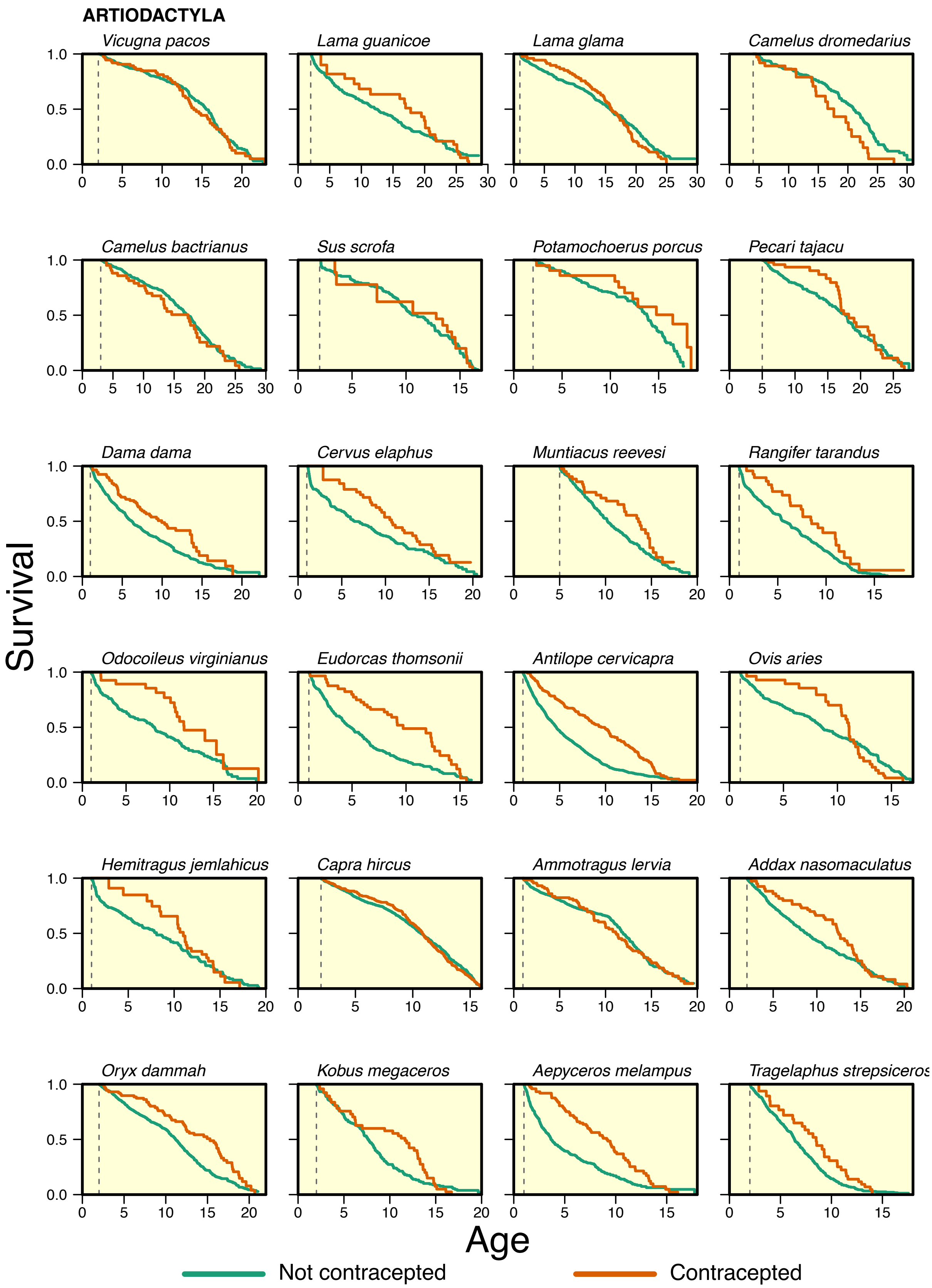
nature portfolio

https://doi.org/10.1038/s41586-025-09836-9

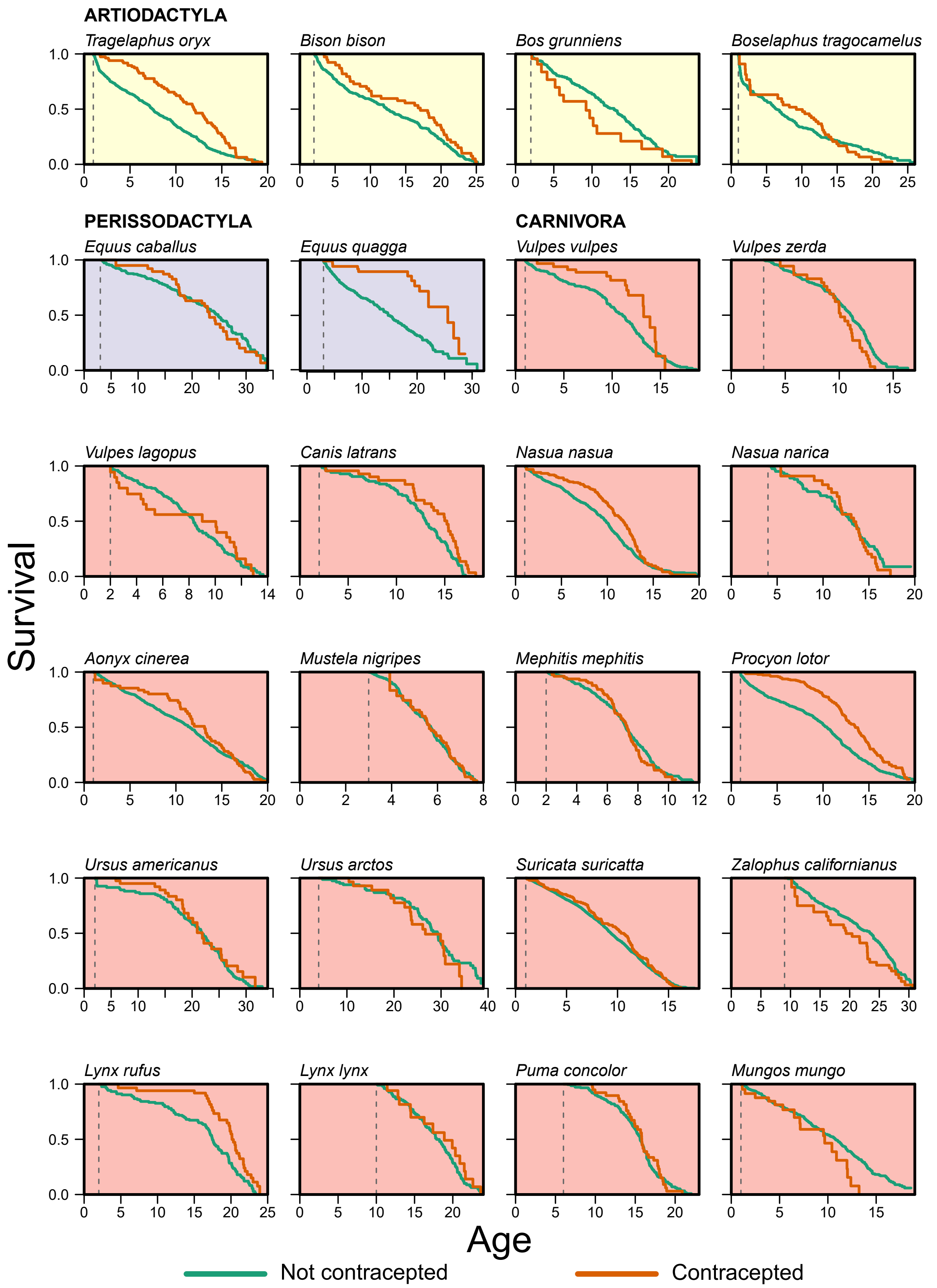
Supplementary information

Sterilization and contraception increase lifespan across vertebrates

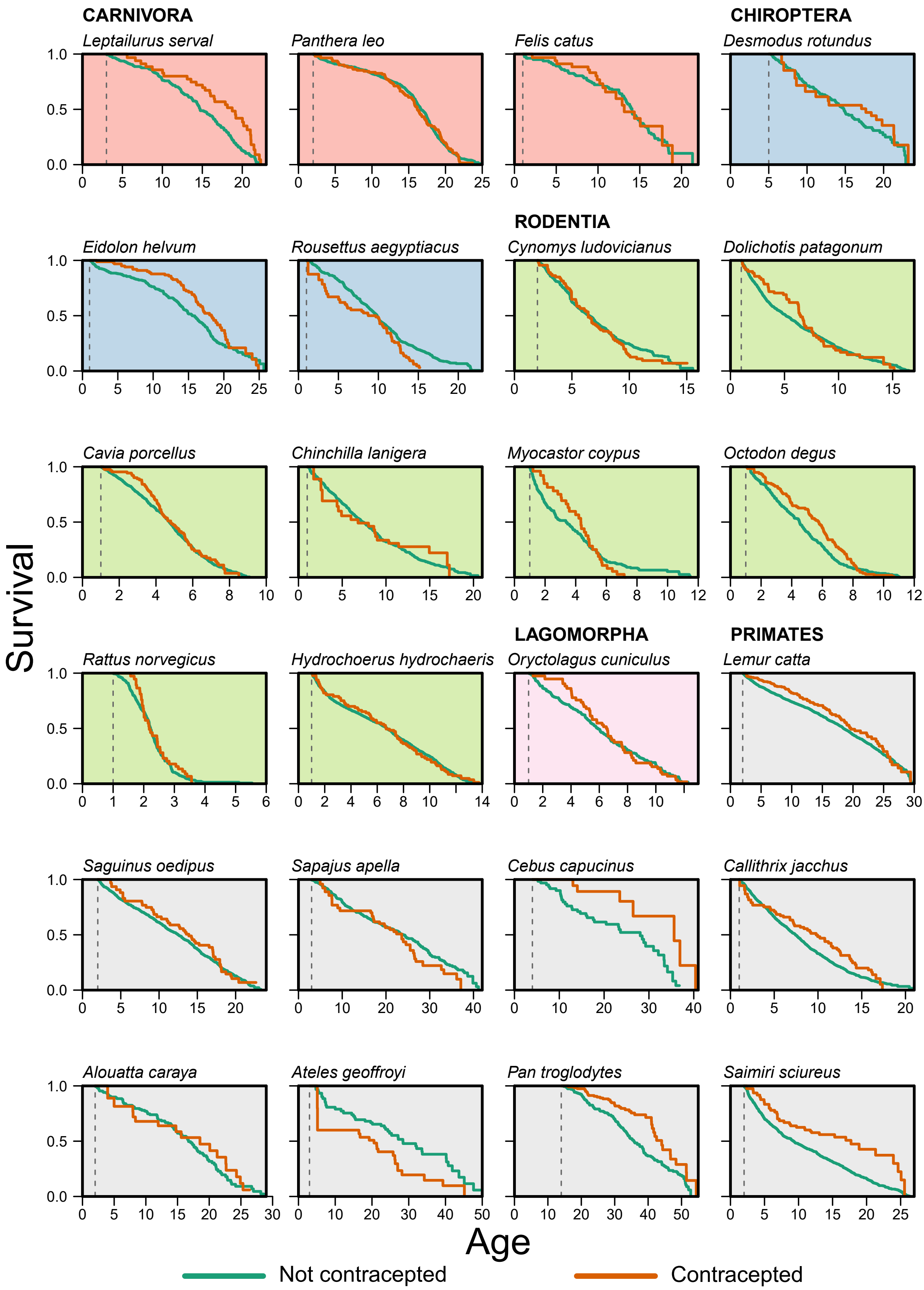
In the format provided by the authors and unedited



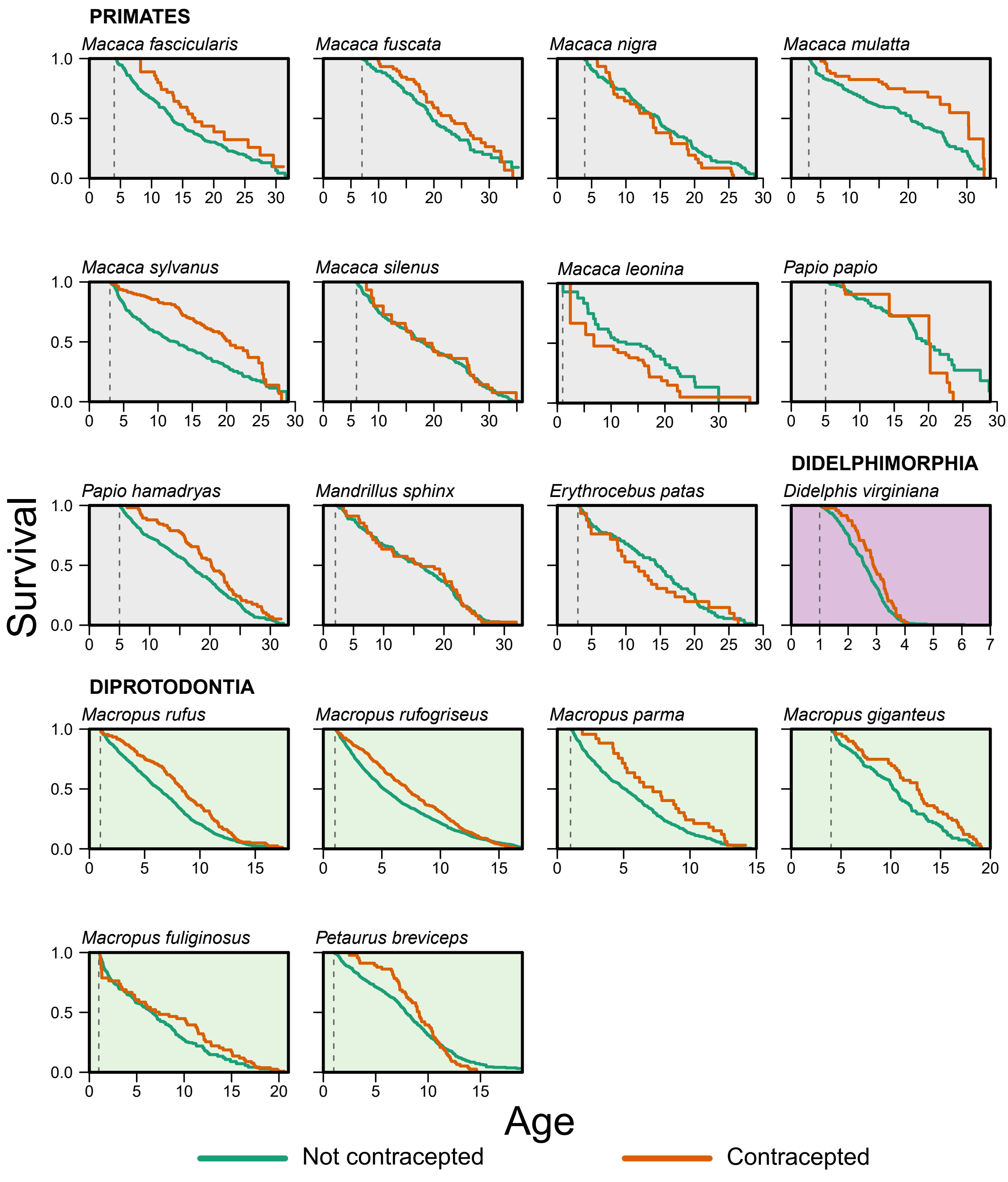
Supplementary Fig 1A. Survival curves for males of individual species that have undergone surgical contraception or have not had contraception. Lines show estimated mean survival at each age. Vertical dashed lines show the age at maturity or minimum age at contraception. Sample sizes for individual species are found in the supplementary information.



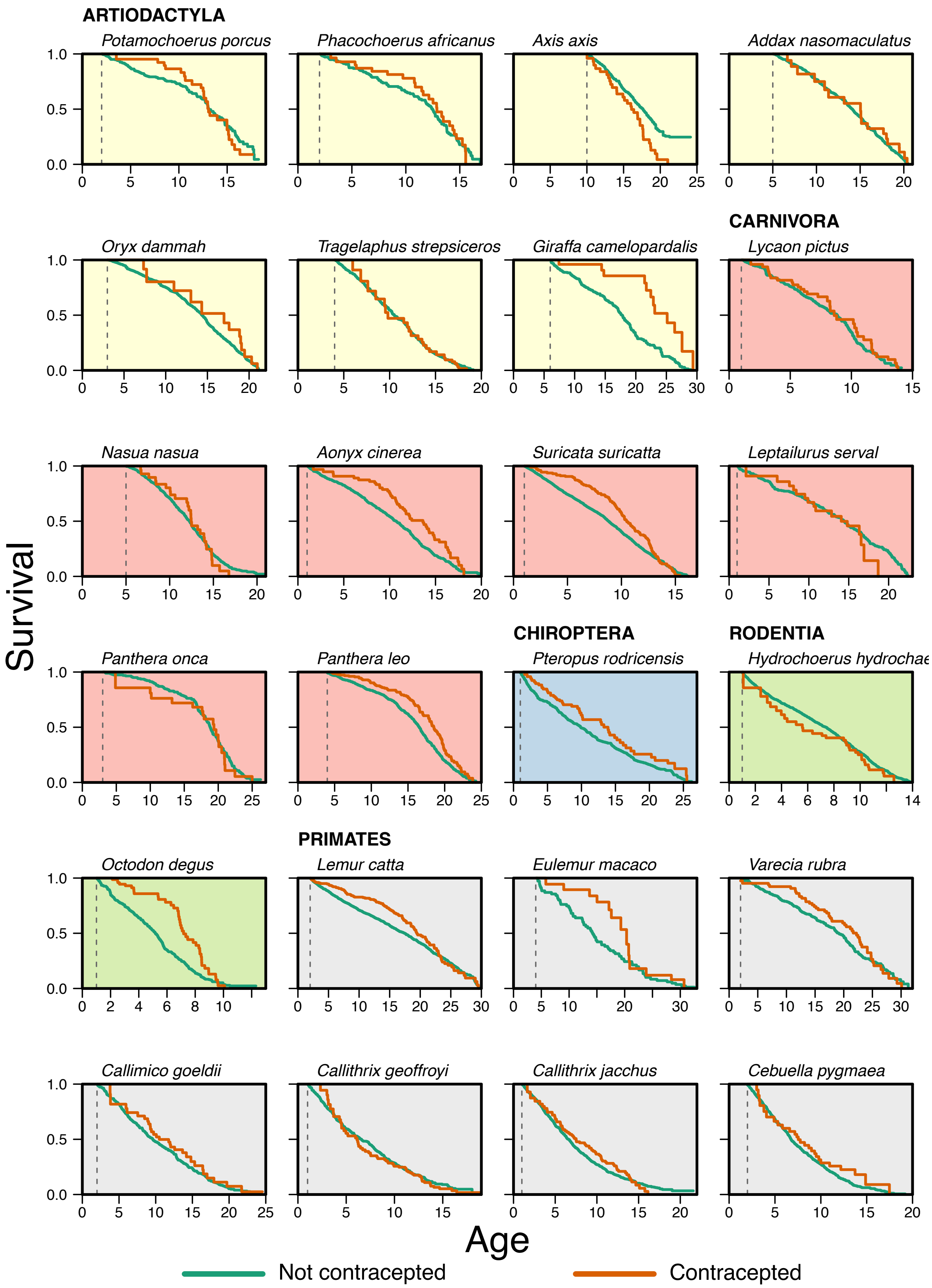
Supplementary Fig 1B. Survival curves for males of individual species that have undergone surgical contraception or have not had contraception. Lines show estimated mean survival at each age. Vertical dashed lines show the age at maturity or minimum age at contraception. Sample sizes for individual species are found in the supplementary information.



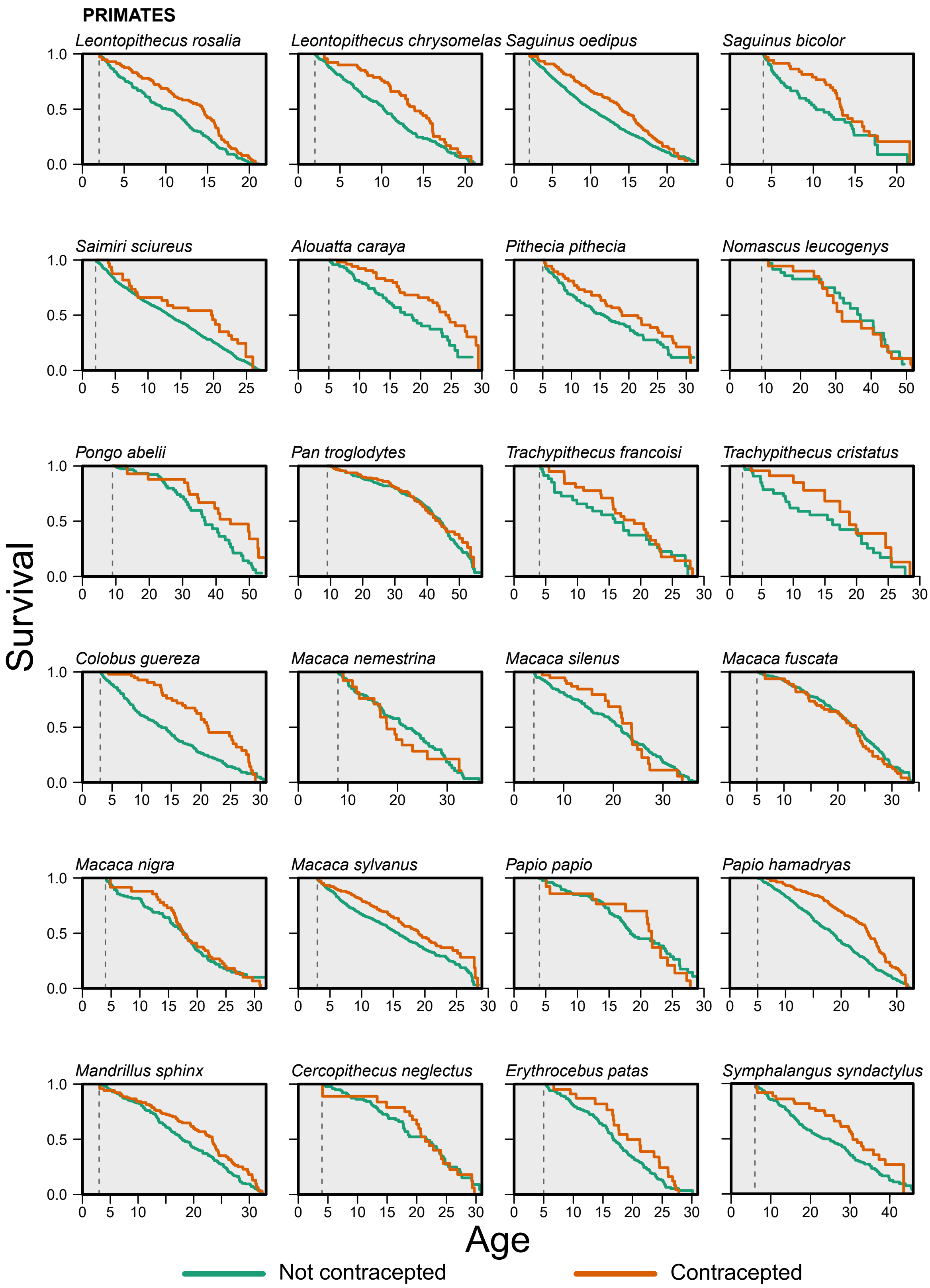
Supplementary Fig 1C. Survival curves for males of individual species that have undergone surgical contraception or have not had contraception. Lines show estimated mean survival at each age. Vertical dashed lines show the age at maturity or minimum age at contraception. Sample sizes for individual species are found in the supplementary information.



