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SHORT REPORT

Inbreeding in the last ruling dynasty of Portugal: The house of Braganza

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Abstract

Objectives: To determine whether: (1) there are high levels of inbreeding in a European royal dynasty that continues until the 20th century, and (2) whether inbreeding is negatively associated with pre-reproductive survival and longevity.

Methods: Genealogical information of all Braganza monarchs (1640-1910) was used to compute the individual's inbreeding coefficient (F) and the coefficient of kinship (θ) of the marriage which were examined in relation to two life-history traits.

Results: Mean F of the monarchs was 0.0530, close to that corresponding to the progeny of first cousins ($F = 0.0625$). A statistically significant effect of maternal inbreeding on offspring longevity ($P = 0.037$) and a significant effect of individual's F on survival from birth to 10 years ($P = 0.023$) were detected.

Conclusions: Another European royal dynasty besides the Habsburgs had high levels of inbreeding, especially high during a period after the early modern age. The lineage showed evidence of inbreeding depression. Results support the hypothesis that an increase in maternal homozygosity affects the lifespan of the progeny.

1 | INTRODUCTION

Inbreeding, defined as mating between individuals related by common ancestry, often leads to a reduction in fitness in offspring, a phenomenon known as inbreeding depression (Charlesworth & Willis, 2009). There is ample evidence of inbreeding depression in survival (Berra, Álvarez, & Ceballos, 2010; Bittles & Black, 2010; Bittles, 2012; Ceballos & Álvarez, 2013), reproductive success (Álvarez, Ceballos, & Berra, 2015; Ober, Hyslop, & Hauck, 1999; Postma, Martini, & Martini, 2010; Robert et al., 2009) and other complex traits (Fareed & Afzal, 2014; Joshi et al., 2015; Verweij et al., 2014) across human populations. Given the detrimental effects of inbreeding observed in many animals (Keller & Waller, 2002), inbreeding depression is recognized as a significant evolutionary factor that may drive the evolution of mate choice (Blouin & Blouin, 1988). However, our species has favored consanguineous marriages for cultural reasons, particularly in certain populations or lineages, such as royal and upper bourgeoisie dynasties, which have used the matrimonial policy as a strategy to retain economic/political power (Goody, 1983; Kuper, 2009). Therefore, the royal dynasties offer an excellent opportunity to study the effects of inbreeding along a lineage. The only European royal dynasty whose levels of inbreeding have been studied in detail are the Habsburgs, which showed evidence of inbreeding depression affecting survival (Álvarez & Ceballos, 2015; Álvarez, Ceballos, & Quinteiro, 2009; Ceballos & Álvarez, 2013). Here, we studied another dynasty, the House of Braganza (1640-1910), that has given rise to speculation about detrimental effects of inbreeding in some of its members (Peters & Willis, 2013). In particular, we study levels of inbreeding and its possible effects in survival and longevity in order to find out whether or not the Habsburg case is a European singularity.

2 | METHODS

2.1 | Genealogical and demographic data

Genealogical and demographic information for the Braganza dynasty was obtained and contrasted from different publicly-

available online sources: The Geneall Project, The Peerage, and Wikipedia. After cleaning and validation, genealogical data were used to construct a large family tree, which included all the ancestors of each king and queen in at least seven complete generations. The individual's inbreeding coefficient (F) and the coefficient of kinship (θ) of marriage were computed using FSpeed Pro computer program (Tenset Technologies). θ between two individuals is the probability that two alleles at the same locus drawn at random, one from each individual, are identical by descent, and, therefore, the F of an individual is the θ of his/her parents (Lynch & Walsh, 1998).

Demographic data consisted of the birth and death dates of the Braganza monarchs and their children. The Braganza monarchs contracted a total of 17 marriages: four of them had no children, three had one or two children, and 10 had three or more children. Survival analysis was performed only with families with at least three children. Progeny mortality data of the monarchs were classified into two categories: infant deaths excluding miscarriages (in the first year of life) and children deaths (between one to 10 years). The 10 families included in the survival analysis had 72 pregnancies with 17 infant deaths, 6 child deaths, and 49 survivals at age 10. The mean number of pregnancies per family was 7.2 ± 0.65 and the average progeny survival was 0.766 for infants, 0.894 for children, and 0.679 for survival from birth to 10 years. Longevity was computed from full birth and deaths dates of the offspring of the 13 marriages with children, a total of 75 individuals. The mean longevity was 31.2 years in men and 42.0 in women.

