Behavioral Ecology

The official journal of the
ISBE
International Society for Behavioral Ecology

Behavioral Ecology (2022), 33(1), 245–251. https://doi.org/10.1093/beheco/arab130

Original Article

MHC-II distance between parents predicts sex allocation decisions in a genetically monogamous bird

Maxime Pineaux,[a] Thomas Merkling,[a] Etienne Danchin,[a] Scott A. Hatch,[b] Sarah Leclaire,[a,†] and Pierrick Blanchard[a,†]

[a]Laboratoire Évolution & Diversité Biologique (EDB UMR 5174), Université Fédérale de Toulouse Midi-Pyrénées, CNRS, IRD, UPS. 118 route de Narbonne, Bât. 4R1, 31062 Toulouse cedex 9, France and [b]Institute for Seabird Research and Conservation, 12850 Mountain Place, Anchorage, AK 99516, USA

Received 1 February 2021; revised 26 August 2021; editorial decision 13 October 2021; accepted 20 October 2021; Advance Access publication 9 November 2021.

Theory predicts that parental heritable characteristics should shape sex allocation decisions when their effects on reproduction or survival are offspring sex-dependent. Numerous studies have questioned to what extent characteristics displayed by one of the parents matched theoretical expectations. This contrasts with the handful of studies that investigated whether compatibility between parents could also trigger selective pressures for sex allocation adjustments. We studied the genetically monogamous black-legged kittiwake (*Rissa tridactyla*), where previous data revealed that female chicks suffered higher fitness costs from low diversity at genes of the major histocompatibility complex (MHC) than male chicks. We predicted, and found in our dataset, that MHC-similar parents, producing low MHC-diverse offspring, should avoid the production of females. The relation between MHC-distance between parents (i.e. the functional distinctness of their MHC alleles) and offspring sex was not linear, such that MHC-dissimilar parents also overproduced sons. Overall, our results suggest that the genetically monogamous black-legged kittiwake parents flexibly adapt their reproduction and circumvent the costs of suboptimal pairing by manipulating offspring sex.

Key words: **compatibility, heterozygote advantage, MHC, monogamy, sex allocation.**

INTRODUCTION

Sex allocation theory predicts that parents should adjust their investment in daughters and sons depending on the fitness costs and benefits associated with each sex (Trivers and Willard 1973; Charnov 1982; Frank 1990). Published data and theoretical models revealed that such sex-specific costs-benefits ratios are shaped by diverse abiotic and biotic parameters (reviewed in West 2009). These include parental heritable genetic or non-genetic characteristics when their effects on reproduction or survival are offspring sex-dependent (Cockburn et al. 2002; West 2009, chapter 6). One textbook example refers to situations where sons inherit elaborate ornaments from their father (e.g., Burley 1981). When these translate into increased reproductive success, such parents have been found to overproduce sons (West 2009, chapter 6; Bowers et al. 2013; but see Booksmythe et al. 2017).

Besides individual parental characteristics, only a handful of studies investigated whether compatibility between parents could also trigger selective pressures for sex allocation adjustments (Pryke and Griffith 2009a, b; Brekke et al. 2010; Rioux-Paquette et al. 2011; Sardell and DuVal 2014). This possibility was elegantly highlighted in Gouldian finches (*Erythrura gouldiae*), where daughters suffer higher viability costs from a Z-linked genetic incompatibility between red and black color morphs than sons (Pryke and Griffith 2009a, b). As predicted by sex allocation theory, females paired with a genetically incompatible male (i.e., an opposite-color morph) overproduced sons (Pryke and Griffith 2009a).

The major histocompatibility complex (hereafter, MHC) is a key group of genes involved in the activation of immune responses against parasites (Murphy and Weaver 2017). Here also, compatibility between parents plays a pivotal role in an evolutionary context as MHC-dissimilar mates are more likely to produce offspring carrying a higher diversity of MHC-alleles (Setchell et al. 2013), thereby able to recognize and eliminate a broader range of pathogens (Doherty and Zinkernagel 1975; Wakeland et al. 1990; Oliver et al. 2009). This increased resistance to diseases ultimately translates into an overall higher reproductive success and survival for more MHC-diverse individuals (Wedekind 1994; Brouwer et al. 2010; Thoss et al. 2011; Lenz et al. 2013). However, the fitness

Address correspondence to M. Pineaux. E-mail: m.pineaux@gmail.com.
†Co-last authors.

costs and benefits of MHC-diversity may differ among individuals depending on their exposure and immune responses to parasites (Roved et al. 2017; Whittingham et al. 2018, Pineaux et al. 2020), which are known to vary according to key characteristics such as personality (Boyer et al. 2010), social status (Habig and Archie 2015), or sex (Zuk 2009, Klein and Flanagan 2016). Some previous results revealed that sex could modulate the association between MHC-diversity and fitness, with males (Schaschl et al. 2012; Roved et al. 2018) or females (Hoover et al. 2018; Pineaux et al. 2020) suffering increased fitness costs from low MHC-diversity compared to the other sex. For instance, in Alpine chamois, males may benefit from higher levels of MHC-diversity than females because male-male contests increase males' risk of wounds and thus infections, and deplete males' energetic reserves, thereby possibly leading to less energy available for allocation to immune functions (Schaschl et al. 2012 and references therein). In a sex allocation context, this predicts that parents able to adjust offspring sex in relation to the expected fitness return of either sex given their MHC-compatibility should be advantaged. Although the MHC has been a trending topic in evolutionary ecology for two decades (Milinski 2006; Kamiya et al. 2014), no study has yet investigated whether MHC-compatibility between parents could drive sex allocation decisions.