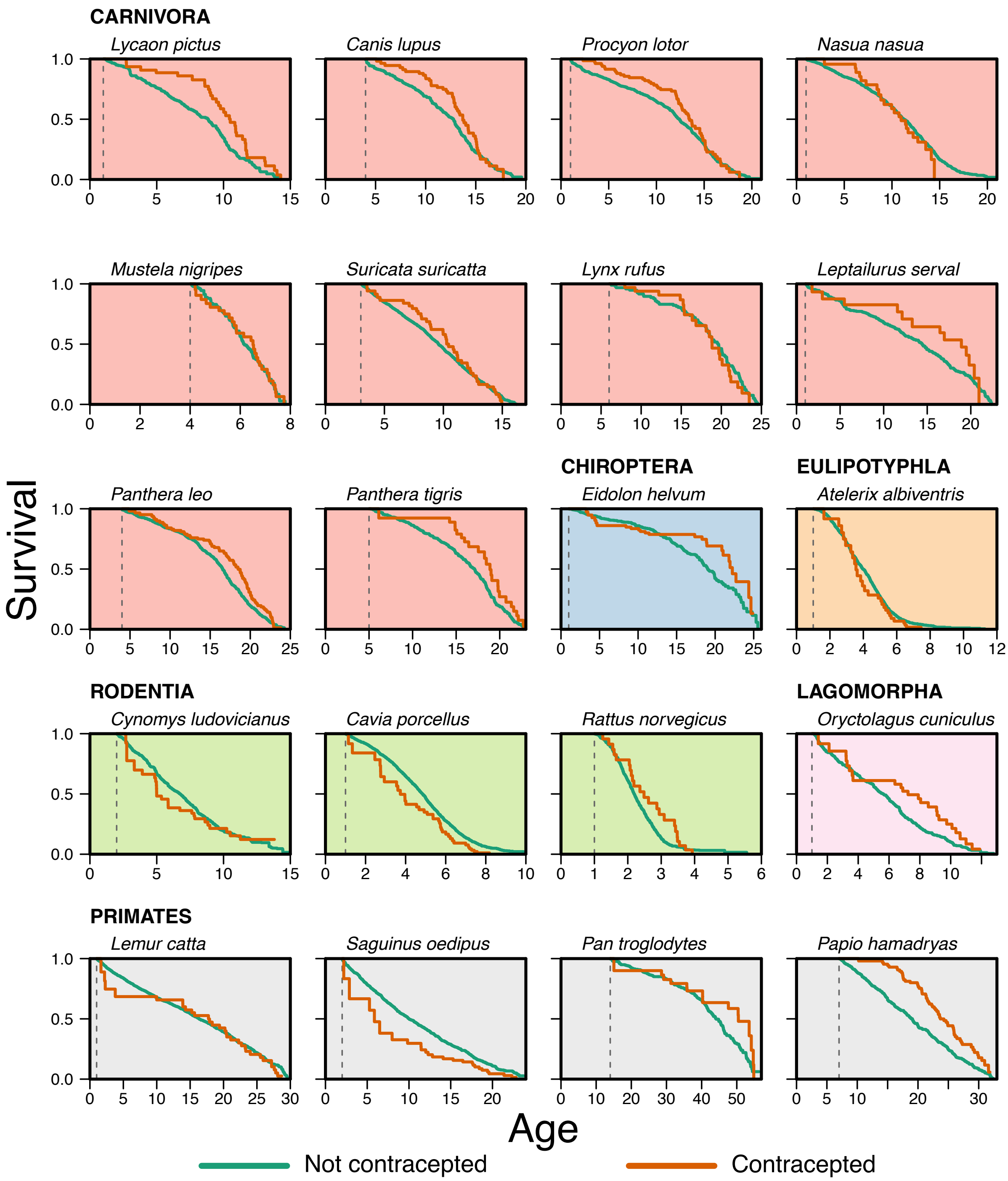
Supplementary Fig 1D. Survival curves for males of individual species that have undergone surgical contraception or have not had contraception. Lines show estimated mean survival at each age. Vertical dashed lines show the age at maturity or minimum age at contraception. Sample sizes for individual species are found in the supplementary information.



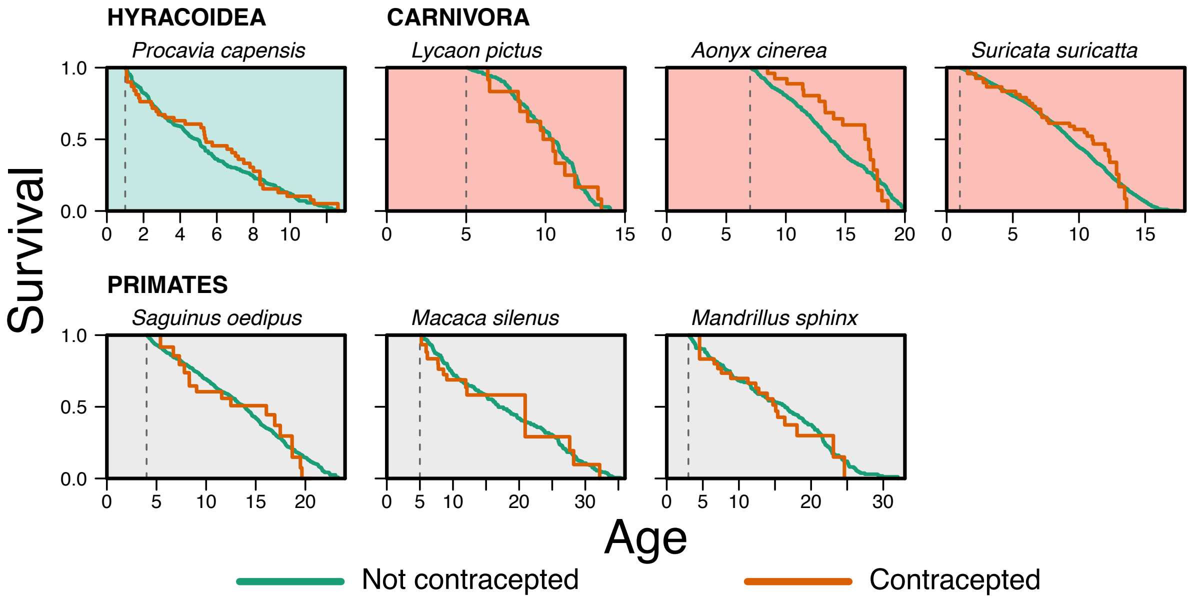
Supplementary Fig 2A. Survival curves for females of individual species that have undergone hormonal contraception or have not had contraception. Lines show estimated mean survival at each age. Vertical dashed lines show the age at maturity or minimum age at contraception. Sample sizes for individual species are found in the supplementary information.



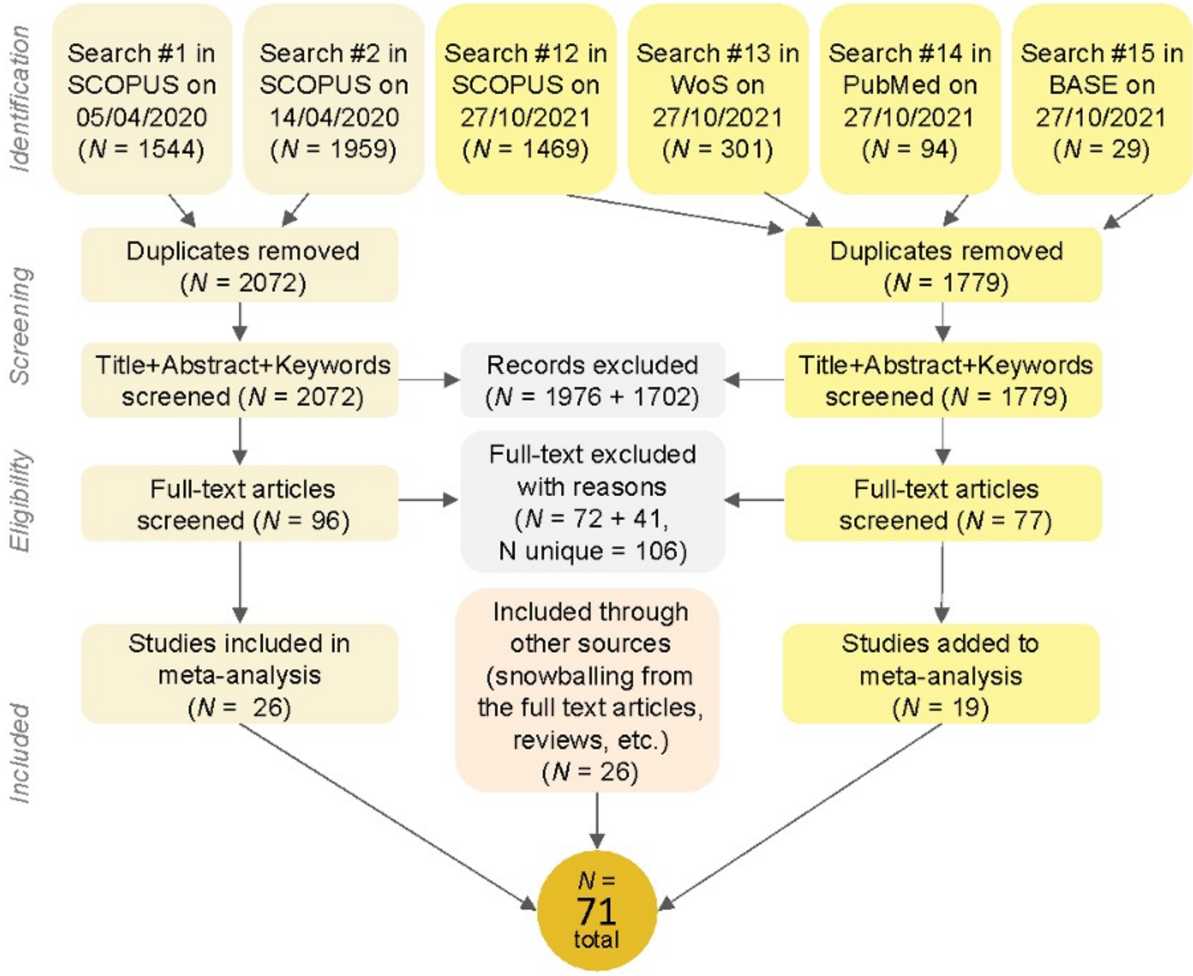
Supplementary Fig 2B. Survival curves for females of individual species that have undergone hormonal contraception or have not had contraception. Lines show estimated mean survival at each age. Vertical dashed lines show the age at maturity or minimum age at contraception. Sample sizes for individual species are found in the supplementary information.



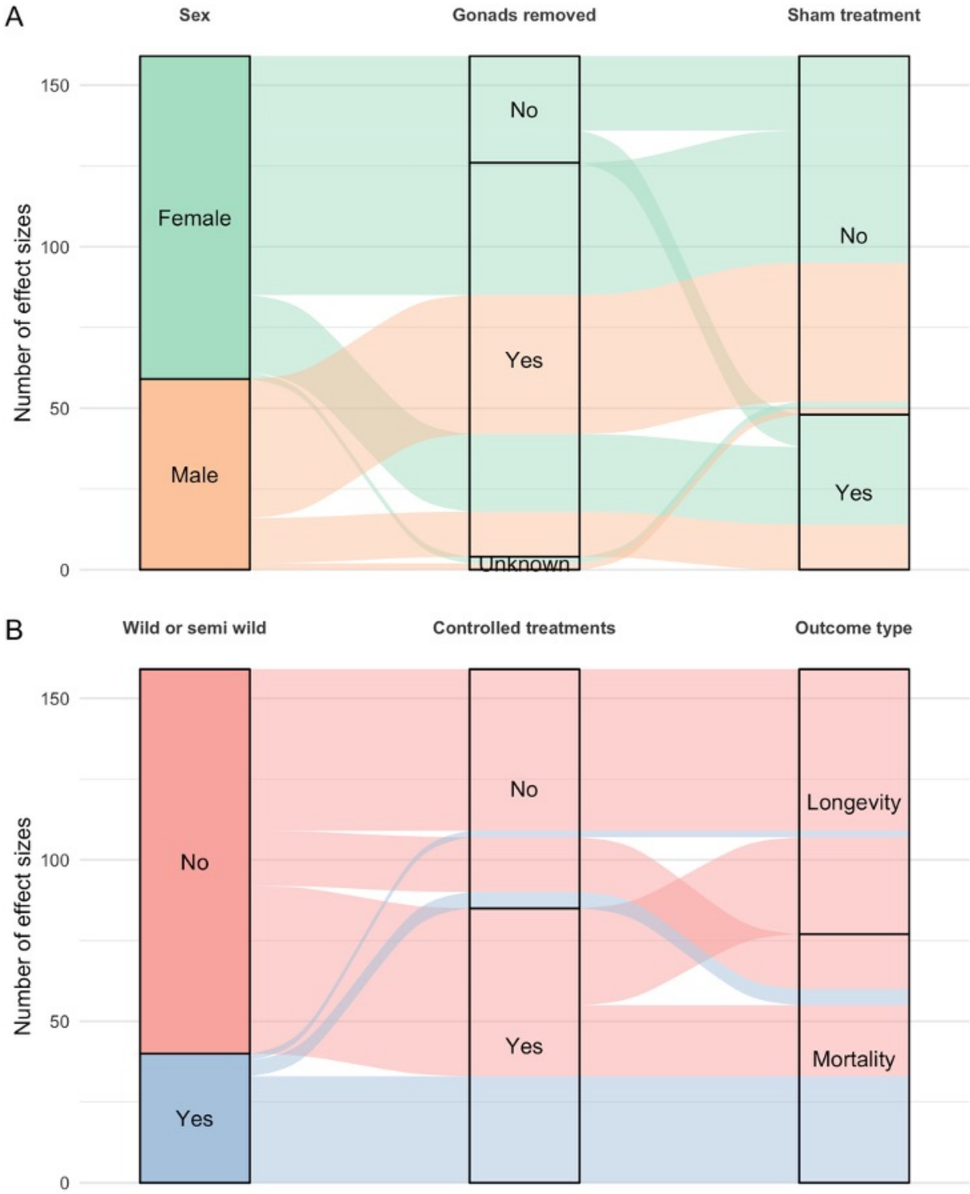
Supplementary Fig 3. Survival curves for females of individual species that have undergone surgical contraception or have not had contraception. Lines show estimated mean survival at each age. Vertical dashed lines show the age at maturity or minimum age at contraception. Sample sizes for individual species are found in the supplementary information.



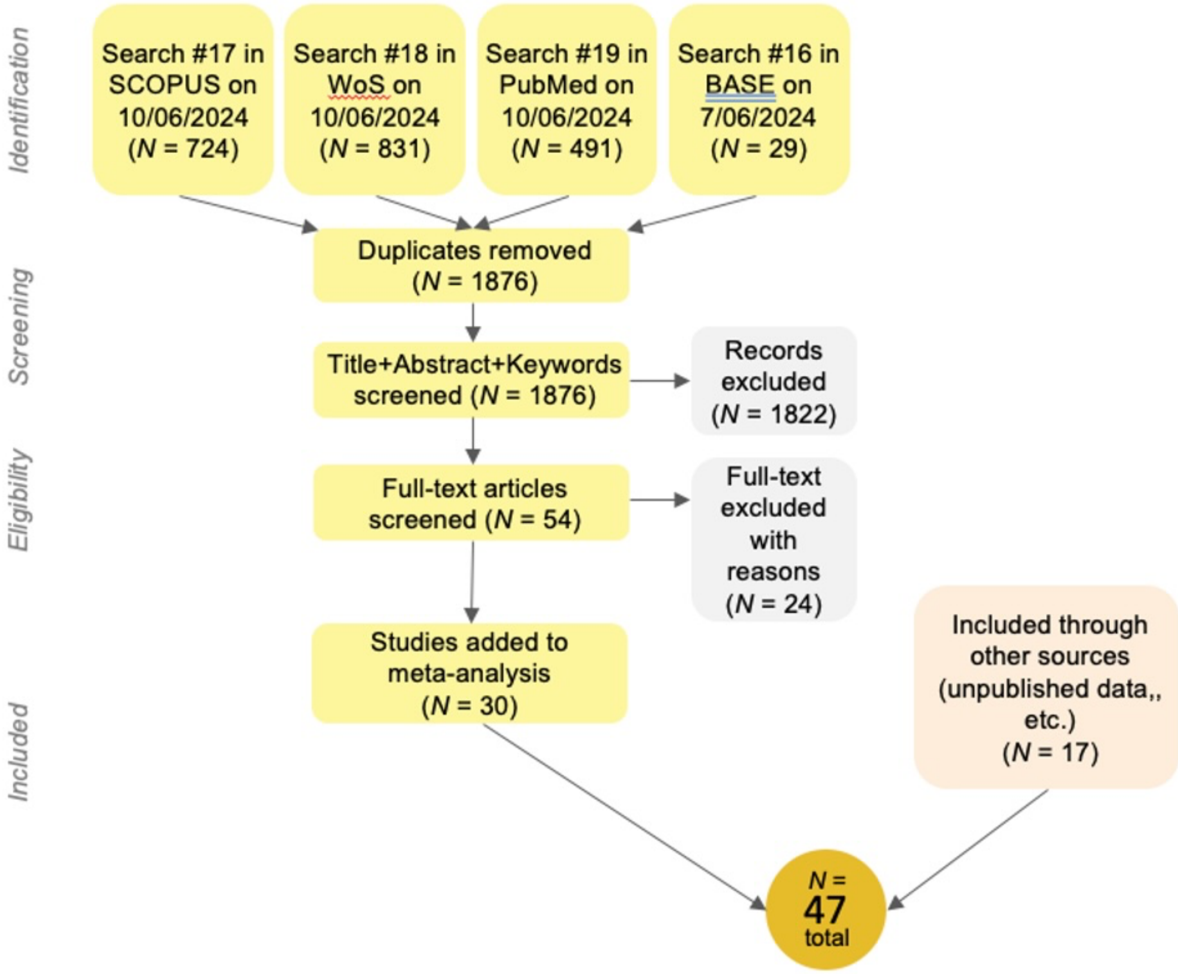
Supplementary Fig 4. Survival curves for males of individual species that have undergone hormonal contraception or have not had contraception. Lines show estimated mean survival at each age. Vertical dashed lines show the age at maturity or minimum age at contraception. Sample sizes for individual species are found in the supplementary information.



Supplementary Fig 5. PRISMA diagram of literature searches and screening for meta-analysis of published literature on changes in survival with sterilization. N represents the number of references in the upper half of the diagram and the number of full-text studies in the bottom half.



Supplementary Fig 6. Overlaps between pairs of six categorical moderators used in the meta-analysis of published literature of changes in lifespan with sterilization. The chart shows the overlap between effect sizes that are used across different moderators in the meta-analysis, in relation to sex-differences, removal of gonads and sham surgery (A) and for the environmental condition that animals were exposed to, whether the experiment was controlled or not and the type of mortality outcome that was assessed (B).



Supplementary Fig 7. PRISMA diagram of literature searches and screening for meta-analysis of published literature on changes in healthspan with gonadectomy. N represents the number of references that were screened at each stage of the meta-analysis search, screening and eligibility assessment.

Supplementary Table 1. Information on use of castration and vasectomy for male surgical sterilization in zoo-housed mammals. Species were classified into their likelihood of using castration or vasectomy using two search approaches. First, reproductive managers and veterinary staff at zoos that are members of the Species360 were contacted and asked to provide feedback on the use of different methods of surgical sterilization for the different species included in the dataset. On the basis of this feedback, and their own knowledge, the veterinary team that are coauthors on this manuscript classified species into different groups according to the likely use of different methods of sterilization. A literature search for published articles and internet sources for different methods of sterilization that are used at a species or taxonomic level (for example, across all ungulates) was also conducted to determine evidence of the application of different types of sterilization in the listed species. The combined searches show that castration is used in the majority of instances. For cross-species comparison species were classified according to whether they were likely castrated, vasectomized, or whether a mix of both approaches could be used.

Species	Likelihood of Castration or Vasectomy	Expert opinion from zoo community	Sexual characteristics	Literature search	Reference	Literature reference link
Kobus megaceros	Likely castration but could be mixed	Castration most common in ungulates, but bovid species in which only male grow horns can be vasectomy used to let horns grow normally	Horns only in males	Castration common for ungulates (see ref) but vasectomy might be used in males to maintain horns (information at the level of ungulates)	Lewis K, Parker MO, Proops L, McBride SD. Risk factors for stereotypic behaviour in captive ungulates. Proc Biol Sci. 2022 Sep 28;289(1983):20221311.	

