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The First Two Detected Cases of Polythelia with Possible Polymastia in the Mediterranean Monk Seal (*Monachus monachus*)

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The Mediterranean monk seal (*Monachus monachus*) populations in the eastern Atlantic consisted of thousands of individuals until the arrival of Europeans in the 14th and 15th centuries (González, 2015). Currently, this seal is one of the rarest mammal species worldwide (Karamanlidis & Dendrinos, 2015), with only two Atlantic populations remaining. One is located at Madeira Archipelago (Portugal), with an estimated population of only 20 individuals (Pires et al., 2019), but in the late 1980s was estimated at only six to eight individuals (Pires & Neves, 2001). The other population, at the Cabo Blanco peninsula (Western Sahara/Mauritania) with 350 individuals estimated (CBD-Habitat, unpub. data, 2021), recovered from a mass die-off that reduced its size to 100 individuals in 1997 (Forcada et al., 1999). Both populations have suffered recent "bottlenecks" (Marchessaux, 1989; Forcada et al., 1999; Pires & Neves, 2001) that have minimized their genetic variability, which was already very low (Pastor et al., 2004). In Cabo Blanco, the species' genetic variability declined further after this mortality. In Madeira, the four individuals analyzed were homozygous with an allelic distribution that suggested a genetically impoverished subpopulation of Cabo Blanco (Dayon et al., 2020). According to the latest genetic studies, individuals from both populations share the same haplotype MM05 (Karamanlidis et al., 2016). With this background, we show the first two cases of polythelia with possible polymastia recorded in two breeding females of this species and hypothesize about its potential consequences.

The Mediterranean monk seal is a Phocidae, but it differs from the other members of this family because these females have four nipples instead of two, with two on each side of the belly with the navel in the center. This trait is shared with Hawaiian monk seals (*Neomonachus schauinslandi*) and bearded seals (*Erignathus barbatus*), which, though an exception among phocids, is common in otariids and odobenids (Riedman, 1990).

Mediterranean monk seals exhibit a maternal-care system that lies between the phocid and otariid patterns, perhaps being closer to the latter (Pastor et al., 2011). Lactation length is much longer (4 mo) than usual among phocids, including the Baikal seal (*Pusa sibirica*), whose pups are weaned in 2 to 3 mo. And, indeed, this species lies at the lower end of the duration range for otariid nursing as the northern fur seal (*Callorhinus ursinus*) and Antarctic fur seal (*Arctocephalus gazella*) have pups weaned at 4 mo old (King, 1983; Riedman, 1990; Boyd, 1991; Pastor et al., 2011; Jefferson et al., 2015). For this reason, monk seals are considered the most primitive members of the Phocidae family and probably still present ancestral mothering and lactation patterns from which the other phocids evolved (Repenning & Ray, 1997).

Having glands that secrete milk to nourish neonatal offspring characterizes all mammals. The number of mammary glands and nipples depends on the number of offspring that females of a certain species must nurse (Koyama et al., 2013). During embryogenesis, nipples arise from a pair of mammary ridges extending along the ventral body wall from mid-axilla to the inguinal area. Extra mammary glands (polymastia) may also arise from these ridges, leading to supernumerary nipples (polythelia). Supernumerary nipples may be unilateral or bilateral (McLaughlin et al., 2008).

Polythelia is a congenital anomaly of the mammary glands wherein there are accessory nipples along the milk line apart from the normal nipples. It is a benign condition with chances of malignancy in the accessory nipples (Arunagiri & Anbalagan, 2016). The term *polythelia* covers a range of eight types, from morphologically and functionally normal additional sinuses (Type 1) to

"polythelia pilosa," a patch of hair only, but with the characteristic that they are all associated with histologically identifiable glandular tissue (see Hsu et al., 2000).

For the first time, two cases of polythelia (supernumerary nipples) with possibility of polymastia (supernumerary mammary glands) has been detected for this species. In November 2016, in the Madeira monk seal population (Portugal), an identified lactating female, "Maminhas," was nursing her pup; she presented three nipples along the right embryonic lactiferous line and two along the left line (Figure 1A). Each nipple was observed with milk suggesting that this female had five functioning mammary glands.

The second case was observed at the Cabo Blanco monk seal colony (Western Sahara/Mauritania) through photo-identification pictures taken of breeding female 2363, "Oca," in March 2020. Although monitored since 2011, and having had at least three pups in 2012, 2014, and 2020, no lactation of this female had been recorded, and it was not possible to know if the 5th nipple was active in milk production (Figure 1B).

There is no proof of polymastia in either of the two cases described, only for polythelia. Without any evidence of the existence of additional glandular tissue by anatomical and physiological analysis, it is impossible to confirm these nipples offered milk—even in the case of the female from Madeira, through whose numerary nipples milk flowed, though those nipples appeared functional. But this does not mean that the phenomenon is accompanied by polymastia as it may happen that the supernumerary nipple is connected to the mammary gland of another nipple, that it has a separate supernumerary gland, that it has no gland, or that it has an abortive one (Martin et al., 2016).