We investigated MHC-based sex allocation decision in the genetically monogamous black-legged kittiwake (*Rissa tridactyla*), a species in which MHC-II diversity is associated with survival and other fitness proxies in female offspring, but not male offspring (Pineaux et al. 2020). This association also depends on female position in the laying sequence (two eggs being the typical clutch size in kittiwakes). In males and first-laid females, the probability of survival before fledging is 70%, irrespective of their MHC-II diversity. In second-laid female offspring, the probability of survival is similarly around 70% in the most MHC-diverse half, whereas it drops to 40% in the least MHC-diverse half (see Figure 1 in Pineaux et al. 2020). This may result from condition-dependent parasite infections differentially affecting females and males in relation to hatching order. We therefore predicted that MHC-II distance between parents, by determining offspring MHC-II diversity, would influence offspring sex in relation to laying position. Specifically, we expected a balanced sex ratio in MHC-dissimilar parents, whereas relatively more sons should be produced by MHC-similar parents at the second position of the laying sequence.

MATERIALS AND METHODS

Study site

The study was conducted during the 2009–2013 and 2016–2018 breeding seasons (May–August) on a colony of black-legged kittiwakes nesting on an abandoned U.S. Air Force radar tower on Middleton Island (59°26′N, 146°20′W), Gulf of Alaska. The nest sites created on the upper walls of the tower can be observed from inside through sliding one-way mirrors and birds can be individually identified using color and metal bands (Gill and Hatch 2002).

General procedure

We checked nest sites twice daily (9:00 and 18:00) to record laying and hatching events. On the day of laying, we individually labeled A- and B-eggs (first-and second-laid eggs, respectively) with a non-toxic marker. We determined offspring sex molecularly using DNA extracted from a drop of blood from the metatarsal vein a few hours after hatching, or from embryo tissues or blood vessels from eggshells when eggs did not hatch (see Merkling et al. 2012 for a detailed sexing protocol). Regarding adults, we used DNA extracted from a blood sample collected with a syringe or capillaries from the brachial vein to determine sex using the same molecular method as for chicks.

Molecular analysis of MHC-II

The DNA samples were used to amplify 258 bp fragments (218 bp excluding primers) of the exon 2 of the black-legged kittiwake MHC class-IIB. We used the MHC class-IIB specific primers (forward: 5′ GCACGAGCAGGGTATTTCCA and reverse: 5′ GTTCTGCCACACACTCACC) designed by Leclaire et al. (2014), which amplify at least four MHC class-IIB loci (Pineaux et al. 2020). Samples were sequenced in two runs with an Illumina MiSeq platform, using the 2 × 300 bp protocol (Fasteris SA, Plan-les-Ouates, Switzerland; see Pineaux et al. 2020, for a detailed sequencing protocol). Amplicon sequences were analyzed with ampliSAS, a three-step pipeline that consists of read demultiplexing, unique sequence clustering, and erroneous sequence filtering (Sebastian et al. 2016). The reproducibility of genotype between the two runs (n = 25 DNA samples that were split and processed in independent PCRs) was 100%. We obtained 83 different MHC class II alleles and, in the subsample used in this study, the mean number of alleles per individual was 3.29 ± 0.76 (± SD; range: 1–5).

We calculated the functional MHC-II distance between mates in pairs for which the MHC class-IIB region was sequenced for both mates, using the approach described in Strandh et al. (2012). To obtain functional alleles, we translated MHC-II DNA sequences into amino acid sequences and considered DNA sequences as functionally identical if they had the same amino-acids in the peptide-binding regions (PBRs; inferred from Leclaire et al. 2014). This gives us a total of 68 functional alleles. To calculate functional distance, we first follow the approach of Schwensow et al. (2007) to describe the chemical binding properties of each amino acid in the PBRs using five physico-chemical descriptors (z-descriptors; Sandberg et al. 1998). Then, following the approach of Strandh et al. (2012), the resulting Sandberg matrix was used to construct an alternative maximum-likelihood phylogenetic tree with "Rcontml" in the R package *Rphylip* (Revell and Chamberlain 2014). This tree represents clusters of functionally-similar MHC sequences and was used as a reference to calculate the functional distance between MHC-sequence repertoires of parents with unweighted UniFrac analyses ("GUniFrac" package in R; Chen 2021). Functional MHC-II distance between parents varied from 0 to 1 (mean ± SD: 0.54 ± 0.19). A score of zero means that both parents have exactly the same MHC alleles, whereas the closer to one their score is, the more functionally dissimilar their MHC alleles are. MHC-distance did not significantly vary among years (Kruskal–Wallis, U = 7.15, df = 7, P = 0.41).

The tree was also used to calculate the functional diversity of offspring. To calculate functional MHC-II diversity, we used the minimum total length of all the branches required to span an offspring's MHC-II alleles (i.e., Faith's phylogenetic diversity; Faith 1992) with the R function "pd" in the *picante* R package (Kembel et al. 2010). In other words, for each additional allele, only the part of the peptide-binding characteristics that is not shared with other alleles is summed (Pineaux et al. 2020). The resulting score is thus positively correlated to the range of peptides bound by all the alleles carried by an individual. Offspring functional MHC-II diversity varied from 0.89 to 9.81 (mean ± SD: 5.97 ± 1.12) and did not significantly vary among years (Kruskal–Wallis, U = 2.70, df = 6, P = 0.85).