Tragelaphus strepsiceros	Likely castration but could be mixed	Castration most common in ungulates, but bovid species in which only male grow horns vasectomy used to let horns grow normally	Horns only in males	Castration common but vasectomy might be used in males to maintain horns	News article (accessed 09/03/2025)	https://www.elpasotimes.com/story/news/local/el-paso/2015/09/23/antelope-dies-during-vasectomy-el-paso-zoo/72708562/
Eudorcas thomsonii	Likely castration but could be mixed	Castration most common in ungulates, but bovid species in which only male grow horns vasectomy used to let horns grow normally	Large horns	Castration common but vasectomy might be used in males to maintain horns (at the level of ungulates)		
Boselaphus tragocamelus	Likely castration but could be mixed	Castration more common in most ungulates	Small horns only in males	Castration common but vasectomy might be used in males to maintain horns (at the level of ungulates)		
Ammotragus lervia	Likely castration but could be mixed	Castration more common in most ungulates	Large horns	Castration common but vasectomy might be used in males to maintain horns (at the level of ungulates)		
Hemitragus jemlahicus	Likely castration but could be mixed	Castration more common in most ungulates	Large horns	Castration common but vasectomy might be used in males to maintain horns (at		

				the level of ungulates)
Addax nasomaculatus	Likely castration but could be mixed	Castration more common in most ungulates	Large horns	Castration common but vasectomy might be used in males to maintain horns (at the level of ungulates)
Oryx dammah	Likely castration but could be mixed	Castration more common in most ungulates	Large horns	Castration common but vasectomy might be used in males to maintain horns (at the level of ungulates)
Tragelaphus oryx	Likely castration but could be mixed	Castration more common in most ungulates	Large horns	Castration common but vasectomy might be used in males to maintain horns (at the level of ungulates)
Aepyceros melampus	Likely castration but could be mixed	Castration more common in most ungulates	Large horns	Castration common but vasectomy might be used in males to maintain horns (at the level of ungulates)
Antilope cervicapra	Likely castration but could be mixed	Castration most common in ungulates, but bovid species in which only male grow horns vasectomy used to	Horns only in males	Castration common but vasectomy might be used in males to maintain horns (at the level of ungulates)

		let horns grow normally			
Bos grunniens	Likely castration but could be mixed	Castration more common in most ungulates	Large horns	Lots of reference to Yaks being castrated (Dorji et al. 2022)	Dorji, N., Derks, M., Koerkamp, P. G., & Bokkers, E. A. M. (2022). Welfare and management practices of free-ranging yaks (Bos grunniens) in Bhutan. Animal Welfare, 31(4), 467-481.
Ovis aries	Likely castration but could be mixed	Castration more common in most ungulates	Horns depend on breed	Very likely to be castrated as a domestic species but vasectomy used to create teaster males for agricultural purposes	
Capra hircus	Likely castration but could be mixed	Castration more common in most ungulates	Horns depend on breed	Very likely to be castrated as a domestic	
Bison bison	Likely castration but could be mixed	Castration more common in most ungulates	Horns	Likely castrated as widely discussed in McCorkell et al in relation to other	Church, J., Galbraith, J., Rob McCorkell, D.

				cattle. No mention of vasectomy.	V. M., Rioja-Lang, F. C., & Silzer, M. (2016). Code of practice for the care and handling of bison: Review of scientific research on priority issues.	
Camelus dromedarius	Likely castration but could be mixed	Castration more common in most ungulates		Castration common in lamas, alpacas and camels	FAO website (accessed 09/03/2025)	https://www.fao.org/4/T0690E/t0690e09.htm
Lama glama	Likely castration but could be mixed	Castration more common in most ungulates		Castration common in lamas, alpacas and camels	FAO website (accessed 09/03/2025)	https://www.fao.org/4/T0690E/t0690e09.htm
Camelus bactrianus	Likely castration but could be mixed	Castration more common in most ungulates		Castration common in lamas, alpacas and camels	FAO website (accessed 09/03/2025)	https://www.fao.org/4/T0690E/t0690e09.htm
Vicugna pacos	Likely castration but could be mixed	Castration more common in most ungulates		Castration common in lamas, alpacas and camels	FAO website (accessed 09/03/2025)	https://www.fao.org/4/T0690E/t0690e09.htm
Lama guanicoe	Likely castration but could be mixed	Castration more common in most ungulates		Castration common in lamas, alpacas and camels	FAO website (accessed 09/03/2025)	https://www.fao.org/4/T0690E/t0690e09.htm
Rangifer tarandus	Likely castration but could be mixed	Mixed feedback. Vasectomy might be preferred to manage antler/velvet issues, but might depend on age	Antlers in males and females	Likely castrated as difficult to handle otherwise during the rut. Review of reindeer populations in Switzerland shows	Luginbühl, C., Gross, J., Wenker, C., Hoby, S., Basso, W., & Zanolari, P.	

				only castrated individuals (Luginbul et al 2023). Also evidence of this as a main management strategy associated with domestication	(2023). Reindeer Husbandry in Switzerland — Management, Feeding, and Endoparasite Infections. Animals, 13(9), 1444.	
Odocoileus virginianus	Likely castration but could be mixed	Mixed feedback. Vasectomy might be preferred to manage antler/velvet issues, but might depend on age	Antlers in males	Both possible. Vasectomy done on wild populations and maintains antlers but males can be dangerous in captivity without castration	Deer Industry Association Australia factsheet (accessed 09/03/2005)	
Dama dama	Likely castration but could be mixed	Mixed feedback. Vasectomy might be preferred to manage antler/velvet issues, but might depend on age	Antlers in males	Both possible. Vasectomy done on wild populations and maintains antlers but males can be dangerous in captivity without castration	Deer Industry Association Australia factsheet (accessed 09/03/2005)	
Cervus elaphus	Likely castration but could be mixed	Mixed feedback. Vasectomy might be preferred to manage antler/velvet issues, but might depend on age	Antlers in males	Both possible. Vasectomy done on wild populations and maintains antlers but males can be dangerous in	Deer Industry Association Australia factsheet (accessed 09/03/2005)	https://www.deerfarming.com.au/wp-content/uploads/2016/11/14-Castration.pdf

				captivity without castration.	
Muntiacus reevesi	Likely castration but could be mixed	Mixed feedback. Vasectomy might be preferred to manage antler/velvet issues, but might depend on age	Antlers in males	Vasectomy done on wild populations and maintains antlers but males can be dangerous in captivity without castration.	Deer Industry Association Australia factsheet (accessed 09/03/2005)
Sus scrofa	Likely castration but could be mixed	Castration more common in most ungulates	Mane, larger size and prominent canines in males	Castration commonly done in pigs.	
Potamochoerus porcus	Likely castration but could be mixed	Castration more common in most ungulates		Likely to be castrated like other members of Suidae.	
Pecari tajacu	Likely castration but could be mixed	Castration more common in most ungulates		Evidence of some populations being castrated.	Lewis K, Parker MO, Proops L, McBride SD. Risk factors for stereotypic behaviour in captive ungulates. Proc Biol Sci. 2022 Sep 28;289(1983):20221311.

Vulpes zerda	Likely castration but could be mixed	Castration preferred in carnivores (except lions)		Neutering in foxes commonly referred to, but it is also possible that they would be vasectomized. Referred to in canid management guide (Boutelle and Bertschinger 2010)	Boutelle, S. M., & Bertschinger, H. J. (2010). Reproductive management in captive and wild canids: contraception challenges. In ternational Zoo Yearbook, 44(1), 109-120.
Vulpes lagopus	Likely castration but could be mixed	Castration preferred in carnivores (except lions)		Neutering in foxes commonly referred to but it is also possible that they would be vasectomized. Referred to in canid management guide (Boutelle and Bertschinger 2010)	Boutelle, S. M., & Bertschinger, H. J. (2010). Reproductive management in captive and wild canids: contraception challenges. In ternational Zoo Yearbook, 44(1), 109-120.
Vulpes vulpes	Likely castration but could be mixed	Castration preferred in carnivores (except lions)		Neutering in foxes commonly referred to but it is also possible that they would be	Boutelle, S. M., & Bertschinger, H. J. (2010). Reproductive

				vasectomized. Referred to in canid management guide (Boutelle and Bertschinger 2010)	management in captive and wild canids: contraception challenges. In ternational Zoo Yearbook, 44(1), 109-120.	
Canis latrans	Likely castration but could be mixed	Castration preferred in carnivores (except lions)		Castration because of relatedness to domestic dogs, but vasectomy is possible to maintain group behaviour		
Leptailurus serval	Castration	Castration preferred in carnivores (except lions)		Servals castrated as pets to stop spraying (see online pet guide)	Online pet guide (accessed 09/03/2025)	https://exoticpetwonderland.org/exotic-pet-care-guides/pet-serval-care-guide/#:~:text=Spaying%20and%20Neutering%20Servals,-Spaying%20or%20neutering&text=This%20cuts%20down%20on%20hormonal,marking%20should%20still%20be%20expected
Felis catus	Castration	Castration preferred in carnivores (except lions)		Domestic cat where castration is the means of neutering		
Lynx rufus	Castration	Castration preferred in carnivores (except lions)		Commonly neutered when kept as exotic pets. Also reference to neutered bobcats	Urban edge wildlife guide (accessed 09/03/2025)	https://exoticpetwonderland.org/exotic-pet-care-guides/pet-bobcat-care-

				in zoos (i.e. Brevard zoo website; big cat rescue, urban edge wildlife)		guide/#elementor-toc__heading-anchor-7
Lynx lynx	Likely castration but could be mixed	Castration preferred in carnivores (except lions)		Castration described method of choice for surplus males but vasectomy described (Krelekamp husbandry guide)	Krelekamp, C. J. (2004). Husbandry guidelines Eurasian lynx (Lynx lynx sspp.). Europe an Association of Zoos and Aquaria (EAZA). Amsterdam, Netherlands, 9-34.	
Puma concolor	Likely castration but could be mixed	Mostly castration		Vasectomy is the most common means of contraception (Deco-Souza et al 2024)	Deco-Souza, T., Cazati, L., Requena, L. A., Jorge-Neto, P. N., Polizelle, S. R., Caramalac, S. M., ... & de Araujo, G. R. (2024). Laparoscopic tubal ligature in pumas (Puma	

					concolor): A technique allied with conservation. Theriogenology Wild, 4, 100083.	
Panthera leo	Likely vasectomized	Most likely vasectomy	Mane in males	Vasectomy preferred to maintain mane		
Mungos mungo	Castration	Castration		Vasectomy not recommended in mongoose due to effects on females (Mongoose care manual)	Mongoose care manual (accessed 09/03/2025)	https://nagonline.net/wp-content/uploads/2014/05/MustelidCareManual2010-NAG-EDIT.pdf
Suricata suricatta	Likely castration but could be mixed	Castration		Vasectomy maybe preferred to avoid disturbing group behaviour (RVC)	Royal veterinary college meerkat care guide (accessed 09/03/2025)	https://www.rvc.ac.uk/Media/Default/Beaumont%20Sainsbury%20Animal%20Hospital/EXOTICS/Animal%20Care%20Factsheets/Meerkat-care-and-advice-Dec-2022.pdf
Mephitis mephitis	Castration	Castration preferred in carnivores (except lions)		Skunks typically neutered as pets. Vasectomy not recommended in mustelids due to the prolonged pseudopregnancy that is induced in females (Mustelid care manual, 2010)	Mustelid care manual (accessed 09/03/2025)	https://nagonline.net/wp-content/uploads/2014/05/MustelidCareManual2010-NAG-EDIT.pdf

Mustela nigripes	Likely castration but could be mixed	Castration preferred in carnivores (except lions)		vasectomy not recommended in mustelids although pet owners sometimes vasectomize to bring female out of season (https://www.rspca.org.uk/adviceandwelfare/pets/ferrets/health/neutering)	RSPCA guide (accessed 09/03/2025)	https://www.rspca.org.uk/adviceandwelfare/pets/ferrets/health/neutering
Aonyx cinereus	Likely castration but could be mixed	Castration preferred in carnivores (except lions)		No info		
Zalophus californianus	Castration	Castration preferred in carnivores (except lions)		Three papers referring to castrated sea lions in different studies from zoos. No mention of vasectomy can be found	Brochon J, Coureaud G, Hue C, Crochu B, Charrier I. Odor discrimination in terrestrial and aquatic environments in California sea lions (Zalophus californianus) living in captivity. Physiol Behav. 2021 Jun	

					1;235:113408. doi: DOI: 10.1016/j.physbeh.2021.113408; Kastelein, R. A., et al. "Food consumption and growth of California sea lions (Zalophus californianus californianus)." Zoo Biology: Published in affiliation with the American Zoo and Aquarium Association 19.2 (2000): 143-159
Nasua narica	Castration	Castration preferred in carnivores (except lions)		Castrated as a common means of contraception in zoos and to reduce aggression as pets	Mayol, J. 2019. Information on measures and related costs in relation to species

				included on the Union list: Nasua nasua. Technical note prepared by IUCN for the European Commission.	
Nasua nasua	Castration	Castration preferred in carnivores (except lions)	Castrated as a common means of contraception in zoos and to reduce aggression as pets	Mayol, J. 2019. Information on measures and related costs in relation to species included on the Union list: Nasua nasua. Technical note prepared by IUCN for the European Commission.	
Procyon lotor	Castration	Castration preferred in carnivores (except lions)	Sounds like castration very common for behavioural reasons	Exotic pet care (accessed 09/03/2025)	https://exoticpetwonderland.org/exotic-pet-care-guides/pet-raccoon-care-guide/#:~:text=fleas%20in%20raccoons.-,Spaying%20and%20Neutering%20Raccoons,good%20age%20range%20for%20sterilization.

Ursus arctos	Likely castration but could be mixed	Can be vasectomized when young then castrated as adults		Castration used if you don't want to induce ovulation in females but vasectomy also mentioned (see bear sanctuaries standard guide)	Bear standard guide (accessed 09/03/2025)	https://sanctuaryfederation.org/wp-content/uploads/2020/02/Bear-Standards-2019.pdf
Ursus americanus	Likely castration but could be mixed	Can be vasectomized when young then castrated as adults		Castration used if you don't want to induce ovulation but vasectomy also mentioned (vet team said both can be conducted on the same animal; also see bear sanctuaries standard guide)	Bear standard guide (accessed 09/03/2025)	https://sanctuaryfederation.org/wp-content/uploads/2020/02/Bear-Standards-2019.pdf
Desmodus rotundus	Likely castration but could be mixed	Castration most likely		No info		
Rousettus aegyptiacus	Likely castration but could be mixed	Castration most likely		No info		
Eidolon helvum	Likely castration but could be mixed	Castration most likely		There are reports of castrated bats in facilities	Carter G, Leffer L. Social Grooming in Bats: Are Vampire Bats Exceptional? PLoS One. 2015 Oct 7;10(10):e01	

					38430. doi: 10.1371/journal.pone.0138430. PMID: 26445502
Didelphis virginiana	Castration	Castration		Castration is recommended for opossum (described in Exotic small mammal care and husbandry, Banks et al 2016)	Banks, R. E., Sharp, J. M., Doss, S. D., & Vanderford, D. A. (2013). Exotic small mammal care and husbandry. John Wiley & Sons.
Notamacropus parma	Castration	Castration		Orchidectomy the method of choice for macropods (Diaz et al 2023)	Díaz-Portalatín, N., Sadar, M. J., Anderson, K., & Brandão, J. (2023). Orchiectomy and scrotal ablation using a bipolar electrothermal sealing device in macropods: Four cases. Journal of Exotic Pet

					Medicine, 47, 34-38.
Macropus fuliginosus	Castration	Castration		Orchidectomy the method of choice for macropods (Diaz et al 2023)	Díaz-Portalatín, N., Sadar, M. J., Anderson, K., & Brandão, J. (2023). Orchiectomy and scrotal ablation using a bipolar electrothermal sealing device in macropods: Four cases. Journal of Exotic Pet Medicine, 47, 34-38.
Macropus giganteus	Castration	Castration		Orchidectomy the method of choice for macropods (Diaz et al 2023)	Díaz-Portalatín, N., Sadar, M. J., Anderson, K., & Brandão, J. (2023). Orchiectomy and scrotal ablation using a bipolar electrothermal sealing

				device in macropods: Four cases. Journal of Exotic Pet Medicine, 47, 34-38.
Osphranter rufus	Castration	Castration	Orchidectomy the method of choice for macropods (Diaz et al 2023)	Díaz-Portalatín, N., Sadar, M. J., Anderson, K., & Brandão, J. (2023). Orchiectomy and scrotal ablation using a bipolar electrothermal sealing device in macropods: Four cases. Journal of Exotic Pet Medicine, 47, 34-38.
Notamacropus rufogriseus	Castration	Castration	Orchidectomy the method of choice for macropods (Diaz et al 2023)	Díaz-Portalatín, N., Sadar, M. J., Anderson, K., & Brandão, J. (2023). Orchiectomy

					and scrotal ablation using a bipolar electrothermal sealing device in macropods: Four cases. Journal of Exotic Pet Medicine, 47, 34-38.
Petaurus breviceps	Castration	Castration		Commonly castrated to remove male odor (Isaza 2020)	Isaza, N., & Isaza, R. (2020). Neutering Procedures and Considerations in Rabbits and Other Small Mammals. High-Quality, High-Volume Spay and Neuter and Other Shelter Surgeries, 295-323.
Oryctolagus cuniculus	Castration			Domestic species commonly castrated	

Equus caballus	Likely castration but could be mixed	Castration more common in most ungulates		Domestic species commonly castrated	
Equus quagga	Likely castration but could be mixed	Castration more common in most ungulates		Castration commonly used to control captive zebra populations (Martinez-Dias)	Martínez-Boví, R., Barba, M., Plaza-Dávila, M., Barja, I., Carbonell, M. D., Gerique, C., & Guillén-Salazar, F. (2022). Effect of immunocastration with a gonadotropin-releasing hormone vaccine in a male zebra Equus quagga boehmi. Journal of Zoo and Aquarium Research, 10(4), 210-214.
Alouatta caraya	Likely castration but could be mixed	Castration more likely but approach can differ by institution			

Ateles geoffroyi	Likely castration but could be mixed	Castration most likely				
Saguinus oedipus	Likely castration but could be mixed	Castration more likely but approach can differ by institution				
Callithrix jacchus	Likely castration but could be mixed	Castration more likely but approach can differ by institution				
Sapajus apella	Likely castration but could be mixed	Castration more likely but approach can differ by institution				
Cebus capucinus	Likely castration but could be mixed	Castration more likely but approach can differ by institution				
Saimiri sciureus	Likely castration but could be mixed	Castration more likely but approach can differ by institution				
Papio hamadryas	Either	Vasectomy could be used to keep males looking like males	Mane in males	Hamadryas baboon care manual states either castration or vasectomy can be used. koot et al 2016 report on castrated males in European zoos for behavioural reasons	Koot, Susanne, et al. "A note on the social compatibility of an all-male group of hamadryas baboons (Papio	

				hamadryas)." Journal of Zoo and Aquarium Research 4.1 (2016): 7-13.	
Macaca leonina	Likely castration but could be mixed				
Erythrocebus patas	Likely castration but could be mixed				
Macaca silenus	Likely castration but could be mixed	Previous manager stated castration at one institution			
Macaca fascicularis	Likely castration but could be mixed				
Macaca nigra	Likely castration but could be mixed				
Macaca fuscata	Likely castration but could be mixed	Manager at one institution stated castration to control behaviour			
Macaca sylvanus	Likely castration but could be mixed				
Macaca mulatta	Likely castration but could be mixed				

Papio papio	Likely castration but could be mixed				
Mandrillus sphinx	Likely vasectomized	Likely vasectomy although various approaches can be used in some institutions	Color dimorphism in males		
Pan troglodytes	Could be either	Currently vasectomy but prior to the early 2000s castration was the main method			
Lemur catta	Likely castration but could be mixed	Suggested castration most likely			
Hydrochoerus hydrochaeris	Castration	Castration		castration used to curb aggressive behaviours (Yu et al 2020)	Yu, J. H., Brown, J., Boisseau, N., Barthel, T., & Murray, S. (2021). Effects of Lupron and surgical castration on fecal androgen metabolite concentrations and intermale aggression in capybaras

					(Hydrochoerus hydrochaeris). Zoo biology, 40(2), 135-141.	
Cavia porcellus	Castration	Castration		Domestic likely to be castrated		
Dolichotis patagonum	Likely castration but could be mixed	Castration		Website describes a male being neutered to change behaviour	Ambassador animal resource and information center (accessed 10/03/2025)	https://ambassadoranimalsag.wordpress.com/2019/11/14/patagonian-cavy/
Chinchilla lanigera	Castration	Castration		Domestic species likely to be castrated as pets		
Rattus norvegicus	Likely castration but could be mixed	Castration		Would assume zoos would castrate but vasectomy is easy too		
Myocastor coypus	Likely castration but could be mixed	Castration		In rodents castration would be common although there is an article on male vasectomy on free-ranging populations (von Degerfeld et al 2021)	Von Degerfeld MM, Serpieri M, Banchi P, Quaranta G. Field anesthesia of free-ranging nutrias (myocastor coypus) for surgical reproduction	

					control. J Wildl Dis. 2021 Jul 1;57(3):495-502. doi: 10.7589/JWD-D-20-00174. PMID: 34015804.	
Octodon degus	Castration	Castration		Article outlining castration and saying this is a common method (Malbrue et al). No information on vasectomy	Malbrue, Raphael A., et al. "Evaluation of orchiectomy and ovariectomy surgical techniques in degus (Octodon Degus)." Journal of Exotic Pet Medicine 30 (2019): 22-28.	
Cynomys ludovicianus	Castration	Castration		Likely castation and decribed in Isaza 2020	Isaza, N., & Isaza, R. (2020). Neutering Procedures and Consideration	

	s in Rabbits and Other Small Mammals. High-Quality, High-Volume Spay and Neuter and Other Shelter Surgeries, 295-323.	

Supplementary Table 2. Average effect size estimates (yi) and the corresponding sampling variances (vi) for change in life-expectancy with male surgical sterilization or female hormonal contraception for zoo housed species. The data is presented in the same order as the corresponding phylogenetic tree shown in Figure 1a. #N/A stands for "Not Available" (no usable data).

