connecting the blind protrusions of the Baka's branch of Danube River (47°52'20.40"N, 17°31'13.01"E). The distance between burrows was 170 meters. Adults were mist-netted in the vicinity of their nesting burrow. Offspring were gently extracted from the nesting chamber at the age of at least 14 days using a bent iron wire. Parents and their offspring were ringed and blood sampled before release. DNA for parentage assignment was extracted by extraction kits (E.Z.N.A.[®] Tissue DNA Kit). Forward primers were fluorescently labelled, and multiplex PCR kit (QIAGEN Multiplex PCR Plus kit) was used to amplify four microsatellite loci. PCR amplifications were performed in 10 µl final volume, containing 1 µl of DNA, 0.1 µl of primer amplifying locus AACC-106, 0.3 µl of primer amplifying locus Be 2.46, 0.2 µl of primer amplifying locus BB 111, 0.2 µl of primer amplifying locus CAM 17, 5 µl of Master Mix, 1 µl Q solution a 2.2 µl of nuclease free water. Fragmentary analysis was carried out commercially in the Comenius University Science Park (Bratislava, Slovakia). Results of fragmented analyses were visualised by software Gene Marker. Alleles of putative parents were compared to those of nestlings to determine if or not they were offspring within the pair. Nestlings failed to be considered

as within-pair if their alleles mismatched with those of their parent at least in one loci.

Our preliminary observations (Rubáčová & Melišková 2020) were not confirmed by subsequent molecular analyzes. However, molecular data revealed that the male M12059 bred for an extremely long time (a total of 185 days), he established five broods, raised 19 young, but he did not breed with only one, but with two females. Moreover, the male acquired a new mate during the incubation period of the third breeding attempt (see table 1 in Rubáčová & Melišková 2020). One male breeding with multiple females was documented in other kingfisher populations several times (Čech 2007, 2009, 2017, Libois 2018). However, the relatively short distance (170 m) between the two nesting burrows together with raising many offspring from both broods indicates that the possibility of simultaneous bigamy is relatively low. It is known that birds are unlikely to respect each other without any aggressive attacks leading to the interruption of breeding or the rearing of a lower number of young for a distance shorter than 200 m (Čech 2009). The distance between burrows of two females mating with the same male is usually several hundred meters to kilometers (Čech 2017, Libois 2018). Although there is evidence of one case of simultaneous bigamy, where the distance between burrows was 150 meters (Čech 2017), which does not completely rule out this possibility, but a partner exchange is more likely. Further, it is not clear whether the female really left, or the male lost his mate in some way (e.g. by predation). Naturally, there is evidence of occasions when a female leaving a male from the first breeding and finding a new partner for the second breeding (Čech 2006, 2009, 2017, Libois 2018). Even though, it appeared in the year with low population density, after the harsh winter in 2017, during which two-third of the kingfisher population disappeared (Rubáčová et al. 2021). It is similar to case of the Piping Plover (*Charadrius melodus*), where in normally monogamous pairs breeding once a year became double breeding with the mate exchange in years with small population densities (Hunt

et al. 2015). Further, it is known for many bird species that the abandoned parent successfully raises the young (Winkler 1987), but often in fewer numbers than the parents would raise together. By this way, they reduce the probability of being abandoned (Houston & Davies 1985, Witthling et al. 1994, Rosmanith et al. 2009). After establishing another brood with a new partner, the kingfisher male raised six young from the full clutch of seven eggs from previous brood, which is common for kingfisher (Čech 2009, own unpublished data.), as the male is an efficient hunter of a large prey (Čech & Čech 2017), for which it is not a problem to catch a sufficient number of fish and provide them to chicks (Vilches 2012, 2013). Finally, such behaviour indicates that leaving a partner is a minority strategy for kingfisher (Čech 2009, Libois 2018, own unpublished data). From this point of view, the male seems to have lost his mate rather than been abandoned.

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Súhrn

Rybárik riečny patrí medzi málo druhov, ktoré hniezdia niekoľkokrát do roka, pričom často jednotlivé hniezdenia prekrývajú. Dĺžka prekrývania hniezdení sa pohybuje od 5 do 23 dní a počet úspešných znášok sa pohybuje od jednej do štyroch. V roku 2018 bol zdokumentovaný prípad hniezdného páru, u ktorého sa na základe označenia jedincov a ich pozorovaním, ale už nie ich spätným odchytom predpokladalo, že hniezdil extrémne dlho a založil až päť znášok za sezónu. Dve hniezdenia boli neúspešné a z ďalších troch úspešne vychovali 19 mláďat. Cieľom tejto práce bolo zistiť, či u sledovaného hniezdného páru nedošlo k mimo-párovým interakciám alebo k výmene partnera v priebehu dlhej hniezdnej sezóny, ktoré sme pozorovaním nezaznamenali. Molekulárne analýzy odhalili, že samec M12059 síce hniezdil extrémne dlho

(spolu 185 dní), založil päť znášok, v ktorých vychoval 19 mláďat, avšak nehniedzil len s jednou samicou, ale s dvomi. Pomerne krátka vzdialenosť (170 m) medzi dvomi hniezdnymi norami spolu s vyvedením vysokého počtu mláďat z oboch hniezdení znižuje pravdepodobnosť, že by išlo o simultánnu bigamiu, pretože je známe, že ak rybárik hniezdia menej než 200 m od seba, správajú sa veľmi agresívne voči sebe navzájom, čo vyúsťuje do zníženého počtu vychovaného potomstva a často až do likvidácie znášok (Čech 2009). Preto predpokladáme, že došlo skôr k výmene partneriek a to konkrétne medzi tretím a štvrtým hniezdením, ktoré sa medzi sebou prekrývali 23 dní. Dĺžka prekryvu naznačuje, že samec musel novú partnerku nájsť ešte v priebehu inkubácie (viď tab. 1 v Rubáčová a Melišková 2020). Treba však dodať, že nedokážeme s istotou povedať, či došlo k opusteniu partnera a získaniu nového, alebo samec o partnerku nejakým spôsobom prišiel (napr. predáciou) a tak si našiel aj v priebehu náročnej hniezdnej fázy novú. Na základe štúdií ukazujúcich, že opustenie partnera je minoritnou stratégiou tohto druhu (Čech 2009, Libois 2018, naše nepublikované údaje), usudzujeme vyššiu pravdepodobnosť toho, že samec skôr o samicu prišiel, než že by ho bola opustila.

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