Sample size

We obtained MHC-II distance for 293 pairs that produced 548 two-eggs clutches, totaling 933 chicks and 163 unhatched eggs. Clutch size ranges from one to three eggs in this species, two-eggs clutches being the most common clutch size in this population (Gill and Hatch 2002; 81% of the clutches in these study years). We did not include three-eggs clutches in the analyses because they were too rare (n = 9), nor we included one-egg clutches (n = 119) as our previous study shows no sex-specific effect of MHC-II diversity on fitness in offspring raised alone (Pineaux et al. 2020). Still, including one egg-clutches in the main analysis testing for effects of MHC-II distance on offspring sex produces the same results (data not shown). We sexed 913 out of the 933 chicks (97% of chicks) and 45 embryos out of the 163 unhatched eggs (27% of unhatched eggs). We used these 958 sexed embryos or chicks in our main analysis relating offspring sex to MHC-II distance between parents. Additionally, in order to investigate the relationship between MHC-II distance between parents and chick MHC-II diversity, we used a subsample of those offspring (n = 471) that had been sequenced for the MHC-II as part of our previous study linking offspring MHC-II diversity to fitness (Pineaux et al. 2020).

Data analysis

First, we tested whether MHC-II diversity of offspring was positively associated with MHC-II distance between parents using a linear mixed model (LMM) built in the *lme4* package (Bates et al. 2015) in R 4.0.1 (R Core Team 2020). Predictor variables included MHC-II distance between parents, offspring sex, laying position, and interactions between these variables. Clutch identity (ID) and pair ID were included as random effects to consider the nonindependence of chicks born during the same breeding season (clutch ID) or born from the same parents in different years (pair ID). However, variance estimates of the clutch ID random effect was practically zero and was thus removed. We standardized fixed predictors by centering and dividing them by two standard deviations using the *arm* package (Gelman and Su 2018). Model selection followed a backward-stepwise approach using the "step" function with Kenward-Roger's approximation of denominator degrees of freedom in the R package *lmerTest* (Kuznetsova et al. 2017). We checked for normality and homoscedasticity of residuals and for normal distribution of random effects in the initial model.

Then, we used the same backward-stepwise approach to test the association between offspring sex and MHC-II distance between parents according to laying position. We built a generalized linear mixed model (GLMM) with a binomial error distribution and a logit link function (i.e., offspring sex was either 0 = female or 1 = male) in the *lme4* package (Bates et al. 2015). Predictor variables included MHC-II distance between parents, laying position and their interaction. We transformed the parental MHC-II distance using a restricted cubic spline (RCS) because we did not expect the association between parental MHC-II distance and offspring sex to be linear at the second position of the laying sequence (see predictions in the introduction). RCS transforms an explanatory variable by dividing the range of values in intervals, fits a separate curve in each interval but still results in a smooth and continuous fitted curve. Intervals are delimited by knots. We used the default knot positions from the *rms* R package (Harrell 2020) and we found the optimal number of knots to be three by comparing the Akaike Information Criteria (AIC) of models with three, four or five knots. Year was included as a continuous variable, given that an increase in the probability of producing sons with time was found in this population during the period considered (Merkling et al. 2019). We also included clutch ID and pair ID as random effects and we checked for normal distribution of these random effects. We standardized fixed variables using the *arm* package (Gelman and Su 2018). We assessed significance of each predictor variable by the change in deviance after removal of that variable (Likelihood-Ratio Test [LRT]) using a chi-square test. A variable was eliminated from the model if $P > 0.05$.

Following recommendations (Krackow and Neuhauser 2008), we performed the same analyses on two datasets, an "unrestricted dataset" (N = 958) containing both complete (where both offspring had been sexed) and incomplete clutches (where only one offspring had been sexed), and a "restricted dataset" (N = 820) containing only complete broods. We also re-ran analyses twice on a modified form of our unrestricted dataset by assuming that all unsexed offspring (N = 138) were females or, alternatively, males. These additional analyses allowed us to investigate whether the reported patterns could result from sex bias in mortality and/or sexing success.

RESULTS

Offspring MHC-II diversity was positively associated with the MHC-II distance between parents. All other explicative variables were lost in the backward-stepwise procedure (Figure 1; Table 1). This analysis may face collinearity issues since chick sex was related to MHC-II distance between parents in our data (see the test of our main prediction below), both parameters being included

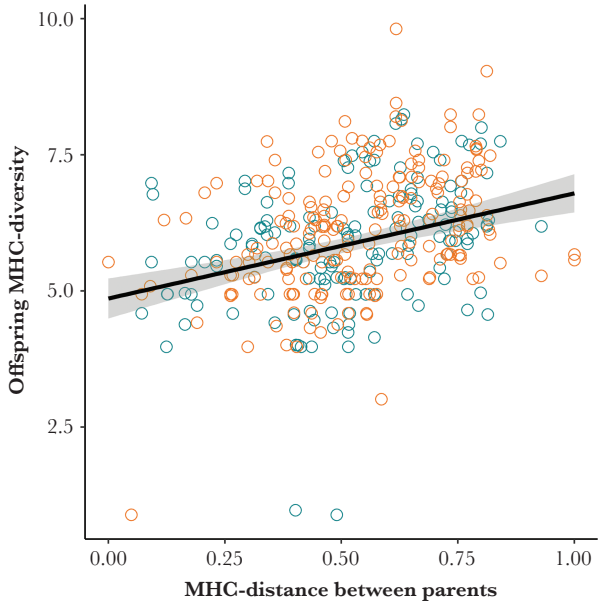

Figure 1
Offspring MHC-II diversity covaries with the MHC-II distance between parents in both female (orange) and male (blue) offspring. The line shows the predictions from a LMM including MHC-II distance between parents as a predictor variable. There was no significant interaction between offspring sex and MHC-II distance (Table 1). The pair ID random effect was not considered in the models used for graphic representations but was accounted for in the analysis. Removing the three extremely low MHC-II diverse offspring did not change the results. Shaded areas represent confidence intervals.