Order	Species name	Male surgical effect size (yi)	Male surgical variance (vi)	Female hormonal effect size (yi)	Female hormonal variance (vi)
Artiodactyla	Capra_hircus	-0.028	0.001	#N/A	#N/A
Artiodactyla	Hemitragus_jemlahicus	-0.039	0.022	#N/A	#N/A
Artiodactyla	Ovis_aries	-0.035	0.012	#N/A	#N/A
Artiodactyla	Ammotragus_lervia	-0.056	0.004	#N/A	#N/A
Artiodactyla	Oryx_dammah	0.24	0.004	0.053	0.01
Artiodactyla	Addax_nasomaculatus	0.129	0.008	0.082	0.009
Artiodactyla	Antilope_cervicapra	0.499	0.004	#N/A	#N/A
Artiodactyla	Eudorcas_thomsonii	0.437	0.014	#N/A	#N/A
Artiodactyla	Kobus_megaceros	0.141	0.014	#N/A	#N/A
Artiodactyla	Aepyceros_melampus	0.498	0.01	#N/A	#N/A
Artiodactyla	Bos_grunniens	-0.199	0.028	#N/A	#N/A
Artiodactyla	Bison_bison	0.141	0.008	#N/A	#N/A
Artiodactyla	Tragelaphus_oryx	0.303	0.006	#N/A	#N/A
Artiodactyla	Tragelaphus_strepsiceros	0.158	0.015	0.002	0.013
Artiodactyla	Boselaphus_tragocamelus	-0.098	0.032	#N/A	#N/A

Artiodactyla	Cervus_elaphus	0.104	0.023	#N/A	#N/A
Artiodactyla	Dama_dama	0.329	0.012	#N/A	#N/A
Artiodactyla	Axis_axis	#N/A	#N/A	-0.517	0.036
Artiodactyla	Muntiacus_reevesi	0.235	0.013	#N/A	#N/A
Artiodactyla	Odocoileus_virginianus	0.371	0.025	#N/A	#N/A
Artiodactyla	Rangifer_tarandus	0.329	0.016	#N/A	#N/A
Artiodactyla	Giraffa_camelopardalis	#N/A	#N/A	0.378	0.011
Artiodactyla	Potamochoerus_porcus	0.174	0.007	-0.045	0.007
Artiodactyla	Sus_scrofa	-0.022	0.017	#N/A	#N/A
Artiodactyla	Phacochoerus_africanus	#N/A	#N/A	0.042	0.006
Artiodactyla	Pecari_tajacu	0.091	0.008	#N/A	#N/A
Artiodactyla	Lama_glama	-0.077	0.004	#N/A	#N/A
Artiodactyla	Lama_guanicoe	0.003	0.022	#N/A	#N/A
Artiodactyla	Vicugna_pacos	-0.03	0.003	#N/A	#N/A
Artiodactyla	Camelus_bactrianus	-0.068	0.007	#N/A	#N/A
Artiodactyla	Camelus_dromedarius	-0.186	0.012	#N/A	#N/A
Perissodactyla	Equus_quagga	0.489	0.017	#N/A	#N/A
Perissodactyla	Equus_caballus	-0.063	0.008	#N/A	#N/A
Carnivora	Nasua_narica	-0.137	0.012	#N/A	#N/A
Carnivora	Nasua_nasua	0.132	0.002	-0.019	0.01
Carnivora	Procyon_lotor	0.244	0.002	#N/A	#N/A

Carnivora	Mustela_nigripes	0.019	0.005	#N/A	#N/A
Carnivora	Aonyx_cinerea	0.008	0.005	0.156	0.004
Carnivora	Mephitis_mephitis	-0.04	0.003	#N/A	#N/A
Carnivora	Zalophus_californianus	-0.134	0.013	#N/A	#N/A
Carnivora	Ursus_arctos	-0.078	0.008	#N/A	#N/A
Carnivora	Ursus_americanus	0.084	0.006	#N/A	#N/A
Carnivora	Canis_lupus	#N/A	#N/A	#N/A	#N/A
Carnivora	Canis_latrans	0.128	0.005	#N/A	#N/A
Carnivora	Lycaon_pictus	#N/A	#N/A	0.061	0.006
Carnivora	Vulpes_zerda	-0.166	0.008	#N/A	#N/A
Carnivora	Vulpes_vulpes	0.269	0.009	#N/A	#N/A
Carnivora	Vulpes_lagopus	0.019	0.015	#N/A	#N/A
Carnivora	Lynx_lynx	0.081	0.012	#N/A	#N/A
Carnivora	Lynx_rufus	0.156	0.004	#N/A	#N/A
Carnivora	Puma_concolor	0.001	0.004	#N/A	#N/A
Carnivora	Felis_catus	0.03	0.015	#N/A	#N/A
Carnivora	Leptailurus_serval	0.179	0.004	-0.084	0.016
Carnivora	Panthera_leo	-0.028	0.002	0.12	0.001
Carnivora	Panthera_onca	#N/A	#N/A	-0.01	0.005
Carnivora	Panthera_tigris	#N/A	#N/A	#N/A	#N/A
Carnivora	Mungos_mungo	-0.158	0.018	#N/A	#N/A
Carnivora	Suricata_suricatta	0.03	0.002	0.093	0.001
Chiroptera	Eidolon_helvum	0.114	0.003	#N/A	#N/A
Chiroptera	Pteropus_rodricensis	#N/A	#N/A	0.202	0.008

Chiroptera	Rousettus_aegyptiacus	-0.204	0.008	#N/A	#N/A
Chiroptera	Desmodus_rotundus	0.257	0.015	#N/A	#N/A
Primates	Macaca_nigra	-0.192	0.02	-0.015	0.012
Primates	Macaca_fuscata	0.123	0.011	-0.055	0.005
Primates	Macaca_mulatta	0.293	0.01	#N/A	#N/A
Primates	Macaca_fascicularis	0.147	0.038	#N/A	#N/A
Primates	Macaca_silenus	0.063	0.016	0.05	0.011
Primates	Macaca_nemestrina	#N/A	#N/A	-0.08	0.032
Primates	Macaca_leonina	-0.254	0.097	#N/A	#N/A
Primates	Macaca_sylvanus	0.371	0.008	0.157	0.005
Primates	Papio_hamadryas	0.174	0.005	0.258	0.002
Primates	Papio_papio	-0.051	0.023	0	0.014
Primates	Mandrillus_sphinx	0.031	0.008	0.167	0.004
Primates	Erythrocebus_patas	-0.065	0.023	0.153	0.011
Primates	Cercopithecus_neglectus	#N/A	#N/A	0.012	0.01
Primates	Trachypithecus_cristatus	#N/A	#N/A	0.22	0.028
Primates	Trachypithecus_francoisi	#N/A	#N/A	0.063	0.024
Primates	Colobus_guereza	#N/A	#N/A	0.371	0.009
Primates	Pan_troglodytes	0.299	0.006	0.021	0.003
Primates	Pongo_abelii	#N/A	#N/A	0.207	0.008
Primates	Nomascus_leucogenys	#N/A	#N/A	0.003	0.019

Primates	Symphalangus_syndactylus	#N/A	#N/A	0.216	0.016
Primates	Callithrix_jacchus	0.224	0.009	0.06	0.009
Primates	Callithrix_geoffroyi	#N/A	#N/A	-0.088	0.013
Primates	Cebuella_pygmaea	#N/A	#N/A	0.191	0.018
Primates	Callimico_goeldii	#N/A	#N/A	0.124	0.013
Primates	Leontopithecus_chrysomelas	#N/A	#N/A	0.212	0.007
Primates	Leontopithecus_rosalia	#N/A	#N/A	0.205	0.005
Primates	Saguinus_bicolor	#N/A	#N/A	0.26	0.022
Primates	Saguinus_oedipus	0.051	0.007	0.157	0.004
Primates	Cebus_capucinus	0.327	0.016	#N/A	#N/A
Primates	Sapajus_apella	-0.119	0.012	#N/A	#N/A
Primates	Saimiri_sciureus	0.434	0.009	0.203	0.01
Primates	Ateles_geoffroyi	-0.261	0.039	#N/A	#N/A
Primates	Alouatta_caraya	0.061	0.014	0.279	0.013
Primates	Pithecia_pithecia	#N/A	#N/A	0.204	0.017
Primates	Eulemur_macaco	#N/A	#N/A	0.324	0.023
Primates	Lemur_catta	0.097	0.002	0.109	0.002
Primates	Varecia_rubra	#N/A	#N/A	0.069	0.005
Rodentia	Octodon_degus	0.123	0.005	0.286	0.007
Rodentia	Myocastor_coypus	-0.026	0.031	#N/A	#N/A
Rodentia	Chinchilla_lanigera	0.138	0.027	#N/A	#N/A
Rodentia	Dolichotis_patagonum	0.092	0.01	#N/A	#N/A

Rodentia	Hydrochoerus_hydrochaeris	-0.018	0.005	-0.065	0.011
Rodentia	Cavia_porcellus	0.036	0.003	#N/A	#N/A
Rodentia	Cynomys_ludovicianus	-0.081	0.009	#N/A	#N/A
Rodentia	Rattus_norvegicus	0.021	0.01	#N/A	#N/A
Lagomorpha	Oryctolagus_cuniculus	-0.018	0.009	#N/A	#N/A
Hyracoidea	Procavia_capensis	#N/A	#N/A	#N/A	#N/A
Diprotodontia	Macropus_parma	0.257	0.015	#N/A	#N/A
Diprotodontia	Macropus_rufogriseus	0.163	0.002	#N/A	#N/A
Diprotodontia	Macropus_rufus	0.194	0.003	#N/A	#N/A
Diprotodontia	Macropus_fuliginosus	0.202	0.013	#N/A	#N/A
Diprotodontia	Macropus_giganteus	0.189	0.008	#N/A	#N/A
Diprotodontia	Petaurus_breviceps	-0.059	0.007	#N/A	#N/A
Didelphimorphia	Didelphis_virginiana	0.09	0.006	#N/A	#N/A

Supplementary Table 3. Testes mass and body mass for males of species where the effect of surgical castration on life-expectancy has been assessed. Information on average testes mass and body mass was found through searches of the published literature. The reference where the data was sourced from is shown, with the full references listed under the table.

species	Combined_Testes_Mass(g)	Male_Body_Mass(g)	Reference
Didelphis virginiana	3	3750	Anderson_et_al_2004-J-Zool.
Procavia capensis	17.2	2025	Anderson_et_al_2004-J-Zool.
Vulpes lagopus	4.064433292	4797.334486	Baker_2020_Ecol_Lett
Addax nasomaculatus	43.2	117000	Ferrandiz-Rovira_et_al_2014-BMC_Evol_Biol
Hemitragus jemlahicus	26.7	103300	Ferrandiz-Rovira_et_al_2014-BMC_Evol_Biol
Kobus megaceros	52	105500	Ferrandiz-Rovira_et_al_2014-BMC_Evol_Biol
Mephitis mephitis	5.05	5050	Iossa_et_al_2008-Funct-Ecol
Procyon lotor	13.72	5130	Iossa_et_al_2008-Funct-Ecol
Suricata suricatta	1.3	731	Iossa_et_al_2008-Funct-Ecol
Ateles geoffroyi	13.4	7940	Kenagy_Trombulak_1986_J-Mammal
Callithrix jacchus	1.3	320	Kenagy_Trombulak_1986_J-Mammal
Cervus elaphus	217	121800	Kenagy_Trombulak_1986_J-Mammal
Chinchilla lanigera	5.2	422.3	Kenagy_Trombulak_1986_J-Mammal
Odocoileus virginianus	85.5	44800	Kenagy_Trombulak_1986_J-Mammal
Oryctolagus cuniculus	6.06	2888	Kenagy_Trombulak_1986_J-Mammal

Ovis aries	318.8	35000	Kenagy_Trombulak_1986_J-Mammal
Pecari tajacu	11.3	12300	Kenagy_Trombulak_1986_J-Mammal
Saimiri sciureus	3.17	779	Kenagy_Trombulak_1986_J-Mammal
Tragelaphus strepsiceros	92	248000	Kenagy_Trombulak_1986_J-Mammal
Vulpes vulpes	9	5069	Kenagy_Trombulak_1986_J-Mammal
Alouatta caraya	18.37	6420	Lüpold _et_al_2019-PRSB
Erythrocebus patas	7.2	13000	Lüpold _et_al_2019-PRSB
Lemur catta	17.8	2700	Lüpold _et_al_2019-PRSB
Macaca fascicularis	35.2	4420	Lüpold _et_al_2019-PRSB
Macaca fuscata	72.3	15400	Lüpold _et_al_2019-PRSB
Macaca mulatta	46	9200	Lüpold _et_al_2019-PRSB
Macaca silenus	42	5900	Lüpold _et_al_2019-PRSB
Mandrillus sphinx	58.86	26900	Lüpold _et_al_2019-PRSB
Pan troglodytes	128.9	44670	Lüpold _et_al_2019-PRSB
Papio hamadryas	27.1	20170	Lüpold _et_al_2019-PRSB
Saguinus oedipus	3.4	520	Lüpold _et_al_2019-PRSB
Sapajus apella	9.1	4800	Lüpold _et_al_2019-PRSB
Aepyceros melampus	120	62500	Lüpold_2013-Evolution
Ammotragus lervia	18.85	68000	Lüpold_2013-Evolution
Bison bison	523.6	731400	Lüpold_2013-Evolution

Camelus bactrianus	95	816000	Lüpold_2013-Evolution
Camelus dromedarius	121.7	800000	Lüpold_2013-Evolution
Canis latrans	22.26	11400	Lüpold_2013-Evolution
Capra hircus	156.8	25420	Lüpold_2013-Evolution
Dama dama	133	63700	Lüpold_2013-Evolution
Equus caballus	416	468000	Lüpold_2013-Evolution
Felis catus	2.34	3100	Lüpold_2013-Evolution
Lama glama	48	133000	Lüpold_2013-Evolution
Lycaon pictus	28.6	31464.3	Lüpold_2013-Evolution
Lynx lynx	6.13	17833	Lüpold_2013-Evolution
Lynx rufus	2.14	10450	Lüpold_2013-Evolution
Mustela nigripes	2.1	950	Lüpold_2013-Evolution
Myocastor coypus	9.6	5520	Lüpold_2013-Evolution
Nasua nasua	4.84	4400	Lüpold_2013-Evolution
Oryx dammah	53.4	129000	Lüpold_2013-Evolution
Panthera leo	27.6	183740	Lüpold_2013-Evolution
Puma concolor	14.9	45000	Lüpold_2013-Evolution
Rangifer tarandus	132	145000	Lüpold_2013-Evolution
Sus scrofa	128.2	39700	Lüpold_2013-Evolution
Ursus americanus	56	125000	Lüpold_2013-Evolution
Ursus arctos	68.6	180900	Lüpold_2013-Evolution
Vicugna pacos	34	63000	Lüpold_2013-Evolution
Hydrochoerus	68.8	43100	Moreira_et_al_1997_J_Mammal

hydrochaeris			
Antilope cervicapra	NA	NA	NA
Aonyx cinereus	NA	NA	NA
Bos grunniens	NA	NA	NA
Boselaphus tragocamelus	NA	NA	NA
Cebus capucinus	NA	NA	NA
Dolichotis patagonum	NA	NA	NA
Equus quagga	NA	NA	NA
Eudorcas thomsonii	NA	NA	NA
Lama guanicoe	NA	NA	NA
Leptailurus serval	NA	NA	NA
Macaca leonina	NA	NA	NA
Macaca nigra	NA	NA	NA
Macaca sylvanus	NA	NA	NA
Macropus fuliginosus	NA	NA	NA
Mungos mungo	NA	NA	NA
Muntiacus reevesi	NA	NA	NA
Nasua narica	NA	NA	NA
Notamacropus parma	NA	NA	NA

Notamacropus rufogriseus	NA	NA	NA
Osphranter rufus	NA	NA	NA
Papio papio	NA	NA	NA
Potamochoerus porcus	NA	NA	NA
Tragelaphus oryx	NA	NA	NA
Vulpes zerda	NA	NA	NA
Zalophus californianus	NA	NA	NA
Octodon degus	2.16	161.6119493	Obregón_Ramirez_1997_Andrologia_&_Bauer_etal_2014_GCE
Desmodus rotundus	0.083	33.7	Pitnick_et_al_2006-PRSB
Eidolon helvum	5.5	325	Pitnick_et_al_2006-PRSB
Rousettus aegyptiacus	3.5	142	Pitnick_et_al_2006-PRSB
Cavia porcellus	3.73	714.5	Ramm_et_al_2005_PRSB
Cynomys ludovicianus	4	815	Ramm_et_al_2005_PRSB
Rattus norvegicus	4	500	Ramm_et_al_2005_PRSB
Macropus giganteus	42.02	40720	Rose_et_al_1997_J-Zool
Petaurus breviceps	0.14	136.3	Rose_et_al_1997_J-Zool

-Anderson, M. J., Nyholt, J., & Dixson, A. F. (2004). Sperm competition affects the structure of the mammalian vas deferens. *Journal of Zoology*, *264*(1), 97-103.
-Baker, J., Humphries, S., Ferguson-Gow, H., Meade, A., & Venditti, C. (2020). Rapid decreases in relative testes mass among monogamous birds but not in other vertebrates. *Ecology letters*, *23*(2), 283-292.

Bauer, C. M., Hayes, L. D., Ebensperger, L. A., & Romero, L. M. (2014). Seasonal variation in the degu (Octodon degus) endocrine stress response. *General and Comparative Endocrinology*, *197*, 26-32.
Ferrandiz-Rovira, M., Lemaître, J. F., Lardy, S., López, B. C., & Cohas, A. (2014). Do pre-and post-copulatory sexually selected traits covary in large herbivores?. *BMC evolutionary biology*, *14*, 1-9.
Iossa, G., Soulsbury, C. D., Baker, P. J., & Harris, S. (2008). Sperm competition and the evolution of testes size in terrestrial mammalian carnivores. *Functional Ecology*, *22*(4), 655-662.
Kenagy, G. J., & Trombulak, S. C. (1986). Size and function of mammalian testes in relation to body size. *Journal of Mammalogy*, *67*(1), 1-22.
Lüpold, S. (2013). Ejaculate quality and constraints in relation to sperm competition levels among eutherian mammals. *Evolution*, *67*(10), 3052-3060.
Lüpold, S., Simmons, L. W., & Grueter, C. C. (2019). Sexual ornaments but not weapons trade off against testes size in primates. *Proceedings of the Royal Society B*, *286*(1900), 20182542.
Moreira, J. R., Clarke, J. R., & Macdonald, D. W. (1997). The testis of capybaras (Hydrochoerus hydrochaeris). *Journal of Mammalogy*, *78*(4), 1096-1100.
Obregón, E. B., & Ramirez, O. (1997). Ageing and testicular function in Octodon degus. *Andrologia*, *29*(6), 319-326.
Pitnick, S., Jones, K. E., & Wilkinson, G. S. (2006). Mating system and brain size in bats. *Proceedings of the Royal Society B: Biological Sciences*, *273*(1587), 719-724.
Ramm, S. A., Parker, G. A., & Stockley, P. (2005). Sperm competition and the evolution of male reproductive anatomy in rodents. *Proceedings of the Royal Society B: Biological Sciences*, *272*(1566), 949-955.
Rose, R. W., Nevison, C. M., & Dixson, A. F. (1997). Testes weight, body weight and mating systems in marsupials and monotremes. *Journal of Zoology*, *243*(3), 523-531.

Supplementary Table 4. Search string development for querying online databases of literature of survival changes with sterilization. Final search strings used to retrieve bibliographic records for screening are in bold.

database, date	nr	search string	n hits	comments
Scopus, 5/04/2020	1	**TITLE-ABS-KEY ((sterili* OR ovariec* OR gonadec* OR testosterone OR eunuch* OR castrat*) AND (longevity OR " life expectancy" OR lifespan OR "life extension") AND NOT (cancer)) AND PUBYEAR < 2020**	1544	first search
Scopus, 14/04/2020	2	**TITLE-ABS-KEY ((sterili* OR ovariec* OR gonadec* OR testosterone OR eunuch* OR castrat* OR neuter* OR germline OR "fertility control") AND (longevity OR "life expectancy" OR lifespan OR "life extension" OR "survival rate" OR "live longer") AND NOT (cancer)) AND PUBYEAR < 2020 AND (EXCLUDE (EXACTKEYWORD , "Animal Tissue") OR EXCLUDE (EXACTKEYWORD , "Signal Transduction") OR EXCLUDE (EXACTKEYWORD , "Instrument Sterilization") OR EXCLUDE (EXACTKEYWORD , "Protein Expression") OR EXCLUDE (EXACTKEYWORD , "Gene Expression") OR EXCLUDE (EXACTKEYWORD , "Blood") OR EXCLUDE (EXACTKEYWORD , "Ovary") OR EXCLUDE (EXACTKEYWORD , "Child") OR EXCLUDE (EXACTKEYWORD , "Cytology") OR EXCLUDE (EXACTKEYWORD , "Gene Expression Regulation") OR EXCLUDE (EXACTKEYWORD , "Bone density") OR EXCLUDE (EXACTKEYWORD , "Enzyme Activity") OR EXCLUDE (EXACTKEYWORD , "Cell Differentiation") OR EXCLUDE (EXACTKEYWORD , "Histology") OR EXCLUDE (EXACTKEYWORD , "Oocyte") OR EXCLUDE (EXACTKEYWORD , "Osteolysis") OR EXCLUDE (EXACTKEYWORD , "Stem Cells"))**	1959	second search

Scopus, 27/10/2021	3	((TITLE-ABS-KEY ((sterili* OR ovariec* OR gonadec* OR testosterone OR eunuch* OR castrat*) AND (longevity OR "life expectancy" OR lifespan OR "life extension") AND NOT (cancer)))) OR ((TITLE-ABS-KEY ((sterili* OR ovariec* OR gonadec* OR testosterone OR eunuch* OR castrat* OR neuter* OR germline OR "fertility control") AND (longevity OR "life expectancy" OR lifespan OR "life extension" OR "survival rate" OR "live longer") AND NOT (cancer))) AND PUBYEAR < 2020) AND (EXCLUDE (EXACTKEYWORD , "Animal Tissue") OR EXCLUDE (EXACTKEYWORD , "Signal Transduction") OR EXCLUDE (EXACTKEYWORD , "Instrument Sterilization") OR EXCLUDE (EXACTKEYWORD , "Protein Expression") OR EXCLUDE (EXACTKEYWORD , "Gene Expression") OR EXCLUDE (EXACTKEYWORD , "Blood") OR EXCLUDE (EXACTKEYWORD , "Ovary") OR EXCLUDE (EXACTKEYWORD , "Child") OR EXCLUDE (EXACTKEYWORD , "Cytology") OR EXCLUDE (EXACTKEYWORD , "Gene Expression Regulation") OR EXCLUDE (EXACTKEYWORD , "Bone density") OR EXCLUDE (EXACTKEYWORD , "Enzyme Activity") OR EXCLUDE (EXACTKEYWORD , "Cell Differentiation") OR EXCLUDE (EXACTKEYWORD , "Histology") OR EXCLUDE (EXACTKEYWORD , "Oocyte") OR EXCLUDE (EXACTKEYWORD , "Osteolysis") OR EXCLUDE (EXACTKEYWORD , "Stem Cells"))	2072	#1 OR #2: combined first two searches
Scopus, 27/10/2021	4	PMID ("5835018") OR DOI ("10.1530/jrf.0.0140113") OR DOI ("10.7554/eLife.56177") OR DOI ("10.1016/0531-5565(76)90007-3") OR DOI ("10.1093/geronj/20.1.96") OR DOI ("10.1093/geronj/24.4.395") OR DOI ("10.1371/journal.pone.0061082") OR DOI ("10.1111/j.1469-7998.1997.tb02806.x") OR DOI ("10.1016/j.cub.2012.06.036") OR DOI ("10.18632/oncotarget.2984") OR DOI ("10.1093/gerona/glp134") OR DOI ("10.1007/s11357-020-00308-8") OR DOI ("10.1038/366215a0") OR DOI ("10.2307/4179") OR DOI ("10.1097/AOG.0b013e3182864350")	14	retrieving benchmarking set: 14 out of 15 of test set papers are indexed in Scopus