2.2 | Statistical analysis

Multiple regression analyses were used to characterize the effects of the explanatory variables individual's inbreeding coefficient (F), maternal inbreeding coefficient (F_m), and year of birth (YOB) on pre-reproductive survival and longevity. Regression analysis of log-transformed survival was performed because survival is expected to decrease linearly with inbreeding on a logarithmic scale according to the multilocus viability model based on the assumption of multiplicative fitness interactions among loci (Cavalli-Sforza & Bodmer, 1971; Charlesworth & Willis, 2009; Lynch & Walsh, 1998). On this basis, the effect of inbreeding on survival is measured in terms of the inbreeding load defined as the slope of the regression of the natural logarithm of survival on F. Longevity was adjusted for the effects of sex using linear regression (ie, residuals). In order to detect multicollinearity problems in the multiple regressions, values of tolerance and variance inflation factor (VIF) were computed in all regression analyses. Values of tolerance were always higher than 0.20 (ranged from 0.53 to 0.78) and VIF values were less than 5 (varied between 1.29 and 1.88), revealing that there is no apparent multicollinearity in the analyzed dataset. All statistical analyses were performed using IBM SPSS Statistics software system (version 24.0; SPSS Inc. Chicago).

3 | RESULTS

### 3.1 | Inbreeding in the Braganza dynasty

The royal lineage of the House of Braganza was constituted by 14 monarchs: 12 kings and two regnant queens, who contracted a total of 17 marriages (Supporting Information Table S1). Some of the monarchs, particularly Peter II, Peter IV, and Maria II, married more than once. Consanguineous marriages were frequent in the dynasty (Supporting Information Figure S1). There was an uncle-niece marriage, a first-cousin marriage, and several second-cousin marriages. In a total of 17 marriages, 8 (47.06%) showed $\theta \geq 0.0157$ (ie, the kinship value corresponding to a second cousin marriage), so they can be considered consanguineous. Since several of these marriages were consecutive, some of the monarchs presented high F (Figure 1). The mean F of the monarchs was 0.0530, a value close to that corresponding to the progeny of first cousins ($F = 0.0625$).

3.2 | Effects of inbreeding on survival and longevity

Effects of inbreeding on pre-reproductive survival (Supporting Information Table S2) and longevity (Supporting Information Table S3) were detected through multiple regression analyses (Table 1). The effect of F on survival from birth to 10 years was close to statistical significance (b = -2.313 ± 1.475; $P = 0.084$). When the non-significant explanatory variables, F_m and YOB, were removed from the analysis, the simple regression of survival on F was statistically significant (b = -2.714 ± 1.142, $P = 0.023$). A statistically significant effect of F_m on offspring longevity was detected (b = -174.6 ± 96.3, $P = 0.037$), and this effect was found even for individuals older than 10 years (b = -156.3 ± 87.4, $P = 0.040$). The effect of F on longevity was not significant (Table 1).