Table 1

(a) Effect of predictor variables from the generalized linear mixed model built to explain chick MHC-II diversity and (b) variance and standard deviation associated with random effects in the final model

(a)

Parameter	Estimate	SE	F	P	Step
MHC-II distance: Sex: Hatching order	0.027	0.333	0.641	0.423	1
MHC-II distance: Hatching order	0.028	0.143	0.040	0.842	2
Sex: Hatching order	0.043	0.162	0.071	0.790	3
Hatching order	0.014	0.071	0.036	0.849	4
MHC-II distance: Sex	0.070	0.162	0.183	0.669	5
Sex	−0.122	0.081	2.235	0.135	6
MHC-II distance	**0.859**	**0.133**	**37.945**	**<0.001**	

(b)

Random effect	Variance	SD
Pair ID	0.621	0.788

Variables were eliminated following a backward-stepwise procedure. Step denotes the exclusion sequence of the nonsignificant terms of the model. Values for excluded variables refer to the step before their exclusion. Values included in the final model are in bold.

Table 2

(a) Effect of predictor variables from the generalized linear mixed model built to explain chick sex and (b) variance and standard deviation associated with random effects in the final model

(a)

Parameter	Estimate	SE	Chi²	P	Step
Cubic spline (MHC-II distance): Hatching order	0.133	0.680	0.038	0.845	1
MHC-II distance: Hatching order	-0.066	0.267	0.062	0.803	2
Year	-0.140	0.136	1.060	0.303	3
Hatching order	0.219	0.132	2.745	0.098	4
Cubic spline (MHC-II distance)	**0.849**	**0.358**	**5.793**	**0.016**	

(b)

Random effect	Variance	SD
Clutch ID	0.162	0.127
Pair ID	0.090	0.300

Variables were eliminated following a backward-stepwise procedure. Step denotes the exclusion sequence of the nonsignificant terms of the model. Values for excluded variables refer to the step before their exclusion. Values included in the final model are in bold. When using a restricted cubic spline, one must include the untransformed predictor variable in the model to force (i.e., to restrict) the curve to be linear at the tails of the predictor variable (MHC-II distance here) to avoid unstable estimates. Note that the best way to interpret results from restricted cubic splines is to use a figure, not the estimated coefficients (Shepherd et al. 2017).

concomitantly into the initial model. However, variance inflated factor (VIF) values were < 2, indicating no such an issue (Zuur et al. 2010).

We then investigated our main prediction. Using the unrestricted dataset (containing both complete and incomplete clutches), offspring sex was significantly associated with the cubic spline transformed parental MHC-II distance while the other predictor variables were eliminated in the backward-stepwise procedure (Table 2). The curve was "U" shaped, with knots at an MHC-II distance of 0.30, 0.55, and 0.77 (Figure 2). More MHC-II similar pairs overproduced sons independently of laying position (Figure 2). Among offspring produced by the most MHC-II similar pairs (i.e., first of 30-quantiles), 26/39 (67%) were sons. Contrary as expected, more MHC-II dissimilar pairs did not produce a balanced sex ratio. They produced relatively more sons (Figure 2), with the most MHC-II dissimilar pairs (i.e., last of 30-quantiles) having produced 19/31 (61%) sons.

The analyses performed on the restricted dataset (containing only complete clutches) gave similar results (Supplementary Table S1). Furthermore, assuming that all unsexed offspring were females, or alternatively males, both lead to the same "U" shaped curve, indicating an overproduction of sons in more MHC-II similar pairs and in more MHC-II dissimilar pairs (Supplementary Tables S2 and S3).

DISCUSSION

Our data first confirmed that MHC-II similar kittiwake parents were more likely to produce offspring with low MHC-II diversity. Previous results reported that such a low MHC-II diversity in offspring was