Scopus, 27/10/2021	5	(PMID ("5835018") OR DOI ("10.1530/jrf.0.0140113") OR DOI ("10.7554/eLife.56177") OR DOI ("10.1016/0531-5565(76)90007-3") OR DOI ("10.1093/geronj/20.1.96") OR DOI ("10.1093/geronj/24.4.395") OR DOI ("10.1371/journal.pone.0061082") OR DOI ("10.1111/j.1469-7998.1997.tb02806.x") OR DOI ("10.1016/j.cub.2012.06.036") OR DOI ("10.18632/oncotarget.2984") OR DOI ("10.1093/gerona/glp134") OR DOI ("10.1007/s11357-020-00308-8") OR DOI ("10.1038/366215a0") OR DOI ("10.2307/4179") OR DOI ("10.1097/AOG.0b013e3182864350")) AND (((TITLE-ABS-KEY ((sterili* OR ovariec* OR gonadec* OR testosterone OR eunuch* OR castrat*) AND (longevity OR "life expectancy" OR lifespan OR "life extension") AND NOT (cancer)))) OR ((TITLE-ABS-KEY ((sterili* OR ovariec* OR gonadec* OR testosterone OR eunuch* OR castrat* OR neuter* OR germline OR "fertility control") AND (longevity OR "life expectancy" OR lifespan OR "life extension" OR "survival rate" OR "live longer") AND NOT (cancer))) AND PUBYEAR < 2020)) AND (EXCLUDE (EXACTKEYWORD , "Animal Tissue") OR EXCLUDE (EXACTKEYWORD , "Signal Transduction") OR EXCLUDE (EXACTKEYWORD , "Instrument Sterilization") OR EXCLUDE (EXACTKEYWORD , "Protein Expression") OR EXCLUDE (EXACTKEYWORD , "Gene Expression") OR EXCLUDE (EXACTKEYWORD , "Blood") OR EXCLUDE (EXACTKEYWORD , "Ovary") OR EXCLUDE (EXACTKEYWORD , "Child") OR EXCLUDE (EXACTKEYWORD , "Cytology") OR EXCLUDE (EXACTKEYWORD , "Gene Expression Regulation") OR EXCLUDE (EXACTKEYWORD , "Bone density") OR EXCLUDE (EXACTKEYWORD , "Enzyme Activity") OR EXCLUDE (EXACTKEYWORD , "Cell Differentiation") OR EXCLUDE (EXACTKEYWORD , "Histology") OR EXCLUDE (EXACTKEYWORD , "Oocyte") OR EXCLUDE (EXACTKEYWORD , "Osteolysis") OR EXCLUDE (EXACTKEYWORD , "Stem Cells"))	5	#4 AND #5 - benchmarking two original searches against the test search -> sensitivity 5/14
Scopus, 27/10/2021	6	(TITLE-ABS-KEY (steril* OR ovariec* OR gonadect* OR testosterone OR "sex hormone*" OR "steroid hormones" OR eunuch* OR castrat*)) W/10 (TITLE-ABS-KEY (longevity OR "life expectancy" OR lifespan OR "life extension" OR surviv* OR mortality OR death OR "Duration of life" OR "life span")) AND (LIMIT-TO (DOCTYPE , "ar"))	5690	initial string for expanding and updating Scopus searches

Scopus, 27/10/2021	7	(TITLE-ABS-KEY (steril* OR ovariec* OR gonadect* OR testosterone OR "sex hormone*" OR "steroid hormones" OR eunuch* OR castrat*)) W/10 (TITLE-ABS-KEY (longevity OR "life expectancy" OR lifespan OR "life extension" OR surviv* OR mortality OR death OR "Duration of life" OR "life span")) AND (PMID ("5835018") OR DOI ("10.1530/jrf.0.0140113") OR DOI ("10.7554/eLife.56177") OR DOI ("10.1016/0531-5565(76)90007-3") OR DOI ("10.1093/geronj/20.1.96") OR DOI ("10.1093/geronj/24.4.395") OR DOI ("10.1371/journal.pone.0061082") OR DOI ("10.1111/j.1469-7998.1997.tb02806.x") OR DOI ("10.1016/j.cub.2012.06.036") OR DOI ("10.18632/oncotarget.2984") OR DOI ("10.1093/gerona/glp134") OR DOI ("10.1007/s11357-020-00308-8") OR DOI ("10.1038/366215a0") OR DOI ("10.2307/4179") OR DOI ("10.1097/AOG.0b013e3182864350"))	13	#4 AND #6: benchmarking string #6 -> sensitivity 13/14
Scopus, 27/10/2021	8	((TITLE-ABS-KEY (steril* OR ovariec* OR gonadect* OR testosterone OR "sex hormone*" OR "steroid hormones" OR eunuch* OR castrat*)) W/10 (TITLE-ABS-KEY (longevity OR "life expectancy" OR lifespan OR "life extension" OR surviv* OR mortality OR death OR "Duration of life" OR "life span"))) AND NOT KEY (cancer* OR centenarian* OR oocyte* OR patient* OR diet* OR cohort* OR *therap* OR biofilm* OR product* OR "testosterone association")	3888	
Scopus, 27/10/2021	9	((TITLE-ABS-KEY (steril* OR ovariec* OR gonadect* OR testosterone OR "sex hormone*" OR "steroid hormones" OR eunuch* OR castrat*)) W/10 (TITLE-ABS-KEY (longevity OR "life expectancy" OR lifespan OR "life extension" OR surviv* OR mortality OR death OR "Duration of life" OR "life span"))) AND NOT TITLE-ABS-KEY (*therap* OR diet* OR product* OR hospital* OR plant*)	3064	
Scopus, 27/10/2021	10	((TITLE-ABS-KEY (steril* OR ovariec* OR gonadect* OR testosterone OR "sex hormone*" OR "steroid hormones" OR eunuch* OR castrat*)) W/10 (TITLE-ABS-KEY (longevity OR "life expectancy" OR lifespan OR "life extension" OR surviv* OR mortality OR death OR "Duration of life" OR "life span"))) AND NOT TITLE-ABS-KEY (*therap* OR diet* OR product* OR instrument* OR hospital* OR plant* OR stem OR leaf OR seed* OR *cultur* OR industr* OR material*)	2313	

Scopus, 27/10/2021	11	((TITLE-ABS-KEY (steril* OR ovariec* OR gonadect* OR testosterone OR "sex hormone*" OR "steroid hormones" OR eunuch* OR castrat*)) W/10 (TITLE-ABS-KEY (longevity OR "life expectancy" OR lifespan OR "life extension" OR surviv* OR mortality OR death OR "Duration of life" OR "life span"))) AND (PMID ("5835018") OR DOI ("10.1530/jrf.0.0140113") OR DOI ("10.7554/eLife.56177") OR DOI ("10.1016/0531-5565(76)90007-3") OR DOI ("10.1093/geronj/20.1.96") OR DOI ("10.1093/geronj/24.4.395") OR DOI ("10.1371/journal.pone.0061082") OR DOI ("10.1111/j.1469-7998.1997.tb02806.x") OR DOI ("10.1016/j.cub.2012.06.036") OR DOI ("10.18632/oncotarget.2984") OR DOI ("10.1093/gerona/glp134") OR DOI ("10.1007/s11357-020-00308-8") OR DOI ("10.1038/366215a0") OR DOI ("10.2307/4179") OR DOI ("10.1097/AOG.0b013e3182864350")) AND NOT TITLE-ABS-KEY (*therap* OR diet* OR product* OR instrument* OR hospital* OR plant* OR stem OR leaf OR seed* OR *cultur* OR industr* OR material*)	13	#4 AND #10: benchmarking string #10 -> sensitivity 13/14

Scopus, 27/10/2021	12	**((TITLE-ABS-KEY (steril* OR ovariec* OR gonadect* OR testosterone OR "sex hormone*" OR "steroid hormones" OR eunuch* OR castrat*)) W/10 (TITLE-ABS-KEY (longevity OR "life expectancy" OR lifespan OR "life extension" OR surviv* OR mortality OR death OR "Duration of life" OR "life span"))) AND NOT TITLE-ABS-KEY (*therap* OR diet* OR product* OR instrument* OR hospital* OR plant* OR stem OR leaf OR seed* OR *cultur* OR industr* OR material*) AND NOT ((TITLE-ABS-KEY ((sterili* OR ovariec* OR gonadec* OR testosterone OR eunuch* OR castrat*) AND (longevity OR "life expectancy" OR lifespan OR "life extension") AND NOT (cancer)))) OR ((TITLE-ABS-KEY ((sterili* OR ovariec* OR gonadec* OR testosterone OR eunuch* OR castrat* OR neuter* OR germline OR "fertility control") AND (longevity OR "life expectancy" OR lifespan OR "life extension" OR "survival rate" OR "live longer") AND NOT (cancer))) AND PUBYEAR < 2020) AND (EXCLUDE (EXACTKEYWORD , "Animal Tissue") OR EXCLUDE (EXACTKEYWORD , "Signal Transduction") OR EXCLUDE (EXACTKEYWORD , "Instrument Sterilization") OR EXCLUDE (EXACTKEYWORD , "Protein Expression") OR EXCLUDE (EXACTKEYWORD , "Gene Expression") OR EXCLUDE (EXACTKEYWORD , "Blood") OR EXCLUDE (EXACTKEYWORD , "Ovary") OR EXCLUDE (EXACTKEYWORD , "Child") OR EXCLUDE (EXACTKEYWORD , "Cytology") OR EXCLUDE (EXACTKEYWORD , "Gene Expression Regulation") OR EXCLUDE (EXACTKEYWORD , "Bone density") OR EXCLUDE (EXACTKEYWORD , "Enzyme Activity") OR EXCLUDE (EXACTKEYWORD , "Cell Differentiation") OR EXCLUDE (EXACTKEYWORD , "Histology") OR EXCLUDE (EXACTKEYWORD , "Oocyte") OR EXCLUDE (EXACTKEYWORD , "Osteolysis") OR EXCLUDE (EXACTKEYWORD , "Stem Cells"))**	1469	#3 AND #10 - additional records from Scopus
WoS, 27/10/2021	13	**(TI=(steril* OR ovariec* OR gonadect* OR eunuch* OR castrat*) AND TI=(longevity OR "life expectancy" OR lifespan OR "life extension" OR surviv* OR mortality OR death OR "Duration of life" OR "life span")) NOT TI=(*therap* OR diet* OR product* OR instrument* OR hospital* OR plant* OR stem OR leaf OR seed* OR *cultur* OR industr* OR material* OR cancer*) and Articles (Document Types)**	301	search string for Web of Science

PubMed, 27/10/2021	14	**"Castration"[MeSH] AND ("Longevity"[MeSH] OR "Life Expectancy"[MeSH])**	94	search string for PubMed
BASE, 27/10/2021	15	**subj:castration doctype:18* Language: English Verbatim search: On**	29	search string for Bielfield Academic Search engine - theses only

Supplementary Table 5. Test set of papers (test set) used to benchmark sensitivity of the final search string for Scopus online databases of academic literature for changes in survival with sterilization.

nr	Reference
1	Asdell, S. A., Doornenbal, H., Joshi, S. R., Sperling, G. A. (1967) The effects of sex steroid hormones upon longevity in rats. J Reprod Fertil 14, 113-120
2	Arriola Apelo, S. I., Lin, A., Brinkman, J. A., Meyer, E., Morrison, M., Tomasiewicz, J. L., Pumper, C. P., Baar, E. L., Richardson, N. E., Alotaibi, M., Lamming, D. W. (2020) Ovariectomy uncouples lifespan from metabolic health and reveals a sex-hormone-dependent role of hepatic mTORC2 in aging. eLife 9, e56177, doi:10.7554/eLife.56177
3	Benedusi, V., Martini, E., Kallikourdis, M., Villa, A., Meda, C., Maggi, A. (2015) Ovariectomy shortens the life span of female mice. Oncotarget 6, 10801-10811
4	Drori, D., Folman, Y. (1976) Environmental effects on longevity in the male rat: Exercise, mating, castration and restricted feeding. Exp Gerontol 11, 25-32, doi:http://dx.doi.org/10.1016/0531-5565(76)90007-3
5	Garratt, M., Try, H., Brooks, R. C. (2021) Access to females and early life castration individually extend maximal but not median lifespan in male mice. GeroScience 43, 1437-1446, doi:10.1007/s11357-020-00308-8
6	Gipps, J., Jewell, P. (1979) Maintaining populations of bank voles, Clethrionomys glareolus, in large outdoor enclosures, and measuring the response of population variables to the castration of males. The Journal of Animal Ecology, 535-555
7	Hamilton, J. B., Mestler, G. E. (1969) Mortality and Survival: Comparison of Eunuchs with Intact Men and Women in a Mentally Retarded Population. J. Gerontol. 24, 395-411, doi:10.1093/geronj/24.4.395
8	Hamilton, J. B. (1965) Relationship of Castration, Spaying, and Sex to Survival and Duration of Life in Domestic Cats. J. Gerontol. 20, 96-104, doi:10.1093/geronj/20.1.96
9	Hoffman, J. M., Creevy, K. E., Promislow, D. E. (2013) Reproductive capability is associated with lifespan and cause of death in companion dogs. PLoS ONE 8, e61082, doi:10.1371/journal.pone.0061082

10	Jewell, P. A. (1997) Survival and behaviour of castrated Soay sheep (Ovis aries) in a feral island population on Hirta, St. Kilda, Scotland. J. Zool. 243, 623-636, doi:10.1111/j.1469-7998.1997.tb02806.x
11	Mason, J. B., Cargill, S. L., Anderson, G. B., Carey, J. R. (2009) Transplantation of young ovaries to old mice increased life span in transplant recipients. J Gerontol A Biol Sci Med Sci 64, 1207-1211, doi:10.1093/gerona/glp134
12	Min, K.-J., Lee, C.-K., Park, H.-N. (2012) The lifespan of Korean eunuchs. Curr. Biol. 22, R792-R793, doi:http://dx.doi.org/10.1016/j.cub.2012.06.036
13	Nieschlag, E., Nieschlag, S., Behre, H. M. (1993) Lifespan and testosterone. Nature 366, 215-215
14	Parker, W. H., Feskanich, D., Broder, M. S., Chang, E., Shoupe, D., Farquhar, C. M., Berek, J. S., Manson, J. E. (2013) Long-term mortality associated with oophorectomy compared with ovarian conservation in the nurses' health study. Obstet. Gynecol. 121, 709-716, doi:10.1097/AOG.0b013e3182864350
15	Talbert, G. B. & Hamilton, J. B. (1965) Duration of life in Lewis strain of rats after gonadectomy at birth and at older ages. J Gerontol 20, 489-491

Supplementary Table 6. List of excluded full-text studies with reasons for exclusion for meta-analysis of changes in survival with sterilization.

reference	main exclusion reason
Abel, E.L., Kruger, M.M., Pandya, K. (2012) Sopranos but not tenors live longer. Aging Male 15: 109-110	No survival data split for sterilization
Aboudkhil, S.,Za√Æd, A.,Henry, L.,Bureau, J.-P. (2003) Influence of age, castration, and testosterone on T cell subsets in healthy and leukemia grafted mice. Biology of the Cell 95: 42614	Survival only assessed when tumors experimentally induced
Anderson, C.P., Bernick, S., Nimni, M.E., Lerner, S.P., Finch, C.E., Strates, B.S. (1994) Effects of genotype and ovariectomy on the skeleton of aging mice. European Journal of Experimental Musculoskeletal Research 3: 61-70	No lifespan/survival data
Anonymous (1965) Chapter VI: Rates free of disease and crude survival rates in relation to prognostic factors and primary castration. Acta Radiologica 3: 54-63	People with cancer
Anonymous (1970) [Eunuchs live longer]. Deutsche Medizinische Wochenschrift 95: 1034-1035	Comment
Anonymous (2005) Keeping ovaries may aid long-term health, survival. Mayo Clinic women's healthsource 9: 3	News article; no data
Anonymous (2013) Banfield: spaying, neutering correlate with longer lives. Journal of the American Veterinary Medical Association 243: 32	Review. No error or exact sample sizes, just a poster with average lifespan
Archer, D. (2010) Lifetime exposure to estrogen and obstructive coronary disease in women. Climacteric 13: 398-399	Only cardiovascular event survival curves

Aschekenasy, A. (1955) Effect of alimentary proteins and of various amino acids on the survival and body weight of adrenalectomized and of adrenalectomized-castrated rats. Annales d'endocrinologie 16: 86-87	No castrated group without additional manipulation
Atwood, C.S., Hayashi, K., Meethal, S.V., Gonzales, T., Bowen, R.L. (2017) Does the degree of endocrine dyscrasia post-reproduction dictate post-reproductive lifespan? Lessons from semelparous and iteroparous species. GeroScience 39: 103-116	No sterilization, comparative study
Baba, T., Shimizu, T., Suzuki, Y.-I., Ogawara, M., Isono, K.-I., Koseki, H., Kurosawa, H., Shirasawa, T. (2005) Estrogen, insulin, and dietary signals cooperatively regulate longevity signals to enhance resistance to oxidative stress in mice. Journal of Biological Chemistry 280: 16417-16426	No survival data
Baeza, I., De Castro, N.M., Arranz, L., Fdez-Tresguerres, J., De La Fuente, M. (2011) Ovariectomy causes immunosenescence and oxi-inflamm-ageing in peritoneal leukocytes of aged female mice similar to that in aged males. Biogerontology 12: 227-238	No survival data
Belo, V.S., Struchiner, C.J., Werneck, G.L., Teixeira Neto, R.G., Tonelli, G.B., De Carvalho, C.G., Jr., Ribeiro, R.A.N., Da Silva, E.S. (2017) Abundance, survival, recruitment and effectiveness of sterilization of free-roaming dogs: A capture and recapture study in Brazil. PLoS ONE 12:	No clear separation of sterilization data
Benowitz-Fredericks, Z.M., Kitaysky, A.S., Welcker, J., Hatch, S.A. (2013) Effects of Food Availability on Yolk Androgen Deposition in the Black-Legged Kittiwake (Rissa tridactyla), a Seabird with Facultative Brood Reduction. PLoS ONE 8:	No sterilization
Bentubo, H.D.L., Tomaz, M.A., Bondan, E.F., Lallo, M.A. (2007) Life expectation and causes of death in dogs in the metropolitan area of S,àö¬£o Paulo (Brazil). Ciencia Rural 37: 1021-1026	Could not separate sexes
Bergstedt, R.A., McDonald, R.B., Twohey, M.B., Mullett, K.M., Young, R.J., Heinrich, J.W. (2003) Reduction in sea lamprey hatching success due to release of sterilized males. Journal of Great Lakes Research 29: 435-444	No actual survival data with sample sizes and proportion of groups
Bimonte-Nelson, H.A., Francis, K.R., Umphlet, C.D., Granholm, A.-C. (2006) Progesterone reverses the spatial memory enhancements initiated by tonic and cyclic oestrogen therapy in middle-aged ovariectomized female rats. European Journal of Neuroscience 24: 229-242	Survival only assessed after surgeries and across an injection period
Bromley, C., Gese, E.M. (2001) Effects of sterilization on territory fidelity and maintenance, pair bonds, and survival rates of free-ranging coyotes. Canadian Journal of Zoology 79: 386-392	Cant separate sexes
Bronson, F.H., Matherne, C.M. (1997) Exposure to anabolic-androgenic steroids shortens life span of male mice. Medicine and Science in Sports and Exercise 29: 615-619	No proper sterilization (used hormone)
Calle, E.E., Rodriguez, C., Walker, K.A., Wingo, P.A., Petrelli, J.M., Thun, M.J. (2001) Tubal sterilization and risk of breast cancer mortality in US women. Cancer Causes and Control 12: 127-135	Only studied breast cancer mortality, no survival data

Campigotto, A.J., Poljak, Z., Stone, E.A., Stacey, D., Bernardo, T.M. (2019) Investigation of relationships between body weight and age among domestic cats stratified by breed and sex. Journal of the American Veterinary Medical Association 255: 205-212	No control group to contrast
Cantarero, A., Perez-Rodriguez, L., Romero-Haro, A.A., Chastel, O., Alonso-Alvarez, C. (2019) Carotenoid-based coloration predicts both longevity and lifetime fecundity in male birds, but testosterone disrupts signal reliability. PLoS ONE 14:	No sterilization
Chalas, E. (2013) Ovaries, estrogen, and longevity. Obstetrics and Gynecology 121: 701-702	Comment on another paper
Chandola, A., Thapliyal, J.P., Gupta, S.C. (1973) Male hormone and thyroid function in the Indian garden lizard, Calotes versicolor. Journal of Endocrinology 57: 325-326	No survival data
Choi, C.-I., Lee, Y.-D., Gwag, B.J., Cho, S.I., Kim, S.-S., Suh-Kim, H. (2008) Effects of estrogen on lifespan and motor functions in female hSOD1 G93A transgenic mice. Journal of the Neurological Sciences 268: 40-47	Only in disease model
Comhaire, F. (2016) Hormone replacement therapy and longevity. Andrologia 48: 65-68	No survival data
Dallo, J., Koles, L. (1996) Longevity treatment with (-)deprenyl in female rats: Effect on copulatory activity and lifespan. Acta Physiologica Hungarica 84: 277-278	No proper sterilization
Davis, SA,Pech, RP (2002) Dependence of population response to fertility control on the survival of sterile animals and their role in regulation. Reproduction : 89-103	No actual survival data
Deleuze, S., Brotcorne, F., Polet, R., Soma, G., Rigaux, G., Giraud, G., Cloutier, F., Poncin, P., Wandia, N., Huynen, M.-C. (2021) Tubectomy of pregnant, non-pregnant female balinese macaques (Macaca fascicularis) with post-operative monitoring. Frontiers in Veterinary Science 8: 688656	No suitable data. Contacted authors to check whether survival data of reproductive group is comparable
Doerner, G., Hinz, G. (1975) Androgen dependent brain differentiation and life span. Endokrinologie 65: 378-380	No intact control group
Douet, W.A., Estienne, M.J., Hartsock, T.G. (1995) Effects of castration within 24 hours after birth or at 10 days of age on pre- and postweaning performance in boars. Professional Animal Scientist 11: 230-232	Very early survival during weaning
Drori, D., Folman, Y. (1986) Interactive environmental and genetic effects on longevity in the male rat: Litter size, exercise, electric shocks and castration. Experimental Aging Research 12: 59-64	All animals had undergone some additional manipulation
Dufty Jr., A.M. (1989) Testosterone and survival: A cost of aggressiveness? Hormones and Behavior 23: 185-193	No sterilization
Durbin, PW,Williams, MH,Jeung, N,Arnold, JS (1966) Development of spontaneous mammary tumors over life-span of female Charles River	Studied survival to cancer incidence