4 | DISCUSSION

An extraordinary cultural evolution makes our species a different animal (Boyd, 2018). Probably, inbreeding avoidance mechanisms in response to selection against inbred offspring have evolved in most animals (Blouin & Blouin, 1988; Pusey & Wolf, 1996), but mating between relatives is favored in some human populations depending on the cultural context (Bittles, 2012). Inter-royalty marriage, for example, has been used as a method to maintain dynastic power and to form new political alliances. Therefore, it is expected that royal dynasties show high levels of inbreeding. The only European dynasty studied in this sense, the Habsburgs, showed very high levels of inbreeding for both the Austrian and the Spanish branches of the dynasty (mean $F = 0.0790$ and 0.1287, respectively), as well as a decrease in survival, compared with outbred individuals, of 13.5% at
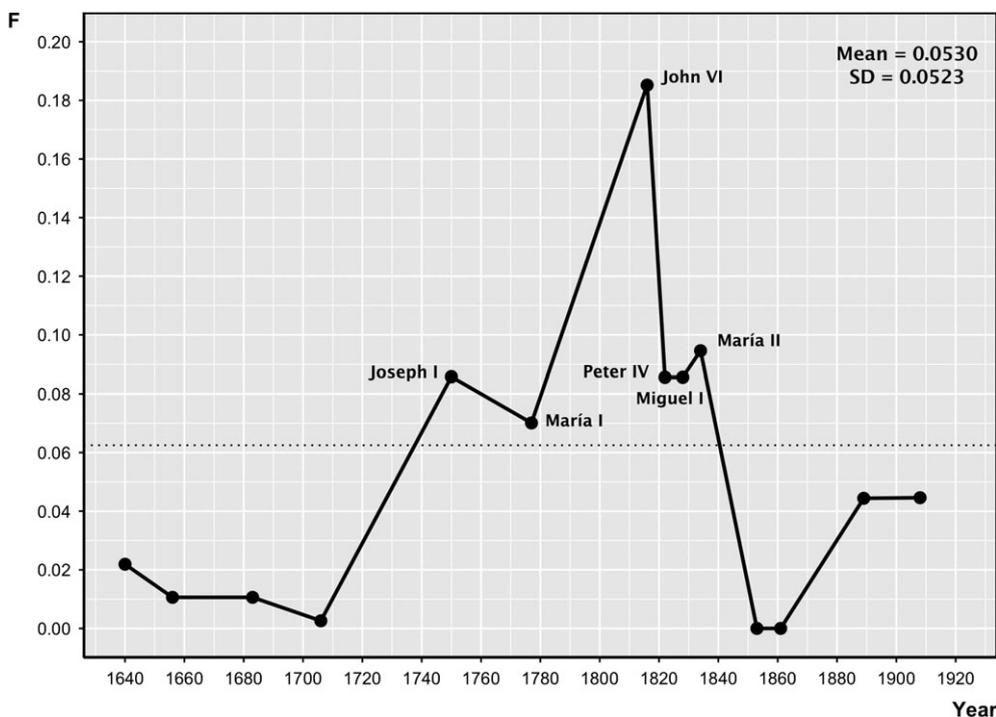
FIGURE 1 Inbreeding coefficient (F) of all the monarchs of the House of Braganza that reigned in Portugal from 1640 to 1910. Only those with the highest values are named. The dotted line represents the level of inbreeding corresponding to progeny of first cousins

the level of inbreeding corresponding to the offspring of first cousins (Ceballos & Álvarez, 2013). The present study shows that, during the second half of the 18th and first half of the 19th centuries, all Braganza monarchs had

TABLE 1 Multiple regression analyses of pre-reproductive survival and longevity as a function of individual's inbreeding coefficient (F), maternal inbreeding coefficient (F_m) and year of birth (YOB)

	Regression coefficient	SE	P
Survival to 10 years			
F	−2.313	1.475	0.084
F_m	−1.112	2.117	0.309
YOB	0.001	0.001	0.260
Infant survival			
F	−1.084	1.908	0.296
F_m	−0.179	2.739	0.475
YOB	−0.000	0.001	0.489
Child survival			
F	−1.230	0.814	0.091
F_m	−0.932	1.169	0.228
YOB	0.001	0.001	0.120
Longevity			
F	−49.6	68.6	0.236
F_m	−174.6	96.3	0.037
YOB	0.112	0.057	0.027
Longevity (>10 years)			
F	17.8	73.2	0.405
F_m	−156.3	87.4	0.040
YOB	0.091	0.049	0.035

Analysis of longevity for individuals older than 10 years is also included.

$F > 0.0625$. In addition, results showed a decrease in survival at a level of $F = 0.0625$ of 10.4%, a value close to that calculated for the Habsburg dynasty (Ceballos & Álvarez, 2013). The adverse effect of inbreeding on survival in the Braganzas was clearly higher than the mean decrease of 3.5% in the offspring of first cousins observed in contemporary human populations (Bittles & Black, 2010).

Interestingly, a significant effect of maternal inbreeding on offspring longevity was detected. On average, a value of $F_m = 0.0625$ led to a decrease in 9.7 years in the lifespan of the progeny. This effect was still found when individuals younger than 10 years were removed from the analysis, so it was not restricted to early life. This effect is clearly noticed in the kings: the Braganza kings with $F_m < 0.0625$ had a mean age of death of 55.5 years while those kings with $F_m > 0.0625$ had a mean longevity of 47.0 years. An effect of maternal inbreeding on offspring traits has been reported in other mammals with complex social system and cognitive abilities (Frère et al., 2010) and with high levels of maternal care (Bérénos et al., 2016; Huisman et al., 2016). In humans, it has been suggested that increased maternal homozygosity provides an unfavorable intrauterine environment that could have adverse effects on the progeny (Tanaka, 1977). Our findings support this hypothesis, although further studies are needed to confirm a significant effect of maternal inbreeding on offspring longevity.

AUTHOR CONTRIBUTIONS

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SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of the article.