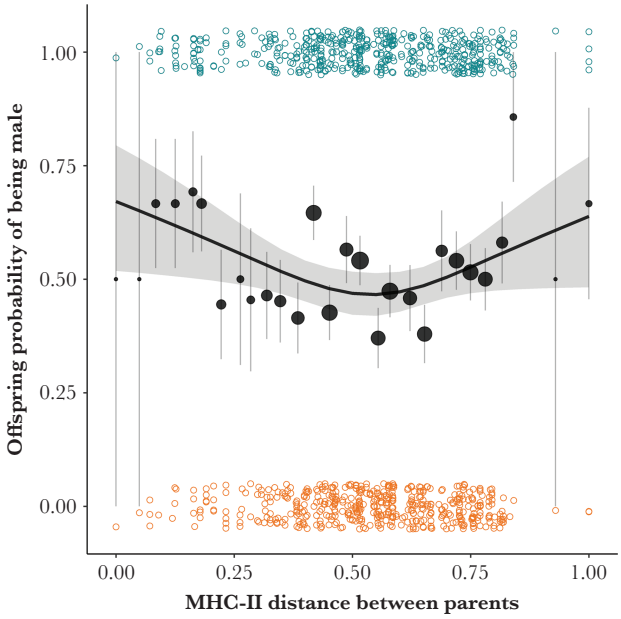

Figure 2
Offspring probability of being male according to MHC-II distance between parents. Each colored dot represents a female chick (orange; n = 472) or a male chick (blue; n = 486). For illustrative purpose, parental MHC-II distance was divided into 30 categories of equal range (0.033), with the black dots representing the mean (± SE) sex ratio per category of parental MHC-II distance, and the size of the dots representing sample size per category. The curve represents predicted values derived from a model including parental MHC-II distance transformed with a restricted cubic spline (see Methods). Shaded areas represent 95% confidence intervals. Random effects (pair ID and clutch ID) were not considered in this model used for graphic representation. Note: the vertical position of colored dots was randomly rearranged to better appreciate the number of chicks in relation to parental MHC-II distance.

associated with increased mortality in daughters hatched in second position as compared to other chick sex-rank categories (Pineaux et al. 2020). Additionally, low MHC-II diversity negatively affected growth and tick resistance in daughters only (Pineaux et al. 2020) In line with sex allocation theory (Cockburn et al. 2002; West 2009, chapter 6), our data revealed that in such a context, MHC-II similar parents avoided production of disadvantaged daughters. Contrary to our expectation, however, we did not find hatching rank to further modulate the association between parental MHC-II distance and offspring sex. The overall increased detrimental effect of low MHC-II diversity in daughters as compared to sons (Pineaux et al. 2020) may have concealed more subtle patterns.

Unexpectedly, our data also revealed that MHC-dissimilar pairs overproduced sons. This may lead to an increased fitness return if MHC-II diverse males have increased survival or reproductive advantages compared to MHC-II diverse females later in life, as shown in other species (Sauermann et al. 2001; Schaschl et al. 2012; Roved et al. 2018). Notably, given that body mass and body size may be more important determinants of male than female reproductive success in adult kittiwakes (Merkling et al. 2012), MHC-diverse males may obtain a competitive and, *in fine*, reproductive advantage, if MHC-diversity is positively associated with such morphological traits, as found in other species (Ditchkoff et al. 2001; Lenz et al. 2009; Dunn et al. 2013). However, when testing this relationship using kittiwake parents involved in this study, we found no such a positive association between MHC-diversity and body mass or body size in adult males (see Supplementary Tables S4 and S5). More research is needed to identify the possible fitness advantages of high MHC diversity in adult males in kittiwakes.

Previous studies investigated sex ratio patterns in this population (Merkling et al. 2012; Merkling et al. 2015; Merkling et al. 2019). Using 10 years of data from a long-term feeding experiment, the most recent results suggested that offspring sex is not shaped by pair's investment capacity (Merkling et al. 2019) despite higher reproductive costs for parents raising sons (Merkling et al. 2015; Merkling et al. 2017). Here, our results indicate that offspring genetic diversity may be a stronger driver of sex allocation decision in this population than pair's ability to provide care.

Sex allocation based on MHC similarity between parents has been suggested in humans, rats and mice because newborn males have been found to be more MHC-diverse than newborn females in these species (Dorak et al. 2002, and references therein). However, whether this result was caused by MHC-similar parents overproducing daughters was not known. A study on humans found that parents sharing the same alleles at two MHC loci produced a female-biased sex ratio whereas parents sharing no allele at these two MHC loci produced a male-biased sex ratio (Astolfi et al. 1990). However, the potential adaptive value of this pattern (e.g., whether males suffered more from low MHC-diversity than females) also remains overlooked (Sauermann et al. 2001; Schaschl et al. 2012; Roved et al. 2018). Clearly, important next steps should involve studies investigating potential fitness pathways and proximate mechanisms underlying sex ratio departure from parity.

Proximate mechanisms of sex ratio adjustments are not well understood and how these could depend on MHC is unknown. Regardless of the parent(s) biasing offspring sex, our results may suggest that kittiwakes can assess the genetic characteristics of their mate (as suggested by Mulard et al. 2009; Pineaux et al. 2019). The covariation between scent-gland compounds and MHC in this species may suggest that odor cues might be used in MHC recognition (Leclaire et al. 2014), as found in several taxa (Wedekind et al. 1995; Olsson et al. 2003; Radwan et al. 2008), including birds (Leclaire et al. 2017). Sex ratio adjustments may also be the result of MHC-specific sperm-ova interactions (Wedekind 1994), in line with previous studies reporting non-random production of blastocysts according to the MHC-distance between gametes (Lenz et al. 2018; Zhu et al. 2019).

Because permanent or temporary constraints may force individuals to mate with suboptimal partners (Stutchbury and Morton 1995; Tinghitella et al. 2013), tactics allowing to lessen associated costs may have emerged. Such constraints are particularly likely to happen in genetically monogamous species such as the kittiwake (Helfenstein et al. 2004). We previously reported that breeding kittiwakes flexibly adapted their breeding timing and copulatory behavior in response to within-pair genetic similarity (Pineaux et al. 2019). The present study suggests another way for kittiwake parents to circumvent fitness costs associated to suboptimal pairing.

SUPPLEMENTARY MATERIAL

Supplementary data are available at *Behavioral Ecology* online.