(Sprague-Dawley) rat - influence of ovariectomy thyroidectomy and adrenalectomy-ovariectomy. Cancer Research 26: 400	
Farrell, A., Gerall, A.A., Alexander, M.J. (1977) Age-related decline in receptivity in normal, neonatally androgenized female and male hamsters. Experimental Aging Research 3: 117-128	No survival data
Garcia, R.C.M., Amaku, M., Biondo, A.W., Ferreira, F. (2018) Dog and cat population dynamics in an urban area: Evaluation of a birth control strategy. Pesquisa Veterinaria Brasileira 38: 511-518	No actual neuter lifespan data
Gese, E.M., Terletzky, P.A. (2021) Space use in free-ranging canids: Are gonadal hormones required for territory maintenance? Canadian Journal of Zoology 99: 939-947	No unsterilized data, although different methods of sterilization, with and without gonadal hormone removal, are compared
Gunther, I., Raz, T., Klement, E. (2018) Association of neutering with health and welfare of urban free-roaming cat population in Israel, during 2012-2014. Preventive Veterinary Medicine 157: 26-33	No survival data
Hogberg, U. (1990) Reproductive mortality and its relation to different methods of birth control. Journal of Biosocial Science 22: 323-331	No overall survival data, just reproductive mortality
Hamilto, K.F., Sacher, G.A., Grahn, D. (1963) A sex difference in mouse survival under daily gamma irradiation and its modification by gonadectomy. Radiation Research 18: 42705	Only study irradiated animals, no normal survival data
Hernandez-Barraza, C.A., Trejo-Martinez, A.B., Loredo-Osti, J., Gutiarrez-Salazar, G. (2016) Evaluation of the productive efficiency of three strains of tilapia sex reversal in a recirculation system (RAS). Latin American Journal of Aquatic Research 44: 869-874	Spanish - cannot see relevant data with my poor Spanish!
Horisberger, U., Stark, K.D., Rufenacht, J., Pillonel, C., Steiger, A. (2004) [Demographic characteristics of dog population in Switzerland]. Schweizer Archiv fur Tierheilkunde 146: 223-32	No survival data for sterilized animals
Huang, W.-H., Liao, A.T., Chu, P.-Y., Yen, I.-F., Liu, C.-H. (2017) A real-time reporting system of causes of death or reasons for euthanasia: A model for monitoring mortality in domesticated cats in Taiwan. Preventive Veterinary Medicine 137: 59-68	Sexes not split
Huang, W.-H., Liao, A.T., Chu, P.-Y., Zhai, S.-H., Yen, I.-F., Liu, C.-H. (2017) A 3-year surveillance on causes of death or reasons for euthanasia of domesticated dogs in Taiwan. Preventive Veterinary Medicine 147: 44105	Sexes not split
Inns, R.W. (1976) Some seasonal changes in Antechinus flavipes (Marsupialia: Dasyuridae). Australian Journal of Zoology 24: 479-489	No survival data
Jewell, P. (1986) Survival in a feral population of primitive sheep on St. Kilda, Outer Hebrides, Scotland. National Geographic Research 2: 402-406	Data already included from a subsequently published paper
Ji, W., Clout, M.N., Sarre, S.D. (2000) Responses of male brushtail possums to sterile females: Implications for biological control. Journal of Applied Ecology 37: 926-934	No survival data

Johnston, N.T., Parkinson, E.A., Tsumura, K. (1993) Longevity and growth of hormone-sterilized kokanee. North American Journal of Fisheries Management 13: 284-290	No proper sterilization (used hormone)
Jones, A.L., Downs, C.T. (2011) Managing feral cats on a university's campuses: How many are there and is sterilization having an effect? Journal of Applied Animal Welfare Science 14: 304-320	No split survival data
Keisler, L.W., Vom Saal, F.S., Keisler, D.H., Walker, S.E. (1991) Hormonal manipulation of the prenatal environment alters reproductive morphology and increases longevity in autoimmune NZB/W mice. Biology of Reproduction 44: 707-716	No sterilization, disease model
Kerber, I.J., Turner, R.J. (2013) Ovaries, estrogen, and longevity and long-term mortality associated with oophorectomy compared with ovarian conservation in the Nurses' Health Study and variation in ovarian conservation in women undergoing hysterectomy for benign indications. Obstetrics and Gynecology 122: 397-398	Comment on another paper
Ketterson, E.D., Nolan Jr, V., Wolf, L., Ziegenfus, C. (1992) Testosterone and avian life histories: effects of experimentally elevated testosterone on behavior and correlates of fitness in the dark-eyed junco (Junco hyemalis). American Naturalist 140: 980-999	No sterilization
Kirkpatrick, J.F., Turner, A. (2008) Achieving population goals in a long-lived wildlife species (Equus caballus) with contraception. Wildlife Research 35: 513-519	No survival data
Knoll, J., Dallo, J., Yen, T.T. (1989) Striatal dopamine, sexual activity and lifespan. Longevity of rats treated with (-) deprenyl. Life Sciences 45: 525-531	No sterilization
Kreisler, R.E., Cornell, H.N., Levy, J.K. (2019) Decrease in population and increase in welfare of community cats in a twenty-three year trap-neuter-return program in Key Largo, FL: The ORCAT program. Frontiers in Veterinary Science 6: 99	No survival data
Kroc, R.L. (1938) Effect of previous castration on weight loss and survival of adrenalectomized rats. Endocrinology 23: 524-526	No survival data without adrenalectomy
Kubicka, L., Golinski, A., John-Alder, H., Kratochvil, L. (2012) Ontogeny of pronounced female-biased sexual size dimorphism in the Malaysian cat gecko (Aeluroscalabotes felinus: Squamata: Eublepharidae): A test of the role of testosterone in growth regulation. General and Comparative Endocrinology 188: 183-188	No proper survival data
Kustritz, M.V. (2007) Determining the optimal age for gonadectomy of dogs and cats. Journal of the American Veterinary Medical Association 231: 1665-75	No data, review
Larsen, L.O. (1974) Effects of Testosterone and oestradiol on gonadectomized and intact male and female river lampreys (Lampetra fluviatilis (L.) Gray). General and Comparative Endocrinology 24: 305-313	No actual castration

Le Bourg, E. (2015) No ground for advocating that Korean eunuchs lived longer than intact men. Gerontology 62: 69-70	No survival data
Ley, S.H., Li, Y., Tobias, D.K., Manson, J.E., Rosner, B., Hu, F.B., Rexrode, K.M. (2017) Duration of reproductive life span, age at menarche, and age at menopause are associated with risk of cardiovascular disease in women. Journal of the American Heart Association 6:	No actual lifespan/survival data
MacLean, R.A., Mathews, N.E., Grove, D.M., Frank, E.S., Paul-Murphy, J. (2006) Surgical technique for tubal ligation in white-tailed deer (Odocoileus virginianus). Journal of Zoo and Wildlife Medicine 37: 354-360	Have included data from cited thesis
Marchant, J. (1967) Development of adrenal cortical carcinoma in c3h mice following castration and the administration of 7,12-dimethylbenz(A)anthracene. British Journal of Cancer 21: 750	No comparable control data
Mason, J.B., Cargill, S.L., Anderson, G.B., Carey, J.R. (2010) Ovarian status influenced the rate of body-weight change but not the total amount of body-weight gained or lost in female CBA/J mice. Experimental Gerontology 45: 435-441	Lifespan data published in Mason et al 2009
Mason, J.B., Cargill, S.L., Griffey, S.M., Reader, J.R., Anderson, G.B., Carey, J.R. (2011) Transplantation of young ovaries restored cardioprotective influence in postreproductive-aged mice. Aging Cell 10: 448-456	Pathology data on Mason et al 2009
Massei, G., Cowan, D.P., Coats, J., Bellamy, F., Quy, R., Pietravalle, S., Brash, M., Miller, L.A. (2012) Long-term effects of immunocontraception on wild boar fertility, physiology and behaviour. Wildlife Research 39: 378-385	No survival data
Matson, K.D., Riedstra, B., Tieleman, B.I. (2016) In ovo testosterone treatment reduces long-term survival of female pigeons: a preliminary analysis after nine years of monitoring. Journal of Animal Physiology and Animal Nutrition 100: 1031-1036	No sterilization
Melez, K.A., Boegel, W.A., Steinberg, A.D. (1980) Therapeutic studies in new zealand mice. Arthritis & Rheumatism 23: 41-47	Disease model
Miller, L.A., Johns, B.E., Elias, D.J., Crane, K.A. (1997) Comparative efficacy of two immunocontraceptive vaccines. Vaccine 15: 1858-1862	No control group
Moskalev, Yu.I., Strel'tsova, V.N., Romanova, L.F. (1967) Effect of ovariectomy on life span and incidence of mammary gland tumors in irradiated rats. Bulletin of Experimental Biology and Medicine 63: 313-315	Survival only studied with irradiation
Munford, R.E. (1957) Effect of age, sex, gonadectomy, sodium chloride intake, and treatment with cortisol acetate upon the lifespan of adrenalectomized mice of an albino strain. The Journal of endocrinology 16: 57-71	No survival data on castrated animals; disease/early death model
Murray, C.F., Fick, L.J., Pajor, E.A., Barkema, H.W., Jelinski, M.D., Windeyer, M.C. (2015) Calf management practices and associations with herd-level morbidity and mortality on beef cow-calf operations. Animal 10: 468-477	No adult survival data

Nassar, R., Mosier, J.E. (1980) Canine population dynamics: a study of the Manhattan, Kansas, canine population. American Journal of Veterinary Research 41: 1798-1803	No survival data
Nassar, R., Mosier, J.E. (1982) Feline population dynamics: a study of the Manhattan, Kansas, feline population. American Journal of Veterinary Research 43: 167-170	No survival data
Neidhart, L., Boyd, R. (2002) Companion animal adoption study. Journal of Applied Animal Welfare Science 5: 175-192	Could not separate sexes
Nguenga, D. (2000) Partial gonadectomy in the catfish Heterobranchus longifilis (Teleostei, Clariidae): Regeneration time, quality and quantity of postsurgical sperm production. Israeli Journal of Aquaculture - Bamidgeh 52: 167-172	Survival was assessed in relation to the surgery
Nieschlag, E., Kramer, U., Nieschlag, S. (2003) Androgens shorten the longevity of women: Sopranos last longer. Experimental and Clinical Endocrinology and Diabetes 111: 230-231	No sterilization comparison
Nilsson, P.M., Nilsson, E., Svanberg, L., Samsioe, G. (2003) Longevity after early surgical menopause-the long-term effect of a permanent cessation of reproductive function and female sex hormone loss. European Journal of Obstetrics, Gynecology, and Reproductive Biology 110: 63-5	No comparable control data with a sample size? - calculation of mean (i.e. one just taken from general population and those living over 50) not the same
Parhon, C.I., Coban, S., Fastlich, G. (1933) Does ovarian castration exert a favourable effect of the duration of the survival of animals which have had thyroparathyroidectomies. Comptes Rendus Des Seances De La Societe De Biologie Et De Ses Filiales 112: 1441-1442	Disease/early death model
Paternostro, G. (1994) Longevity and testosterone [13]. Nature 368: 408	Comment paper
Perez, G.I., Jurisicova, A., Wise, L., Lipina, T., Kanisek, M., Bechard, A., Takai, Y., Hunt, P., Roder, J., Grynpas, M., Tilly, J.L. (2007) Absence of the proapoptotic Bax protein extends fertility and alleviates age-related health complications in female mice. Proceedings of the National Academy of Sciences of the United States of America 104: 5229-5234	No OVX survival data
Planas-Silva, M.D., Rutherford, T.M., Stone, M.C. (2008) Prevention of age-related spontaneous mammary tumors in outbred rats by late ovariectomy. Cancer Detection and Prevention 32: 65-71	No survival data without surgical manipulation or removal of animals due to tumur size
Ratsimamanga, A.R., Ratsimam, S., Nigeondu, M. (1974) study of survival of adrenalectomized and castrated adult rat. Comptes Rendus Hebdomadaires Des Seances De L Academie Des Sciences Serie D 278: 901-905	Castrated animals also had adrenal removed
Reece, J.F., Chawla, S.K., Hiby, E.F., Hiby, L.R. (2008) Fecundity and longevity of roaming dogs in Jaipur, India. BMC Veterinary Research 4: 6	No control group
Reed, W.L., Clark, M.E., Parker, P.G., Raouf, S.A., Arguedas, N., Monk, D.S., Snajdr, E., Nolan Jr., V., Ketterson, E.D. (2006) Physiological effects on demography: A long-term experimental study of testosterone's effects on fitness. American Naturalist 167: 667-683	No sterilization

Reid, S.W.J., Mohammed, H.O. (1997) Longitudinal and cross-sectional Studies to evaluate the risk of sarcoid associated with castration. Canadian Journal of Veterinary Research 61: 89-93	Time to get a disease
Robertson, O.H., Wexler, B.C. (1962) Histological changes in the organs and tissues of senile castrated kokanee salmon (Oncorhynchus nerka kennerlyi). General and Comparative Endocrinology 2: 458-472	No survival data
Roubinian, J.R., Talal, N., Greenspan, J.S., Goodman, J.R., Siiteri, P.K. (1978) Effect of castration and sex-hormone treatment on survival, anti-nucleic acid antibodies, and glomerulonephritis in nzb-nzw F1 mice. Journal Of Experimental Medicine 147: 1568-1583	Disease model
Sanches, I.C., Conti, F.F., Bernardes, N., De O Brito, J., Galdini, E.G., Cavaglieri, C.R., Irigoyen, M.-C., De Angelis, K. (2015) Impact of combined exercise training on cardiovascular autonomic control and mortality in diabetic ovariectomized rats. Journal of Applied Physiology 119: 656-662	Do ovariectomy group without manipulation of diabetes, and no proper survival data
Sandberg, S.I., Barnes, B.A., Weinstein, M.C., Braun, P. (1985) Elective hysterectomy. Benefits, risks, and costs. Medical care 23: 1067-85	No data
Schwabl, H., Holmes, D., Strasser, R., Scheuerlein, A. (2012) Embryonic exposure to maternal testosterone influences age-specific mortality patterns in a captive passerine bird. Age 34: 87-94	No hormone removal/sterilization
Seidler, R.G., Gese, E.M. (2012) Territory fidelity, space use, and survival rates of wild coyotes following surgical sterilization. Journal of Ethology 30: 345-354	Could not separate sexes ad those that died were shot
Seidler, R.G., Gese, E.M., Conner, M.M. (2014) Using sterilization to change predation rates of wild coyotes: A test case involving pronghorn fawns. Applied Animal Behaviour Science 154: 83-92	No survival data
Smith, S.M. (1994) Statistical scrotal effect [9]. Nature 368: 501-502	Comment on previous paper
Sockman, K.W., Schwabl, H. (2000) Yolk androgens reduce offspring survival. Proceedings of the Royal Society B: Biological Sciences 267: 1451-1456	No sterilization
Stupfel, M., Bouley, G., Tantcheva, K. (1970) Castration and mortality of mice due to hypoxia. Journal de Physiologie 62: 218-219	Only assessed survival under hypoxia
Tortosa, A., Zumpano, R., Ardiles, I., Berra, Y., Faigenbaum, A.N., Guido, G.G., Castro, J., Molina, J.L., Marcos, E.R., Degregorio, O.J. (2016) Characterization of pet ownership in Buenos Aires city, Argentina. Revista de Investigaciones Veterinarias del Peru 27: 631-643	No proper survival data
Von Eyben, F.E., Graugaard, C., Vaeth, M. (2006) Erratum: All-cause mortality and mortality of myocardial infarction for 989 legally castrated men (European Journal of Epidemiology (2005) 20 (863-869) DOI: 10.1007/s10654-005-2150-0). European Journal of Epidemiology 21: 83	No comparable control data; Erratum

von Eyben, F.E., Graugaard, C., Vaeth, M. (2005) All-cause mortality and mortality of myocardial infarction for 989 legally castrated men. European Journal of Epidemiology 20: 863-869	No comparable control data
Wallis, L.J., Szabo, D., Erdelyi-Belle, B., Kubinyi, E. (2018) Demographic change across the lifespan of pet dogs and their impact on health status. Frontiers in Veterinary Science 5: 200	No actual lifespan/survival data
Warden, S.J., Galley, M.R., Hurd, A.L., Wallace, J.M., Gallant, M.A., Richard, J.S., George, L.A. (2013) Elevated mechanical loading when young provides lifelong benefits to cortical bone properties in female rats independent of a surgically induced menopause. Endocrinology 154: 3178-3187	No proper survival data and surgical removal of cancers
Waters, D.J., Kengeri, S.S., Maras, A.H., Suckow, C.L., Chiang, E.C. (2017) Life course analysis of the impact of mammary cancer and pyometra on age-anchored life expectancy in female Rottweilers: Implications for envisioning ovary conservation as a strategy to promote healthy longevity in pet dogs. Veterinary Journal (London, England : 1997) 224: 25-37	No data on intact females
Whisson, D.A., Holland, G.J., Carlyon, K. (2012) Translocation of overabundant species: Implications for translocated individuals. Journal of Wildlife Management 76: 1661-1669	No comparable control data
Yonker, J.A., Chang, V., Roetker, N.S., Hauser, T.S., Hauser, R.M., Atwood, C.S. (2013) Hypothalamic-pituitary-gonadal axis homeostasis predicts longevity. Age 35: 129-138	Data for sterilized groups not provided
Zielinski, W.J., Vandenbergh, J.G. (1991) Increased survivorship of testosterone-treated female house mice, Mus musculus, in high-density field conditions. Animal Behaviour 42: 955-967	No manipulation or control
Zielinski, W.J., Vandenbergh, J.G. (1993) Testosterone and competitive ability in male house mice, Mus musculus: Laboratory and field studies. Animal Behaviour 45: 873-891	No manipulation or control

Supplementary Table 7. Full reference list of all studies included in the meta-analysis on changes in survival with sterilization.

Reference
Aida, K., Kato, T. & Awaji, M. Effects of Castration on the Smoltification of Precocious Male Masu Salmon Oncorhynchus masou. *Bulletin of the Japanese Society of Scientific Fisheries* **50**, 565-571 (1984).
Arriola Apelo, S. I. *et al.* Ovariectomy uncouples lifespan from metabolic health and reveals a sex-hormone-dependent role of hepatic mTORC2 in aging. *Elife* **9**, doi:10.7554/eLife.56177 (2020).
Asdell, S. A., Doornenbal, H., Joshi, S. R. & Sperling, G. A. The effects of sex steroid hormones upon longevity in rats. *J. Reprod. Fert.* **14**, 113-120 (1967).
Asdell, S. A. & Joshi, S. R. Reproduction and longevity in the hamster and rat. *Biol Reprod* **14**, 478-480, doi:10.1095/biolreprod14.4.478 (1976).
Benedusi, V. *et al.* Ovariectomy shortens the life span of female mice. *Oncotarget* **6**, 10801-10811, doi:10.18632/oncotarget.2984 (2015).
Bronson, R. T. Age at death of necropsied intact and neutered cats. *American Journal of Veterinary Research* **42**, 1606-1608 (1981).
Bronson, R. T. Variation in age at death of dogs of different sexes and breeds. *Am J Vet Res* **43**, 2057-2059 (1982).
Cargill, S. L., Carey, J. R., Muller, H. G. & Anderson, G. Age of ovary determines remaining life expectancy in old ovariectomized mice. *Aging Cell* **2**, 185-190, doi:10.1046/j.1474-9728.2003.00049.x (2003).
Cheng, C. J. Sex differences in longevity and lifespan-extending drug efficacy in genetically heterogeneous mice: age-specific effects and gonadal contributions. *Thesis, The university of Texas health Science center at San Antonio* (2019).
Collins, G. H. & Kasbohm, J. Population Dynamics and Fertility Control of Feral Horses. *The Journal of Wildlife Management* **81**, 289-296 (2017).
Cox, R. M. & Calsbeek, R. Severe costs of reproduction persist in Anolis lizards despite the evolution of a single-egg clutch. *Evolution* **64**, 1321-1330, doi:10.1111/j.1558-5646.2009.00906.x (2009).
Cox, R. M., Lovern, M. B. & Calsbeek, R. Experimentally decoupling reproductive investment from energy storage to test the functional basis of a life-history trade-off. *J Anim Ecol* **83**, 888-898, doi:10.1111/1365-2656.12228 (2014).
Cox, R. M. *et al.* Experimental evidence for physiological costs underlying the trade-off between reproduction and survival. *Functional Ecology* **24**, 1262-1269, doi:10.1111/j.1365-2435.2010.01756.x (2010).
Cox, R. M., Wittman, T. N. & Calsbeek, R. Reproductive trade-offs and phenotypic selection change with body condition, but not with predation regime, across island lizard populations. *J Evol Biol* **35**, 365-378, doi:10.1111/jeb.13926 (2022).
Deleuze, S. *et al.* Tubectomy of Pregnant and Non-pregnant Female Balinese Macaques (Macaca Fascicularis) With Post-operative Monitoring. *Front Vet Sci* **8**, 688656, doi:10.3389/fvets.2021.688656 (2021).