Despite intense monitoring of both populations for more than 20 years and through sophisticated means of obtaining digital images and direct observation, no cases of polythelia, with possible polymastia, had ever been detected in females in either monk seal population. In these, the arrangement of the nipples on the ventral area is easily observable and already visible from 2 mo of age when they shed their neonatal lanugo for a short-haired gray fur.

This phenotypic phenomenon with genomic base and potential heritability could be transmitted with relative probability in such a small population. Considering that the data for each population is $n = 1$, which is not valid for statistical

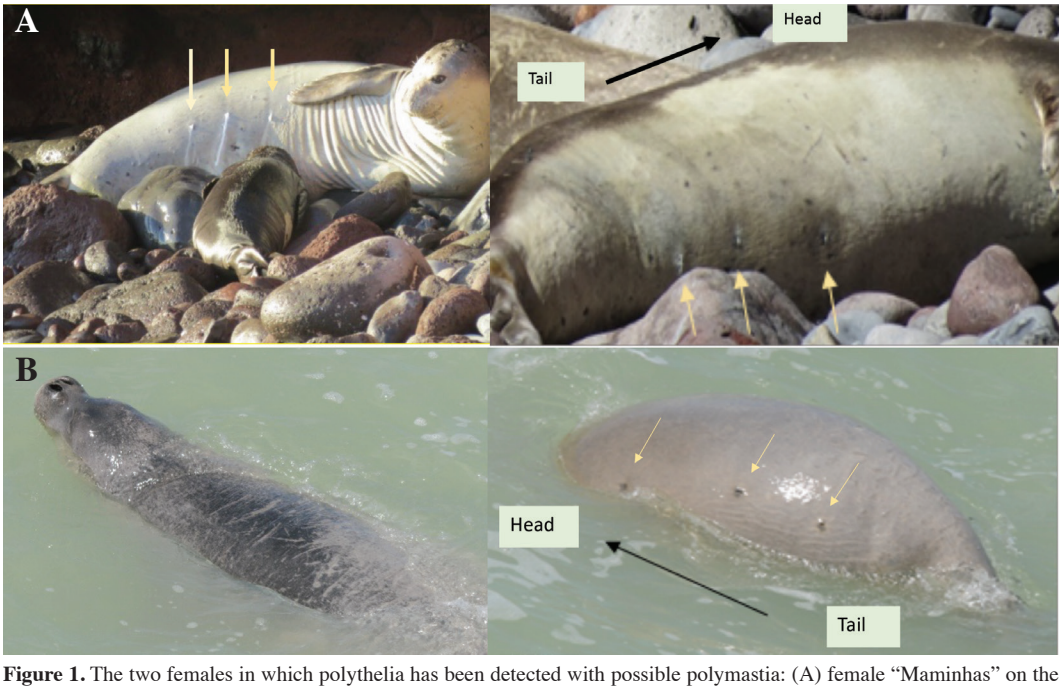

Figure 1. The two females in which polythelia has been detected with possible polymastia: (A) female "Maminhas" on the island of Deserta Grande, Madeira, in 2016, nursing her calf. You can see how a stream of milk comes out of each nipple, which would indicate possible polymastia (yellow arrows). (*Photo credit:* Sergio Pereira and Miguel A. Cedenilla/LIFE MADEIRA MONK SEAL); and (B) female 2363, "Oca," at the Cabo Blanco colony. The three nipples can be clearly seen (yellow arrows). (*Photo credit:* Moulaye Haye/CBD-Habitat)

purposes, and that this phenomenon has only been detected in reproductive females, we dare to infer an incidence rate taking into account the proportion of known breeding females for both populations. The population of Madeira has 13 breeding females, identified in 2019 (*Project Life Madeira Monk Seal Layman's Report*, 2019), which represents a 7.69% incidence of this anomaly. In the Cabo Blanco colony, there are 116 breeding females (CBD-Habitat, unpub. data, 2021), which represents a 0.86% incidence.

We do not know if this condition also affects males—juvenile or adult. In males, the presence of nipples on their external anatomy is undetectable by direct observation and, if present, is likely vestigial. However, from now on, it will be necessary to analyze the phenomenon in male carcasses.

In general, for most mammalian species, polythelia is less conspicuous in males. In addition, the prominence of supernumerary nipples/areolas is hormone dependent and may not appear before puberty, so, in many species, screening is done only in adults (Hsu et al., 2000). Therefore, we are aware of this bias in the sex difference and the possible higher incidence of polythelia in the two monk seal populations.

Supernumerary teats are common in many mammalian species. The association with malignancy is controversial and very rare in the human and animal literature. The frequency of polythelia varies considerably between species and even between breeds. Species used for livestock usually have a high incidence due to genetic management, and the condition might affect animals in terms of causing diseases and mammary infections (Martin et al., 2016). Therefore, given this seal's low genetic variability, its health status must be monitored to evaluate possible anomalies and to take measures, if necessary.

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