FUNDING

Fieldwork was supported by a grant from the Institut Polaire Français Paul-Emile Victor (IPEV "Programme 1162 SexCoMonArc") to S.L. and E.D. This work originated in the laboratory "Evolution et Diversité Biologique" (EDB) and was supported by the French Laboratory of Excellence (LabEx)

Project "TULIP" (ANR-10-LABX-41; ANR-11-IDEX-0002-02), and the Agence Nationale de la Recherche Française (ANR grant "BactOdo", no. ANR-13-PDOC-0002 to S.L.). M.P. was supported by a French doctoral scholarship. The authors declare no conflict of interest.

We are grateful to the Middleton fieldworkers who collected data for this study, as well as the undergraduate students who helped with chick sexing. The study was conducted under the approval of the U.S. Geological Survey Alaska Science Center, the University of Alaska Institutional Animal Care and Use Committees, and the Institut Paul-Emile Victor Ethical Committee, in accordance with United States laws and under permits from the U.S. Fish and Wildlife Service and the State of Alaska. Any use of trade names is for descriptive purposes only and does not imply endorsement by the U.S. Government.

Conflict of interest: The authors declare no conflict of interest.

Data availability: Analyses reported in this article can be reproduced using the data provided by Pineaux et al (2021).

Handling editor: Jan Komdeur

REFERENCES

Astolfi P, Martinetti M, Gigli-Berzolari F, Cuccia M. 1990. The effect of parental and maternal-fetal histocompatibility at MHC on sex ratio in offspring. Tissue Antigens. 35:172–177.

Bates D, Machler M, Bolker BM, Walker SC. 2015. Fitting linear mixed-effects models using lme4. J Stat Softw. 67:1–48.

Booksmythe I, Mautz B, Davis J, Nakagawa S, Jennions MD. 2017. Facultative adjustment of the offspring sex ratio and male attractiveness: a systematic review and meta-analysis. Biol Rev Camb Philos Soc. 92:108–134.

Bowers EK, Munclinger P, Bureš S, Kučerová L, Nádvorník P, Krist M. 2013. Cross-fostering eggs reveals that female collared flycatchers adjust clutch sex ratios according to parental ability to invest in offspring. Mol Ecol. 22:215–228.

Boyer N, Réale D, Marmet J, Pisanu B, Chapuis JL. 2010. Personality, space use and tick load in an introduced population of Siberian chipmunks *Tamias sibiricus*. J Anim Ecol. 79:538–547.

Brekke P, Bennett PM, Wang J, Pettorelli N, Ewen JG. 2010. Sensitive males: inbreeding depression in an endangered bird. Proc Biol Sci. 277:3677–3684.

Brouwer L, Barr I, van de Pol M, Burke T, Komdeur J, Richardson DS. 2010. MHC-dependent survival in a wild population: evidence for hidden genetic benefits gained through extra-pair fertilizations. Mol Ecol. 19:3444–3455.

Burley N. 1981. Sex ratio manipulation and selection for attractiveness. Science. 211:721–722.

Charnov E. 1982. The theory of sex allocation. Princeton: Princeton University Press.

Chen J. 2021. GUniFrac: Generalized UniFrac Distances. In, 1.3 R package. https://cran.r-project.org/package=GUniFrac.

Cockburn A, Legge S, Double M. 2002. Sex ratios in birds and mammals: can the hypotheses be disentangled? In: ICW H, editors. Sex ratios: concept and research methods. Cambridge (UK): Cambridge University Press.

Ditchkoff SS, Lochmiller RL, Masters RE, Hoofer SR, Van Den Bussche RA. 2001. Major-histocompatibility-complex-associated variation in secondary sexual traits of white-tailed deer (*Odocoileus virginianus*): evidence for good-genes advertisement. Evolution. 55:616–625.

Doherty PC, Zinkernagel RM. 1975. Enhanced immunological surveillance in mice heterozygous at the H-2 gene complex. Nature. 256:50–52.

Dorak MT, Lawson T, Machulla HK, Mills KI, Burnett AK. 2002. Increased heterozygosity for MHC class II lineages in newborn males. Genes Immun. 3:263–269.

Dunn PO, Bollmer JL, Freeman-Gallant CR, Whittingham LA. 2013. MHC variation is related to a sexually selected ornament, survival, and parasite resistance in common yellowthroats. Evolution. 67:679–687.

Faith DP. 1992. Conservation evaluation and phylogenetic diversity. Biol Conserv 61:1–10

Frank SA. 1990. Sex allocation theory for birds and mammals. Annu Rev Ecol Systemat. 21:13–55.

Gelman A, Su Y. 2018. arm: Data analysis using regression and multilevel/hierarchical models. In, 1.10-1 R package. https://CRAN.R-project.org/package=arm edn.

Gill VA, Hatch SA. 2002. Components of productivity in black-legged kittiwakes *Rissa tridactyla*: response to supplemental feeding. J Avian Biol. 33:113–126.

Habig B, Archie EA. 2015. Social status, immune response and parasitism in males: a meta-analysis. Philos Trans R Soc Lond B Biol Sci. 370:1669.

Harrell FE. 2020. rms: Regression Modeling Strategies. In, 6.0-1 R package. https://cran.r-project.org/package=rms edn.

Helfenstein F, Tirard C, Danchin E, Wagner RH. 2004. Low frequency of extra-pair paternity and high frequency of adoption in Black-legged Kittiwakes. Condor. 106:149–155.