Dorner, G. Sex hormone dependent brain organization and duration of life. *Z. Alternsforsch.* **27**, 379-380 (1973).

Drori, D. & Folman, Y. Interactive environmental and genetic effects on longevity in the male rat: litter size, exercise, electric shocks and castration. *Exp Aging Res* **12**, 59-64, doi:10.1080/03610738608259437 (1986).

Garratt, M., Try, H. & Brooks, R. C. Access to females and early life castration individually extend maximal but not median lifespan in male mice. *Geroscience* **43**, 1437-1446, doi:10.1007/s11357-020-00308-8 (2021).

Gipps, J. H. W. & Jewell, P. A. Maintaining Populations of Bank Voles, Clethrionomys glareolus, in Large Outdoor Enclosures, and Measuring the Response of Population Variables to the Castration of Males. *Journal of Animal Ecology* **48**, 535-555 (1979).

Hamilton, J. B. Relationship of Castration, Spaying, and Sex to Survival and Duration of Life in Domestic Cats. *J. Gerontol.* **20**, 96-104 (1965).

Hamilton, J. B. & Mestler, G. E. Mortality and Survival: Comparison of Eunuchs with Intact Men and Women in a Mentally Retarded Population. *Journal of Gerontology* **24**, 395-411 (1969).

Hamilton, J. B., S.Hamilton, R. & Mestler, G. E. Duration of Life and Causes of Death in Domestic Cats: Influence of Sex, Gonadectomy, and Inbreeding. *Journal of Gerontology* **24**, 427-437 (1969).

Hoffman, J. M., O'Neill, D. G., Creevy, K. E. & Austad, S. N. Do Female Dogs Age Differently Than Male Dogs? *J Gerontol A Biol Sci Med Sci* **73**, 150-156, doi:10.1093/gerona/glx061 (2018).

Holland, J. M., Mitchell, T. J. & Walburg, H. E. J. Effects of Prepubertal Ovariectomy on Survival and Specific Diseases in Female RFM Mice Given 300 R of X Rays. *radiation research* **69**, 317-327 (1977).

Hotchkiss, C. E. Effect of surgical removal of subcutaneous tumors on survival of rats. *J Am Vet Med Assoc* **206**, 1575-1579 (1995).

Howard, B. V. *et al.* Risk of cardiovascular disease by hysterectomy status, with and without oophorectomy: the Women's Health Initiative Observational Study. *Circulation* **111**, 1462-1470, doi:10.1161/01.Cir.0000159344.21672.Fd (2005).

Huang, W. H., Liao, A. T., Chu, P. Y., Yen, I. F. & Liu, C. H. A real-time reporting system of causes of death or reasons for euthanasia: A model for monitoring mortality in domesticated cats in Taiwan. *Prev Vet Med* **137**, 59-68, doi:10.1016/j.prevetmed.2016.12.011 (2017).

Huang, W. H. *et al.* A 3-year surveillance on causes of death or reasons for euthanasia of domesticated dogs in Taiwan. *Preventive Veterinary Medicine* **147**, 1-10, doi:10.1016/j.prevetmed.2017.08.015 (2017).

Iversen, L., Hannaford, P. C. & Elliott, A. M. Tubal sterilization, all-cause death, and cancer among women in the United Kingdom: evidence from the Royal College of General Practitioners' Oral Contraception Study. *Am J Obstet Gynecol* **196**, 447.e441-448, doi:10.1016/j.ajog.2007.01.022 (2007).

Iwasa, T., Matsuzaki, T., Yano, K. & Irahara, M. The effects of ovariectomy and lifelong high-fat diet consumption on body weight, appetite, and lifespan in female rats. *Horm Behav* **97**, 25-30, doi:10.1016/j.yhbeh.2017.10.005 (2018).

Jacob, J., Aini Herawati, N. U. R., Davis, S. A. & Singleton, G. R. The Impact of Sterilized Females on Enclosed Populations of Ricefield Rats. *Journal of Wildlife Management* **68**, 1130-1137, doi:10.2193/0022-541x(2004)068[1130:Tiosfo]2.0.Co;2 (2004).

Jacob, J., Justianus, M. & Sudarmaji. Effects of imposed sterility on movement patterns of female ricefield rats. *J. Wildl. Manage.* **68**, 1138-1144 (2004).

Jewell, P. A. Survival and behaviour of castrated Soay sheep (Ovis aries) in a feral island population on Hirta, St. Kilda, Scotland. *J. Zool. Lond.* **243**, 623-636 (1997).

Kent, M. S., Burton, J. H., Dank, G., Bannasch, D. L. & Rebhun, R. B. Association of cancer-related mortality, age and gonadectomy in golden retriever dogs at a veterinary academic center (1989-2016). *PLoS One* **13**, e0192578, doi:10.1371/journal.pone.0192578 (2018).

Kirkman, H. & Yau, P. K. S. Longevity of Male and Female, Intact and Gonadectomized, Untreated and Hormone-treated, Neoplastic and Non-neoplastic Syrian Hamsters. *Am. J. Anat.* **135**, 205-220 (1972).

Kirkpatrick, J. F. & Turner, A. Immunocontraception and Increased Longevity in Equids. *Zoo Biology* **26**, 237-244, doi:10.1002/zoo.20109 (2007).

Larsen, L. O. Effects of gonadectomy in the cyclostome, Lampetra jluviatilis. *Gen. Comp. Endocrinol.* **13**, 516-517 (1969).

Larsen, L. O. Development in Adult, Fresh-water River Lampreys and its Hormonal Control. Starvation, Sexual Maturation and Natural Death. *Thesis, Copenhagen* (1973).

Manson, J. E. *et al.* Menopausal hormone therapy and health outcomes during the intervention and extended poststopping phases of the Women's Health Initiative randomized trials. *Jama* **310**, 1353-1368, doi:10.1001/jama.2013.278040 (2013).

Mason, J. B., Cargill, S. L., Anderson, G. B. & R.Carey, J. Transplantation of Young Ovaries to Old Mice Increased Life Span in Transplant Recipients. *Journal of Gerontology* **64**, 1207-1211, doi:10.1093/gerona/glp134 (2009).

Michell, A. R. Longevity of British breeds of dog and its relationships with sex, size, cardiovascular variables and disease. *The Veterinary Record* **145**, 625-629 (1999).

Min, K. J., Lee, C. K. & Park, H. N. The lifespan of Korean eunuchs. *Curr Biol* **22**, R792-793, doi:10.1016/j.cub.2012.06.036 (2012).

Moore, G. E., Burkman, K. D., Carter, M. N. & Peterson, M. R. Causes of death or reasons for euthanasia in military working dogs: 927 cases (1993-1996). *JAVMA* **29** (2001).

Muhlbock, O. Factors Influencing the Life-Span of Inbred Mice. *Gerontologia* **3**, 177-183 (1959).

Nieschlag, E., Nieschlag, S. & Behre, H. M. Lifespan and testosterone. *Nature* **366**, 215 (1993).

O'Neill, D. G., Church, D. B., McGreevy, P. D., Thomson, P. C. & Brodbelt, D. C. Longevity and mortality of owned dogs in England. *The Veterinary Journal* **198**, 638-643, doi:10.1016/j.tvjl.2013.09.020 (2013).

O'Neill, D. G., Church, D. B., McGreevy, P. D., Thomson, P. C. & Brodbelt, D. C. Longevity and mortality of cats attending primary care veterinary practices in England. *Journal of Feline Medecine and Surgery* **17**, 125-133, doi:10.1177/1098612X14536176 (2015).

Ossewaarde, M. E. *et al.* Age at menopause, cause-specific mortality and total life expectancy. *Epidemiology* **16**, 556-562, doi:10.1097/01.ede.0000165392.35273.d4 (2005).

Phelan, J. P. Reproductive Costs and Longevity in the House Mouse. *Thesis, Harvard University* (1995).

Pullinger, B. D. & M.A. Head, M. A. Hepatoma in intact C3Hf male and virgin female mice and after gonadectomy alone and subsequent treatment with oestrogen. *Br J Cancer* **18**, 521-527 (1964).

Ramsey, D. Population dynamics of brushtail possums subject to fertility control. *Journal of Applied Ecology* **42**, 348-360, doi:10.1111/j.1365-2664.2005.01006.x (2005).

Ramsey, D. S. L. *et al.* Long-term fertility control reduces overabundant koala populations and mitigates their impacts on food trees. *Biological Conservation* **253**, doi:10.1016/j.biocon.2020.108870 (2021).

Reedy, A. M., Cox, C. L., Chung, A. K., Evans, W. J. & Cox, R. M. Both sexes suffer increased parasitism and reduced energy storage as costs of reproduction in the brown anole, Anolis sagrei. *Biological Journal of Linnean Society* **117**, 516-527 (2015).

Robertson, O. H. Prolongation of the life span of Kokanee salmon (Oncorhynchus Nerka Kennerlyi) by castration before beginning of gonad development. *Proc Natl Acad Sci U S A* **47**, 609-621, doi:10.1073/pnas.47.4.609 (1961).

Rocca, W. A., Grossardt, B. R., de Andrade, M., Malkasian, G. D. & Melton, L. J., 3rd. Survival patterns after oophorectomy in premenopausal women: a population-based cohort study. *Lancet Oncol* **7**, 821-828, doi:10.1016/s1470-2045(06)70869-5 (2006).

Sato, M., Zeng, G. Q. & Turner, C. H. Biosynthetic human parathyroid hormone (1-34) effects on bone quality in aged ovariectomized rats. *Endocrinology* **138**, 4330-4337, doi:10.1210/endo.138.10.5440 (1997).

Saunders, G. *et al.* The effects of induced sterility on the territorial behaviour and survival of foxes. *Journal of Applied Ecology* **39**, 56-66 (2002).

Sichuk, G., Bettigole, R. E., Der Bela, K. & Fortner, J. G. Influence of sex hormones on thrombosis of left atrium in Syrian (golden) hamsters. *Am. J. Physiol.* **208**, 465- 470 (1965).

Skinner, B. Surgical sterilization of female white-tailed deer in suburban Chicago, Illinois : social causes and biological impacts. *Thesis UW-Madison* (2008).

Slonaker, J. R. The effect of the excision of different sexual organs on the development, growth and longevity of the albino rat. *Am. J. Physiology* (1930).

Storer, J. B., Mitchell, T. J. & Ullrich, R. L. Causes of Death and Their Contribution to Radiation-Induced Life Shortening in Intact and Ovariectomized Mice. *Radiation Research* **89**, 618-643 (1982).

Talbert, G. B. & Hamilton, J. B. Duration of life in Lewis Strain of rats after gonadectomy at birth and at older ages. *Reproduction and aging* (1965).

Tapprest, J. *et al.* Fallen stock data: An essential source of information for quantitative knowledge of equine mortality in France. *Equine Vet J* **49**, 596-602, doi:10.1111/evj.12664 (2017).

Tidière, M. The diversity of actuarial senescence across mammals: Demographic data from captive populations reveal the influence of sexual selection. *Thesis Université de Lyon* (2016).

Twigg, L. E. *et al.* Effects of surgically imposed sterility on free-ranging rabbit populations. *Journal of Applied Ecology* **37**, 16-39 (2000).

Urfer, S. R., Kaeberlein, M., Promislow, D. E. L. & Creevy, K. E. Lifespan of companion dogs seen in three independent primary care veterinary clinics in the United States. *Canine Med Genet* **7**, 7, doi:10.1186/s40575-020-00086-8 (2020).

Urfer, S. R., Wang, M., Yang, M., Lund, E. M. & Lefebvre, S. L. Risk Factors Associated with Lifespan in Pet Dogs Evaluated in Primary Care Veterinary Hospitals. *J Am Anim Hosp Assoc* **55**, 130-137, doi:10.5326/JAAHA-MS-6763 (2019).

Wang, Z., Wu, W. & Cai, D. GnRH pulse frequency and irregularity play a role in male aging. *Nature Aging* **1**, 904-918 (2021).

Waters, D. J., Kengeri, S. S., Maras, A. H. & Chiang, E. C. Probing the perils of dichotomous binning: how categorizing female dogs as spayed or intact can misinform our assumptions about the lifelong health consequences of ovariohysterectomy. *Theriogenology* **76**, 1496-1500, doi:10.1016/j.theriogenology.2011.06.017 (2011).

Williams, C. K. *et al.* Population responses to sterility imposed on female European rabbits. *Journal of Applied Ecology* **44**, 291-301, doi:10.1111/j.1365-2664.2006.01264.x (2007).

Wilson, L. F., Pandeya, N., Byles, J. & Mishra, G. D. Hysterectomy status and all-cause mortality in a 21-year Australian population-based cohort study. *Am J Obstet Gynecol* **220**, 83 e81-83 e11, doi:10.1016/j.ajog.2018.10.002 (2019).

Zakeri, M. *et al.* Pro-neurocognitive and anti-sarcopenic benefits of one-year metformin therapy in ovariectomized aged mice. *Clin Exp Pharmacol Physiol* **46**, 1133-1140, doi:10.1111/1440-1681.13149 (2019).

Supplementary Table 8. List of recommended assays and techniques for measuring healthspan. The list has been aggregated from Richardson, A., Fischer, K. E., Speakman, J. R., de Cabo, R., Mitchell, S. J., Peterson, C. A., Rabinovitch, P., Chiao, Y. A., Taffet, G., Miller, R. A., Rentería, R. C., Bower, J., Ingram, D. K., Ladiges, W. C., Ikeno, Y., Sierra, F., & Austad, S. N. (2016). Measures of Healthspan as Indices of Aging in Mice-A Recommendation. The journals of gerontology. Series A, Biological sciences and medical sciences, 71(4), 427–430. https://doi.org/10.1093/gerona/glv080 and from Bellantuono, I., de Cabo, R., Ehninger, D., Di Germanio, C., Lawrie, A., Miller, J., Mitchell, S. J., Navas-Enamorado, I., Potter, P. K., Tchkonia, T., Trejo, J. L., & Lamming, D. W. (2020). A toolbox for the longitudinal assessment of healthspan in aging mice. Nature protocols, 15(2), 540–574. https://doi.org/10.1038/s41596-019-0256-1.

Last column lists measurements that will be excluded from our meta-analysis, with reason. Listed outcomes for pathology assessment are collated from two additional sources as specifics for pathology assessment are not outlined in the earlier healthspan articles: Pettan-Brewer, C., & M. Treuting, PiperM. (2011). Practical pathology of aging mice. Pathobiology of Aging & Age-Related Diseases, 1(1). https://doi.org/10.3402/pba.v1i0.7202; Snyder JM, Snider TA, Ciol MA, Wilkinson JE, Imai DM, Casey KM, Vilches-Moure JG, Pettan-Brewer C, Pillai SPS, Carrasco SE, Salimi S, Ladiges W. Validation of a geropathology grading system for aging mouse studies. Geroscience. 2019 Aug;41(4):455-465. doi: 10.1007/s11357-019-00088-w.

Outcome Type	Outcome Sub-type	Functional Assay	Direction indicating improved health	Exclude?
Activity, Energetics and Metabolism	Food or Energy Intake	Weight of food consumed		Exclude - unclear interpretation of effect direction
Activity, Energetics and Metabolism	Food or Energy Intake	Energy consumed		Exclude - unclear interpretation of effect direction
Activity, Energetics and Metabolism	Food or Energy Intake	Energy absorbed		Exclude - unclear interpretation of effect direction
Activity, Energetics and Metabolism	Food or Energy Intake	Metabolisable energy intake		Exclude - unclear interpretation of effect direction
Activity, Energetics and Metabolism	Body Composition	Measures on dead animals		Exclude - unclear interpretation of effect direction
Activity, Energetics and Metabolism	Body Composition	Isotope dilution		Exclude - unclear interpretation of effect direction
Activity, Energetics and Metabolism	Body Composition	TOBEC/BIA		Exclude - unclear interpretati

				on of effect direction
Activity, Energetics and Metabolism	Body Composition	DXA		Exclude - unclear interpretation of effect direction
Activity, Energetics and Metabolism	Body Composition	QMS		Exclude - unclear interpretation of effect direction
Activity, Energetics and Metabolism	Body Composition	CT		Exclude - unclear interpretation of effect direction
Activity, Energetics and Metabolism	Energy Expenditure	Doubly-labeled water	Increased	
Activity, Energetics and Metabolism		Indirect calorimetry	Increased	
Activity, Energetics and Metabolism	Activity	Motion sensor systems	Increased	
Activity, Energetics and Metabolism		Implanted transmitter devices (activity)	Increased	
Activity, Energetics and Metabolism		Open field test (activity)	Increased	
Activity, Energetics and Metabolism		Running wheels	Increased	
Activity, Energetics and Metabolism		Video systems (activity)	Increased	
Activity, Energetics and Metabolism		Glucose tolerance test (GTT)	Decreased	
Activity, Energetics and Metabolism		Insulin tolerance test (ITT)	Decreased	
Skeletal Muscle Function		Muscle size	Increased	

Skeletal Muscle Function		Fiber size	Increased	
Skeletal Muscle Function		Fiber pathology/morphology (presence of centralized nuclei, angular atrophied fibers etc)	Increased	
Skeletal Muscle Function		Nerve conduction velocity	Increased	
Skeletal Muscle Function		Strength/motor performance	Increased	
Skeletal Muscle Function	Grip strength	Rota-Rod	Increased	
Skeletal Muscle Function	Grip strength	Cage top (grid hanging test, hanging bar, hanging wire)	Increased	
Skeletal Muscle Function	Grip strength	Hanging bar (wire suspension latency test, wire hang, hang test, or body suspension)	Increased	
Skeletal Muscle Function	Grip strength	Isometric force (*in vitro*)	Increased	
Skeletal Muscle Function	Endurance/Exercise tolerance	P-NMR (mito function)		Exclude unless clear direction of change with age within study
Skeletal Muscle Function	Endurance/Exercise Tolerance	Treadmill to exhaustion	Increased	
Cardiac Function		Echocardiography Heart rate		Exclude as direction of change unclear and different according

			to relative age
Cardiac Function	Echocardiography LV mass (relative to body mass)	Decreased	
Cardiac Function	Echocardiography LV wall thickness	Decreased	
Cardiac Function	Echocardiography IVRT: isovolumetric relaxation time	Decreased	
Cardiac Function	Echocardiography LV fractional shortening (FS)	Increased	
Cardiac Function	Echocardiography Ejection fraction (EF)	Increased	
Cardiac Function	Echocardiography Mycocardial performance index (MPI)	Decreased	
Cardiac Function	Echocardiography Diastolic function Ea/Aa	Increased	
Cardiac Function	Echocardiography E/A ratio	Increased	
Cardiac Function	Heart size relative to tibia or body weight	Decreased	
Cardiac Function	Myoctye fiber size	Decreased	
Cardiac Function	Cardiac fibrosis	Decreased	
Cardiac Function	Histopathological features (mineralization, sarcomere collapse, vacuolization, arteriolosclerosis)	Decreased	

Cardiac Function	Cardiac Catheter	Include only values where there is a clear expected direction of change with ageing	
Cardiac Function	Magnetic resonance imaging and spectroscopy	Include only values where there is a clear expected direction of change with ageing	
Cardiac Function	Isolated heart perfusion (with or without P-MRS)	Include only values where there is a clear expected direction of change with ageing	
Cardiac Function	Blood Pressure		Exclude as does not change with aging and is not informative in mice (see Richardson et al)
Cardiac Function	Aortic Impedance	Decreased	
Pulmonary Function	Histology and Gross Pathology (Elastin content increased collagen to elastin ratio)	Increased	
Pulmonary Function	Histology and Gross Pathology (Collagen to elastin ratio)	Decreased	
Pulmonary Function	Respiratory Impedance/lung capacity		Exclude unless clear direction of change with age within study as there are

		strain and sex differences in response (i.e. Huang et al 2016, Glassberg et al 2007)
Pulmonary Function	Hypoxia and hypercapnia responses	Exclude unless clear direction of change with age within study
Pulmonary Function	Invasive measurements	Exclude unless clear direction of change with age within study
Immune Function	CD4 memory cells, blood	Exclude unless clear direction of change with age within study (see Richardson et al for discussion)
Immune Function	CD8 memory cells, blood	Exclude unless clear direction of change with age within study (see Richardson et al for discussion)
Immune Function	CD4 and CD8 memory, spleen	Exclude unless clear direction of change with age within study (see Richardson

				et al for discussion)
Immune Function		T cell function tests in vitro	Improved responsiveness	
Immune Function		Cell mediated immunity in vivo	Increased responsiveness	
Immune Function		Antibody production in vivo	Increased antibodies	
Sensory Function	Hearing	ABR	Improved hearing range	
Sensory Function	Hearing	DPOAE	Improved hearing range	
Sensory Function	Hearing	Startle test	Increased	
Sensory Function	Olfaction	Odor detection	Increased	
Sensory Function	Olfaction	Odor discrimination	Increased	
Sensory Function	Olfaction	Odor Identification	Increased	
Sensory Function	Vision	Cataracts	Reduced	
Cognition		Fear Conditioning	Better conditioning	
Cognition		Eye Blink Conditioning	Better conditioning	
Cognition		Novel Location Recognition	Greater time with novel location (increased)	
Cognition		Novel Object Recognition	Greater time with novel object (increased)	
Cognition		Avoidance Learning	Fewer trials to learn to avoid (decreased)	
Cognition		Morris Water Maze/other maze tests of memory	Less time/distance (use distance if supplied) to get to platform. Focus on probe test and crossings/distance travelled to get	

			to quadrat if supplied. Less distance/time is better, or more crossings of quadrat
Cognition		Stone Maze	Fewer errors
Cognition		Food Burrowing	Quicker time
Frailty assessment	Howlett and Rockwood clinical frailty index	Frailty score	Decreased
Pathology cross sectional		Presence of kidney lesions (Nephropathy, Pyelonephritis, Infarct, Lymphoid aggregates,Mineralization, Amyloid, Tumor, CLS)	Decreased
Pathology cross sectional		Presence of heart lesions (Cardiomyopathy/myocardial fibrosis, Myocardial inflammation, Valvular myxomatosis,Lymphoid aggregates, Atrial thrombosis,Tumor, CLS)	Decreased
Pathology cross sectional		Presence of liver lesions (Hepatic degeneration/necrosis, Hepatic lipidosis, Central venous congestion, Periportal inflammation Bile duct	Decreased

	hyperplasia/cyst, Lymphoid aggregates,Microgranuloma,Ito cell hyperplasia, Telangiectasia, Tumor,CLS)	
Pathology cross sectional	Presence of lung lesions (Alveolar acidophilic macrophage pneumonia, Alveolar histiocytosis,Foam cells, Heart failure cells (chronic passive congestion), Interstitial pneumonia,Perivascular inflammation,Bronchial/bronchiolar inflammation, Airway metaplasia or hyperplasia, Vascular hypertrophy, Pulmonary fibrosis, Atelectasis, Lymphoid aggregates, Tumor,CLS)	Decreased
Pathology cross sectional	Presence/absence or severity of neoplastic disease (Fibrosarcoma, Hemangioma, Hemangiosarcoma, Hematopoietic neoplasia (Malignant lymphoma or Histiocytic sarcoma), Hepatic adenoma, Leiomyosarcoma, Malignant Pheochromocytom	Decreased

	a, Osteosarcoma, Ovarian granulosa cell tumor Pituitary adenocarcinom, Pituitary adenoma, Pulmonary adenocarcinom, Pulmonary adenoma, Squamous cell carcinoma, Squamous papilloma, Testicular interstitial cell adenoma)	
Pathology cross sectional	Presence/absence or severity of non-neoplastic disease (Acidophilic macrophage pneumonia, Amyloid, glomerular, Amyloid, intestinal, Biliary hyperplasia, Cataracts or cornea opacity, Chronic infarct, Cystic endometrial hyperplasia, Heart lesions, Hepatic cysts, Hydronephrosi, Nephropathy, Polyarteritis, Preputial cystic adenitis,Rectal prolapse, Seminal vesiculitis, cystic degeneration Skin lesions, Systemic antigenic stimulation, Ulcerative keratitis)	Decreased

Pathology cross sectional	Presence of collagen/fibrosis accumulation at a specific time point	Decreased	
Pathology end o life	Presence/severity of pathological observations (listed above) at death	Decreased	
Pathology end o life	Cause of death		Exclude

Supplementary Table 9. List of excluded full-text studies with reasons for exclusion for meta-analysis of changes in healthspan with gonadectomy.