Hoover B, Alcaide M, Jennings S, Sin SYW, Edwards SV, Nevitt GA. 2018. Ecology can inform genetics: disassortative mating contributes to MHC polymorphism in Leach's storm-petrels (*Oceanodroma leucorhoa*). Mol Ecol. 27:3371–3385.

Kamiya T, O'Dwyer K, Westerdahl H, Senior A, Nakagawa S. 2014. A quantitative review of MHC-based mating preference: the role of diversity and dissimilarity. Mol Ecol. 23:5151–5163.

Kembel SW, Cowan PD, Helmus MR, Cornwell WK, Morlon H, Ackerly DD, Blomberg SP, Webb CO. 2010. Picante: R tools for integrating phylogenies and ecology. Bioinformatics. 26:1463–1464.

Klein SL, Flanagan KL. 2016. Sex differences in immune responses. Nat Rev Immunol. 16:626–638.

Krackow S, Neuhauser M. 2008. Insights from complete-incomplete brood sex-ratio disparity. Behav Ecol Sociobiol. 62:469–477.

Kuznetsova A, Brockhoff PB, Christensen RHB. 2017. lmerTest package: tests in linear mixed effects models. J Stat Softw. 82:1–26.

Leclaire S, Strandh M, Mardon J, Westerdahl H, Bonadonna F. 2017. Odour-based discrimination of similarity at the major histocompatibility complex in birds. Proc Biol Sci. 284:5.

Leclaire S, van Dongen WF, Voccia S, Merkling T, Ducamp C, Hatch SA, Blanchard P, Danchin E, Wagner RH. 2014. Preen secretions encode information on MHC similarity in certain sex-dyads in a monogamous seabird. Sci Rep. 4:6920.

Lenz TL, Hafer N, Samonte IE, Yeates SE, Milinski M. 2018. Cryptic haplotype-specific gamete selection yields offspring with optimal MHC immune genes. Evolution. 72:2478–2490.

Lenz TL, Mueller B, Trillmich F, Wolf JB. 2013. Divergent allele advantage at MHC-DRB through direct and maternal genotypic effects and its consequences for allele pool composition and mating. Proc Biol Sci. 280:20130714.

Lenz TL, Wells K, Pfeiffer M, Sommer S. 2009. Diverse MHC IIB allele repertoire increases parasite resistance and body condition in the Long-tailed giant rat (*Leopoldamys sabanus*). BMC Evol Biol. 9:269.

Merkling T, Blanchard P, Chastel O, Glauser G, Vallat-Michel A, Hatch SA, Danchin E, Helfenstein F. 2017. Reproductive effort and oxidative stress: effects of offspring sex and number on the physiological state of a long-lived bird. Funct Ecol. 31:1201–1209.

Merkling T, Hatch S, Leclaire S, Danchin E, Blanchard P. 2019. Offspring sex-ratio and environmental conditions in a seabird with sex-specific rearing costs: a long-term experimental approach. Evol Ecol. 33:417–433.

Merkling T, Leclaire S, Danchin E, Lhuillier E, Wagner RH, White J, Hatch SA, Blanchard P. 2012. Food availability and offspring sex in a monogamous seabird: insights from an experimental approach. Behav Ecol. 23:751–758.

Merkling T, Welcker J, Hewison AJM, Hatch SA, Kitaysky AS, Speakman JR, Danchin E, Blanchard P. 2015. Identifying the selective pressures underlying offspring sex-ratio adjustments: a case study in a wild seabird. Behav Ecol. 26:916–925.

Milinski M. 2006. The major histocompatibility complex, sexual selection, and mate choice. Annu Rev Ecol Syst. 37:159–186.

Mulard H, Danchin E, Talbot SL, Ramey AM, Hatch SA, White JF, Helfenstein F, Wagner RH. 2009. Evidence that pairing with genetically similar mates is maladaptive in a monogamous bird. BMC Evol Biol. 9:1–12.

Murphy K, Weaver C. 2017. Janeway's immunobiology, 9th edn. LLC, New York, NY: Garland Science, Taylor & Francis Group.

Oliver MK, Telfer S, Piertney SB. 2009. Major histocompatibility complex (MHC) heterozygote superiority to natural multi-parasite infections in the water vole (*Arvicola terrestris*). Proc Biol Sci. 276:1119–1128.

Olsson M, Madsen T, Nordby J, Wapstra E, Ujvari B, Wittsell H. 2003. Major histocompatibility complex and mate choice in sand lizards. Proc Biol Sci. 270(Suppl 2):S254–S256.

Pineaux M, Blanchard P, Danchin E, Hatch S, Helfenstein F, Mulard H, White J, Leclaire S, Wagner R. 2019. Behavioural avoidance of sperm ageing depends on genetic similarity of mates in a monogamous seabird. Biol J Linn Soc. 128:170–180.

Pineaux M, Merkling T, Danchin E, Hatch S, Duneau D, Blanchard P, Leclaire S. 2020. Sex and hatching order modulate the association between MHC-II diversity and fitness in early-life stages of a wild seabird. Mol Ecol. 29:3316–3329.

Pineaux M, Merkling T, Danchin E, Hatch S, Leclaire S, Blanchard P. 2021. MHC-II distance between parents predicts sex allocation decisions in a genetically monogamous bird. Behav Ecol. doi:10.5061/dryad.prr4xgxnc

Pryke SR, Griffith SC. 2009a. Genetic incompatibility drives sex allocation and maternal investment in a polymorphic finch. Science. 323:1605–1607.