Reference	**Main exclusion reason**
Alibhai, F.J. *et al.* Female ClockΔ19/Δ19 mice are protected from the development of age-dependent cardiomyopathy. *Cardiovasc Res* **114**, 259-271 (2018).	No appropriate control group
Baeza, I., De Castro, N.M., Arranz, L. & De la Fuente, M. Soybean and green tea polyphenols improve immune function and redox status in very old ovariectomized mice. *Rejuvenation Res* **13**, 665-674 (2010).	Surgery conducted after cut-off age
Baeza, I., De Castro, N.M., Arranz, L., Fdez-Tresguerres, J. & De la Fuente, M. Ovariectomy causes immunosenescence and oxi-inflamm-ageing in peritoneal leukocytes of aged female mice similar to that in aged males. *Biogerontology* **12**, 227-238 (2011).	Surgery conducted after cut-off age
Baeza, I., De Castro, N.M., Giménez-Llort, L. & De la Fuente, M. Ovariectomy, a model of menopause in rodents, causes a premature aging of the nervous and immune systems. *Journal of neuroimmunology* **219**, 90-99 (2010).	Insufficent time between surgery and testing
Barragán-Ceballos, N.G. *et al.* A chronic low dosage of taurine induces muscle weakness in castrated-aged mice. *Translational Medicine of Aging* **5**, 54-61 (2021).	Insufficent time between surgery and testing
Bimonte-Nelson, H.A. *et al.* Ovarian Hormones and Cognition in the Aged Female Rat: I. Long-Term, but Not Short-Term, Ovariectomy Enhances Spatial Performance. *Behavioral Neuroscience* **117**, 1395-1406 (2003).	Surgery conducted after cut-off age
Douillet, C., Borson-Chazot, F., Renaud, S. & Ciavatti, M. Long-term ovariectomy effect on plasma glucose, lipids, antioxidant vitamins, and lipid peroxides: Relationship with aortic cholesterol and fatty acid distribution in rats. *Journal of Clinical Biochemistry and Nutrition* **23**, 15-25 (1997).	Has not measured an included healthspan parameter
Dynlacht, J.R. *et al.* Age and Hormonal Status as Determinants of Cataractogenesis Induced by Ionizing Radiation. I. Densely Ionizing (High-LET) Radiation. *Radiation research* **175**, 37-43 (2011).	No appropriate control group
Frye, C.A., Edinger, K.L., Lephart, E.D. & Walf, A.A. 3α-androstanediol, but not testosterone, attenuates age-related decrements in cognitive, anxiety, and depressive behavior of male rats. *Frontiers in aging neuroscience* **2** (2010).	Insufficent time between surgery and testing
Bimonte-Nelson, H.A. *et al.* Ovarian Hormones and Cognition in the Aged Female Rat: I. Long-Term, but Not Short-Term, Ovariectomy Enhances Spatial Performance. *Behavioral Neuroscience* **117**, 1395-1406 (2003).	Surgery conducted after cut-off age
Douillet, C., Borson-Chazot, F., Renaud, S. & Ciavatti, M. Long-term ovariectomy effect on plasma glucose, lipids, antioxidant vitamins, and lipid peroxides: Relationship with aortic cholesterol and fatty acid distribution in rats. *Journal of Clinical Biochemistry and Nutrition* **23**, 15-25 (1997).	Has not measured an included healthspan parameter

Glassberg, M.K. *et al.* 17β-estradiol replacement reverses age-related lung disease in estrogen-deficient C57BL/6J mice. *Endocrinology* **155**, 441-448 (2014).	No appropriate control group, surgery too late
Hofmann, C., Oslapas, R., Nayyar, R., McCall, A. & Paloyan, E. Testosterone enhancement of thyroid carcinoma in rats. The role of TSH. *Surgery* **100**, 1078-1087 (1986).	Could not access full text
Kimura, T. & Nandi, S. Nature of induced persistent vaginal cornification in mice. iv. changes in the vaginal epithelium of old mice treated neonatally with estradiol or testosterone. *Journal of the National Cancer Institute* **39**, 75-93 (1967).	Has not measured an included healthspan parameter
Lubischer, J.L., Jordan, C.L. & Arnold, A.P. Transient and permanent effects of androgen during synapse elimination in the levator ani muscle of the rat. *J Neurobiol* **23**, 1-9 (1992).	No appropriate control group
Markowska, A.L. & Savonenko, A.V. Effectiveness of estrogen replacement in restoration of cognitive function after long-term estrogen withdrawal in aging rats. *J Neurosci* **22**, 10985-10995 (2002).	Surgery conducted after the cut-off age
Meranze, D.R., Gruenstein, M. & Shimkin, M.B. Effect of age and sex on the development of neoplasms in wistar rats receiving a single intragastric instillation of 7,12-dimethylbenz(a)anthracene. *International Journal of Cancer* **4**, 480-486 (1969).	Effects only assessed in a disease model
Mori, T., Bern, H.A., Mills, K.T. & Young, P.N. Long-term effects of neonatal steroid exposure on mammary-gland development and tumorigenesis in mice. *JNCI-Journal of the national cancer institute* **57**, 1057-1062 (1976).	No appropriate control group
Parkinson, K.C., Peterson, R.L. & Mason, J.B. Cognitive behavior and sensory function were significantly influenced by restoration of active ovarian function in postreproductive mice. *Experimental gerontology* **92**, 28-33 (2017).	No appropriate control group
Pleil, K.E., Glenn, M.J. & Williams, C.L. Estradiol alters Fos-immunoreactivity in the hippocampus and dorsal striatum during place and response learning in middle-aged but not young adult female rats. *Endocrinology* **152**, 946-956 (2011).	No appropriate control group
Said, S.A. *et al.* Effects of long-term dietary administration of estrogen receptor-beta agonist diarylpropionitrile on ovariectomized female ICR (CD-1) mice. *GeroScience* **40**, 393-403 (2018).	No appropriate control group
Sato, M. *et al.* Skeletal efficacy with parathyroid hormone in rats was not entirely beneficial with long-term treatment. *Journal of Pharmacology and Experimental Therapeutics* **302**, 304-313 (2002).	No data on an included healthspan parameter for surgically manipulated animals
Silberberg, R., Thomasson, R. & Silberberg, M. Degenerative joint disease in castrate mice .2. effects of orchiectomy at various ages. *archives of pathology* **65**, 442-444 (1958).	No data on an included healthspan parameter
Strickland, J.E., Saviolakis, G.A., Allen, P.T., Heilman, A. & Arnold, K.F. Spontaneous Adrenal Tumors in the Aged, Ovariectomized Nih Swiss	No data on an included healthspan parameter

Mouse without Enhanced Retrovirus Expression. *Cancer Research* **40**, 3570-3575 (1980).	
Toh, Y.C. Effect of neonatal castration on liver tumor induction by n-2-fluorenylacetamide in suckling balb/c mice. *Carcinogenesis* **2**, 1219-1221 (1981).	Within the groups to be included in the analysis, only one animal showed the pathology
Utsuyama, M., Kobayashi, S. & Hirokawa, K. Induction of thymic hyperplasia and suppression of splenic T cells by lesioning of the anterior hypothalamus in aging Wistar rats. *Journal of Neuroimmunology* **77**, 174-180 (1997).	No data on an included healthspan parameter

Supplementary Table 10. Full reference list of all studies included in the meta-analysis on changes in healthspan with gonadectomy in rodents.

Reference
Andervont, Howard B. 1950. 'Studies on the Occurrence of Spontaneous Hepatomas in Mice of Strains C3H and CBA2', *JNCI: Journal of the National Cancer Institute*, 11: 581-92.
Apelo, SIA, A Lin, JA Brinkman, E Meyer, M Morrison, JL Tomasiewicz, CP Pumper, EL Baar, NE Richardson, M Alotaibi, and DW Lamming. 2020. 'Ovariectomy uncouples lifespan from metabolic health and reveals a sex-hormone-dependent role of hepatic mTORC2 in aging', *eLife*, 9: e56177.
Ayaz O, Banga S, Heinze-Milne S, Rose RA, Pyle WG, and Howlett SE. 2019. 'Long-term testosterone deficiency modifies myofilament and calcium-handling proteins and promotes diastolic dysfunction in the aging mouse heart', *American journal of physiology. Heart and circulatory physiology*, 316: H768-H80.
Banga, S., S.D. Heinze-Milne, J. Godin, and S.E. Howlett. 2021. 'Signs of diastolic dysfunction are graded by serum testosterone levels in aging C57BL/6 male mice', *Mechanisms of Ageing and Development*, 198: 111523.
Banga, S., M. Mishra, S. D. Heinze-Milne, H. J. Jansen, R. A. Rose, and S. E. Howlett. 2023. 'Chronic testosterone deficiency increases late inward sodium current and promotes triggered activity in ventricular myocytes from aging male mice', *Am J Physiol Heart Circ Physiol*, 325: H264-H77.
Borbélyová, V., E. Domonkos, J. Bábíčková, Ľ Tóthová, M. Bosý, J. Hodosy, and P. Celec. 2016. 'No effect of testosterone on behavior in aged Wistar rats', *Aging (Albany NY)*, 8: 2848-61.
Borbélyová, V., Domonkos, E., Bábíčková, J., Tóthová, Ľ., Kačmárová, M., Uličná, O., Ostatníková, D., Hodosy, J. and Celec, P., 2017. Does long-term androgen deficiency lead to metabolic syndrome in middle-aged rats?. *Experimental Gerontology*, *98*, 38-46.
Borbélyová, V., V. Šarayová, E. Renczés, J. Čonka, J. Janko, K. Šebeková, K. Štefíková, D. Ostatníková, and P. Celec. 2021. 'The effect of long-term hypogonadism on body composition and morphometry of aged male Wistar rats', *Physiol Res*, 70: S357-s67.
de Chaves, G., M. Moretti, A. A. Castro, W. Dagostin, G. G. da Silva, C. R. Boeck, J. Quevedo, and E. C. Gavioli. 2009. 'Effects of long-term ovariectomy on anxiety and behavioral despair in rats', *Physiol Behav*, 97: 420-5.

De la Fuente, M., I. Baeza, N. Guayerbas, M. Puerto, C. Castillo, V. Salazar, C. Ariznavarreta, and J.A. F-Tresguerres. 2004. 'Changes with ageing in several leukocyte functions of male and female rats', *Biogerontology*, 5: 389-400.

Domonkos, E., V. Borbélyová, M. Csongová, M. Bosý, M. Kačmárová, D. Ostatníková, J. Hodosy, and P. Celec. 2017. 'Sex differences and sex hormones in anxiety-like behavior of aging rats', *Hormones and Behavior*, 93: 159-65.

Drori, D., and Y. Folman. 1976. 'Environmental effects on longevity in the male rat: Exercise, mating, castration and restricted feeding', *Experimental Gerontology*, 11: 25-32.

Durbin, P.W., M.H. Williams, N. Jeung, and J.S. Arnold. 1966. 'Development of Spontaneous Mammary Tumors over the Life-Span of the Female Charles River (Sprague-Dawley) Rat: The Influence of Ovariectomy, Thyroidectomy, and Adrenalectomy-Ovariectomy', *CANCER RESEARCH*, 26: 400-11.

Fares E, Pyle WG, Ray G, Rose RA, Denovan-Wright EM, Chen RP, and Howlett SE. 2013. 'The impact of ovariectomy on calcium homeostasis and myofilament calcium sensitivity in the aging mouse heart', *PLoS One*, 8: e74719.

Galea, L.A.M., M.M. Roes, C.J. Dimech, C. Chow, R. Mahmoud, S.E. Lieblich, and P. Duarte-Guterman. 2018. 'Premarin has opposing effects on spatial learning, neural activation, and serum cytokine levels in middle-aged female rats depending on reproductive history', *Neurobiology of Aging*, 70: 291-307.

Garratt, M., B. Bower, G. G. Garcia, and R. A. Miller. 2017. 'Sex differences in lifespan extension with acarbose and 17-alpha estradiol: gonadal hormones underlie male-specific improvements in glucose tolerance and mTORC2 signaling', *Aging Cell*, 16: 1256-66.

Garratt, M., D. Leander, K. Pifer, B. Bower, J. J. Herrera, S. M. Day, O. Fiehn, S. V. Brooks, and R. A. Miller. 2019. '17-alpha estradiol ameliorates age-associated sarcopenia and improves late-life physical function in male mice but not in females or castrated males', *Aging Cell*, 18: e12920.

Garratt, M., H. Try, and R.C. Brooks. 2021. 'Access to females and early life castration individually extend maximal but not median lifespan in male mice', *GeroScience*, 43: 1437-46.

Heikkinen, T., J. Puoliväli, and H. Tanila. 2004. 'Effects of long-term ovariectomy and estrogen treatment on maze learning in aged mice', *Experimental Gerontology*, 39: 1277-83.

Heinze-Milne, S.D., S. Banga, J. Godin, and S.E. Howlett. 2022. 'Serum testosterone concentrations are not associated with frailty in naturally ageing and testosterone-deficient older C57Bl/6 mice', *Mechanisms of Ageing and Development*, 203: 111638.

Herrera, J.J., S Louzon, K .Pifer, D. Leander, G. E. Merrihew, J. H. Park, K. Szczesniak, J. A. Whitson, J. E. Wilkinson, O. Fiehn, M. J. MacCoss, S. M. Day, R. A. Miller, and M. Garratt. 2020. 'Acarbose has sex-dependent and independent effects on age-related physical function, cardiac health and lipid biology', *JCI Insight*, 5(21): e137474.

Hewitson TD, Zhao C, Wigg B, Lee SW, Simpson ER, Boon WC, and Samuel CS. 2012. 'Relaxin and castration in male mice protect from, but testosterone exacerbates, age-related cardiac and renal fibrosis, whereas estrogens are an independent determinant of organ size', *Endocrinology*, 153: 188-99.

Holland, J. M., T. J. Mitchell, and Walburg, H. E., Jr. 1977. 'Effects of Prepubertal Ovariectomy on Survival and Specific Diseases in Female RFM Mice Given 300 R of X Rays', *Radiation research*, 69: 317-27.

Hotchkiss, C.E. 1995. 'Effect of surgical removal of subcutaneous tumors on survival of rats', *Journal of the American Veterinary Medical Association*, 206: 1575-79.

Hou, Y., X. Q. Bao, H. L. Wei, Y. Luo, and G. T. Liu. 2011. 'Long-term deprivation of gonadal hormone accelerates brain aging in mice', *Neurological research*, 33: 43-9.

Itoh, N., Y. Itoh, C. E. Meyer, T. T. Suen, D. Cortez-Delgado, M. Rivera Lomeli, S. Wendin, S. S. Somepalli, L. C. Golden, A. MacKenzie-Graham, and R. R. Voskuhl. 2023. 'Estrogen receptor beta in astrocytes modulates cognitive function in mid-age female mice', *Nat Commun*, 14: 6044.

Jianjun J, Na Ge, Yuzhi Wang, Juntao Qi, Guibiao Wen, Xiufen Gu, Xuewen Yu, Muming Shao, Yueming Luo, Kangshuyun Gu, Feng Lin, Shudong Yang, and Wei Wei. 2024. 'Applicability of Castration Model in Sex Difference Studies: Insights from Metabolome and Transcriptome Analyses', *bioRxiv*: 2023.12.27.573488.

Koebele, S.V., V.E. Bernaud, S.N. Northup-Smith, M.N. Willeman, I.M. Strouse, H.L. Bulen, A.R. Schrier, J.M. Newbern, D.F. DeNardo, L.P. Mayer, C.A. Dyer, and H.A. Bimonte-Nelson. 2023. 'Gynecological surgery in adulthood imparts cognitive and brain changes in rats: A focus on hysterectomy at short-, moderate-, and long-term intervals after surgery', *Hormones and Behavior*, 155: 105411.

Lazenby CM, Westwood FR, and Greaves P. 1993. 'Crescentic cataracts in Alderley Park rats', *Veterinary pathology*, 30: 70-4.

Leffa, DD, AP Damiani, DD Damazio, NP Guerra, M Moretti, GGD de Brito, CR Boeck, EC Gavioli, and VM de Andrade. 2014. 'Long-term effects of ageing and ovariectomy on aversive and recognition memory and DNA damage in the hippocampus of female rats', *ACTA NEUROPSYCHIATRICA*, 26: 161-69.

Mason JB, Cargill SL, Griffey SM, Reader JR, Anderson GB, and Carey JR. 2011. 'Transplantation of young ovaries restored cardioprotective influence in postreproductive-aged mice', *Aging Cell*, 10: 448-56.

Moore, E. 2022. 'The Impact Of Long Term Ovariectomy On Diastolic Function And Fibrosis Following Chronic Sympathetic Stimulation In Mice', PhD Thesis. East Tennessee State University.

Morin-Grandmont, A., E. Walsh-Wilkinson, SÈ Thibodeau, D. K. Boudreau, M. Arsenault, Y. Bossé, and J. Couet. 2024. 'A murine model of hypertensive heart disease in older women', *PeerJ*, 12: e17434.

Perez, J., E. Burunat, R.M. Arevalo, and M. Rodriguez. 1989. 'Gonadal influences on behavioral deterioration with aging of male rats', *Hormones and Behavior*, 23: 457-65.

Phelan, J.P. 1995. 'Reproductive costs and longevity in the house mouse', PhD thesis. Harvard University.

Picazo, O., J. Espinosa-Raya, A. Briones-Aranda, and M. Cerbón. 2016. 'Ovariectomy increases the age-induced hyperphosphorylation of Tau at hippocampal CA1', *Cognitive Processing*, 17: 443-49.

Pijacka, W., B. Clifford, C. Tilburgs, J.A. Joles, S. Langley-Evans, and S. McMullen. 2015. 'Protective role of female gender in programmed accelerated renal aging in the rat', *Physiological Reports*, 3: e12342.

Pilgrim, HI. 1957. 'A method of evaluating tumor morbidity as applied to the effect of ovariectomy at different ages on the development of mammary tumors inc-3(h) mice', *CANCER RESEARCH*, 17: 405-08.

Planas-Silva, M.D., T.M. Rutherford, and M.C. Stone. 2008. 'Prevention of age-related spontaneous mammary tumors in outbred rats by late ovariectomy', *Cancer Detection and Prevention*, 32: 65-71.

Pullinger, B. 1961. 'Increase in mammary carcinoma and adenoma and incidences of other tumours in c3hf virgin females after ovariectomy and high dosage with some oestrogens', *British Journal of Cancer* 15: 574-+.

Pullinger, B. D., and M. A. Head. 1964. 'Hepatoma in intact c3hf male and virgin female mice and after gonadectomy alone or sebsequent treatment with oestrogen', *Britich Journal of Cancer*, 13: 521-7.

Reckelhoff, J. F., and C. Baylis. 1993. 'Glomerular metalloprotease activity in the aging rat kidney: inverse correlation with injury', *J Am Soc Nephrol*, 3: 1835-8.

Storer, J. B., T. J. Mitchell, and R. L. Ullrich. 1982. 'Causes of death and their contribution to radiation-induced life shortening in intact and ovariectomized mice', *Radiation research*, 89: 618-43.

Urbieta-Caceres, V. H., F. A. Syed, J. Lin, X. Y. Zhu, K. L. Jordan, C. C. Bell, M. D. Bentley, A. Lerman, S. Khosla, and L. O. Lerman. 2012. 'Age-dependent renal cortical microvascular loss in female mice', *Am J Physiol Endocrinol Metab*, 302: E979-86.

Walsh-Wilkinson, É, M. L. Aidara, A. Morin-Grandmont, SÈ Thibodeau, J. Gagnon, M. Genest, M. Arsenault, and J. Couet. 2022. 'Age and sex hormones modulate left ventricle regional response to angiotensin II in male and female mice', *Am J Physiol Heart Circ Physiol*, 323: H643-H58.

Wang, Z., W. Wu, M.S. Kim, and D. Cai. 2021. 'GnRH pulse frequency and irregularity play a role in male aging', *Nature Aging*, 1: 904-18.

Zakeri, M., I. Fatemi, A. Kaeidi, M. A. Zakeri, E. Hakimizadeh, M. Hassanipour, M. Rahmani, J. Hassanshahi, F. Ayoobi, and M. Allahtavakoli. 2019. 'Pro-neurocognitive and anti-sarcopenic benefits of one-year metformin therapy in ovariectomized aged mice', *Clin Exp Pharmacol Physiol*, 46: 1133-40.