Pryke SR, Griffith SC. 2009b. Postzygotic genetic incompatibility between sympatric color morphs. Evolution. 63:793–798.

Radwan J, Tkacz A, Kloch A. 2008. MHC and preferences for male odour in the bank vole. Ethology. 114:827–833.

R Core Team. 2020. R: a language and environment for statistical computing. Vienna (Austria): R Foundation for Statistical Computing.

Revell LJ, Chamberlain SA. 2014. Rphylip: an R interface for PHYLIP. Methods Ecol Evol. 5:976–981.

Rioux-Paquette E, Festa-Bianchet M, Coltman DW. 2011. Sex-differential effects of inbreeding on overwinter survival, birth date and mass of bighorn lambs. J Evol Biol. 24:121–131.

Roved J, Hansson B, Tarka M, Hasselquist D, Westerdahl H. 2018. Evidence for sexual conflict over major histocompatibility complex diversity in a wild songbird. Proc Biol Sci. 285:20180841.

Roved J, Westerdahl H, Hasselquist D. 2017. Sex differences in immune responses: Hormonal effects, antagonistic selection, and evolutionary consequences. Horm Behav. 88:95–105.

Sandberg M, Eriksson L, Jonsson J, Sjöström M, Wold S. 1998. New chemical descriptors relevant for the design of biologically active peptides. A multivariate characterization of 87 amino acids. J Med Chem. 41:2481–2491.

Sardell RJ, DuVal EH. 2014. Differential allocation in a lekking bird: females lay larger eggs and are more likely to have male chicks when they mate with less related males. Proc Biol Sci. 281:20132386.

Sauermann U, Nürnberg P, Bercovitch FB, Berard JD, Trefilov A, Widdig A, Kessler M, Schmidtke J, Krawczak M. 2001. Increased reproductive success of MHC class II heterozygous males among free-ranging rhesus macaques. Hum Genet. 108:249–254.

Schaschl H, Suchentrunk F, Morris DL, Ben Slimen H, Smith S, Arnold W. 2012. Sex-specific selection for MHC variability in Alpine chamois. BMC Evol Biol. 12:20.

Schwensow N, Fietz J, Dausmann KH, Sommer S. 2007. Neutral versus adaptive genetic variation in parasite resistance: importance of major histocompatibility complex supertypes in a free-ranging primate. Heredity (Edinb). 99:265–277.

Sebastian A, Herdegen M, Migalska M, Radwan J. 2016. AMPLISAS: a web server for multilocus genotyping using next-generation amplicon sequencing data. Mol Ecol Resour. 16:498–510.

Setchell JM, Abbott KM, Gonzalez JP, Knapp LA. 2013. Testing for postcopulatory selection for major histocompatibility complex genotype in a semi-free-ranging primate population. Am J Primatol. 75:1021–1031.

Shepherd BE, Rebeiro PF; Caribbean, Central and South America network for HIV epidemiology. 2017. Brief Report: assessing and interpreting the association between continuous covariates and outcomes in observational studies of HIV using splines. J Acquir Immune Defic Syndr. 74:e60–e63.

Strandh M, Westerdahl H, Pontarp M, Canbäck B, Dubois MP, Miquel C, Taberlet P, Bonadonna F. 2012. Major histocompatibility complex class II compatibility, but not class I, predicts mate choice in a bird with highly developed olfaction. Proc Biol Sci. 279:4457–4463.

Stutchbury BJ, Morton ES. 1995. The effect of breeding synchrony on extra-pair mating systems in songbirds. Behaviour. 132:675–690.

Thoss M, Ilmonen P, Musolf K, Penn DJ. 2011. Major histocompatibility complex heterozygosity enhances reproductive success. Mol Ecol. 20:1546–1557.

Tinghitella RM, Weigel EG, Head M, Boughman JW. 2013. Flexible mate choice when mates are rare and time is short. Ecol Evol. 3:2820–2831.

Trivers RL, Willard DE. 1973. Natural selection of parental ability to vary the sex ratio of offspring. Science. 179:90–92.

Wakeland EK, Boehme S, She JX, Lu CC, McIndoe RA, Cheng I, Ye Y, Potts WK. 1990. Ancestral polymorphisms of MHC class II genes: divergent allele advantage. Immunol Res. 9:115–122.

Wedekind C. 1994. Mate choice and maternal selection for specific parasite resistances before, during and after fertilization. Philos Trans R Soc Lond B Biol Sci. 346:303–311.

Wedekind C, Seebeck T, Bettens F, Paepke AJ. 1995. MHC-dependent mate preferences in humans. Proc Biol Sci. 260:245–249.

West SA. 2009. Sex allocation. Princeton: Princeton University Press.

Whittingham LA, Dunn PO, Freeman-Gallant CR, Taff CC, Johnson JA. 2018. Major histocompatibility complex variation and blood parasites in resident and migratory populations of the common yellowthroat. J Evol Biol. 31:1544–1557.

Zhu Y, Wan QH, Zhang HM, Fang SG. 2019. Reproductive strategy inferred from major histocompatibility complex-based inter-individual, sperm-egg, and mother-fetus recognitions in giant pandas (*Ailuropoda melanoleuca*). Cells. 8:21.

Zuk M. 2009. The sicker sex. PLoS Pathog. 5:e1000267.

Zuur AF, Ieno EN, Elphick CS. 2010. A protocol for data exploration to avoid common statistical problems. Methods Ecol Evol. 1:3–14.