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Uncoupling elephant *TP53* and cancer

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Elephant testicles do not descend, with implications for sperm production being hot enough to compromise germline DNA replication/repair. Uniquely, elephants also possess 20 copies of a gene encoding for the p53 protein. Did elephants evolve multiplication of the *TP53* gene complex to protect their germline rather than to fight cancer?

TP53, an intriguing complex of genes

Each and every division of a cell carries the risk of a mutation, and larger animals have more somatic cell divisions than smaller ones. Consequently, very large animals ought to be more likely to develop cancer. But they do not, if anything the opposite, which is known as Peto's paradox [1]. Elephants (Elephantidae) have become a model organism to explore the implications of this paradox because elephants are not only very large but also, uniquely, carry more than one copy of the *TP53* gene, which encodes for the p53 protein. Named 'Guardian of the Genome' by its co-discoverer David Lane, p53 'protects' dividing cells of soma and germline by suppressing the spread of genetic mutations in an organism by initiating a cascade of DNA repair or, if repair is impossible, by instigating apoptosis to eliminate the dividing cells [2,3].

Not surprisingly, given its ability to guard against cancerous mutations in somatic cell divisions, most research into this protein and its complex molecular interactions focuses on p53's anticancer properties. Indeed, exploring Peto's cancer paradox led to the surprise discovery that elephants have 20 copies of the *TP53* gene compared with a single copy in all other animals examined [4,5]. If nothing else, this feature already marks elephants out as an important natural experiment for anyone interested in the *TP53* gene complex and the molecular biology of p53 proteins. But elephants offer more than multiple *TP53* genes (and retrogenes) encoding (or not) various p53 protein isoforms [6]. In addition to their impressive size, elephants provide another rare, interesting, and potentially important character trait with potentially significant implications for germline stability: nondescending testicles [7].

The hypothesis presented here explores the possibility that the multiplication of *TP53* genes in elephants was not due to selection against cancer in the soma but instead was the result of selection for protection of the germline. After all, p53 is an active key participant in cell divisions in both soma and germline [3]. In this scenario, any benefits of cancer protection would have been a welcome serendipity, perhaps leading to its own evolutionary developments. This view is based on the premise that, because of turnover, selection pressures are stronger on germline mutations than on somatic mutations. Death of individuals from cancer, especially at an advanced age and often post-reproductive age, provides evolution with much lower selection 'pressures' than success of a sperm and its payload in the face of competition and morphogenesis. Moreover, while natural selection acts slowly on whole animals, it can act extremely fast on spermatozoa.

Linking nondescending testicles

Elephants are not only very large and have 20 copies of *TP53*, but they have a trait highly relevant concerning germline stability. The elephant bull's testes do not descend into a scrotum but remain deep inside the body. Whether solely/exclusively the result of their afrothere ancestry [7], perhaps combined with unknown past selection pressures, the fact remains that the elephant's testicles are testicond and ascrotal. In consequence, testes and spermatogonia would share the animal's body temperature (Figure 1). This likely has serious implications for sperm production. Mammalian spermatogenesis is greatly affected by temperature, with only marginal heating of the spermatogonia sperm leading to rapid decreases in sperm quality [8]. It seems that in all mammals the high turnover of proliferating cells requires metabolic temperatures 2–4°C below body temperature. For example, a mouse's core body temperature is 36.6°C, while the testicles are at 34°C; any sperm produced at 37°C has low viability, with high levels of unrepaired DNA and compromised crossover formation, while sperm produced at 38°C will be killed by apoptosis because of excessive loads of mutated DNA [9].

The strong link between temperature and the production of healthy sperm in mammals suggests why maturing testes descend into a scrotum that is cooled to temperatures typically significantly below core body temperature by a combination of airflow and venal–arterial blood heat exchange. Elephants cannot benefit from such scrotal cooling and their testes consequently would shadow core body temperatures, which circle around 36–37°C [10]. Physical activity in the heat can push body temperatures up significantly. In the African savannah an elephant will experience long, sunny days that heat the skin while the body is heated internally by metabolism. In a multi-ton animal, muscle action generates heat when walking slowly on the level and more so when the animal runs or walks uphill [10]. For an elephant, the ear is the most important heat exchanger, with the pinna being well supplied with counterflow arteries and veins; temperatures of 39°C were measured by thermal camera at the access point of this radiator, the ear hole auricle in a

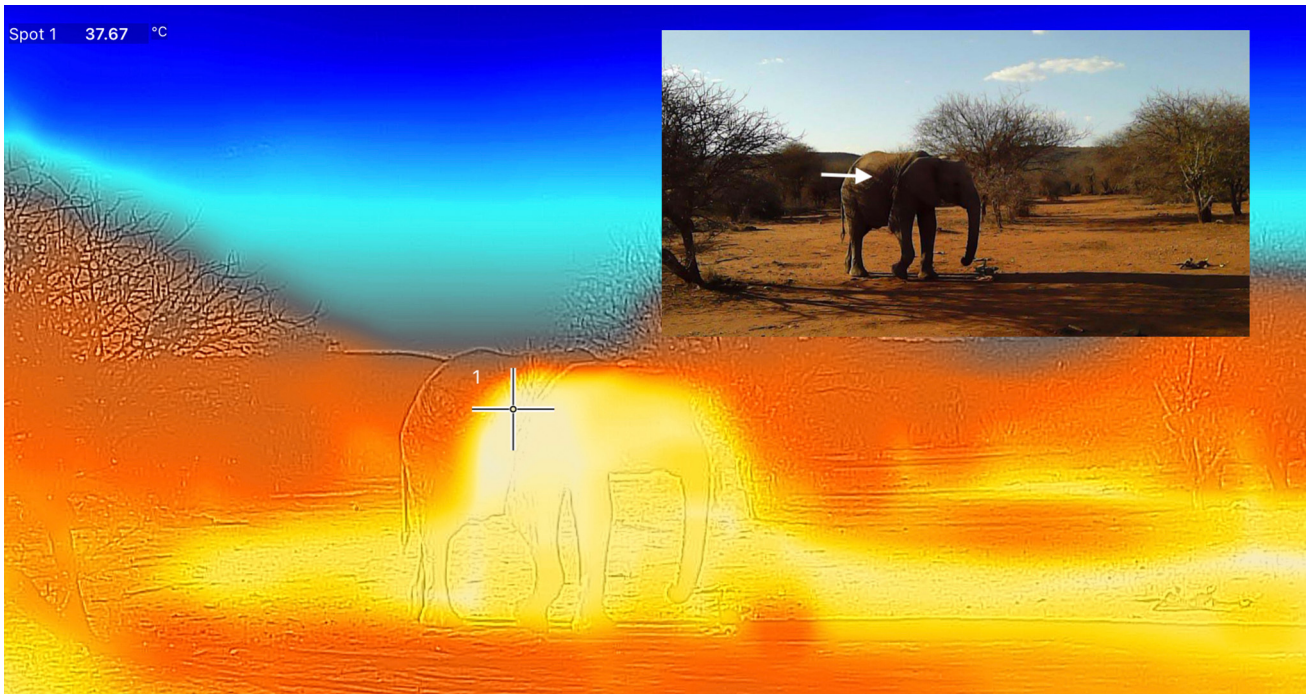

Figure 1. A young elephant shown in a FLIR camera false colour temperature image. The estimated location of the testicles is shown by the arrow in the inset, which shows that the image was taken late afternoon and out of the full sun. The spot measurement on the body above the position of the testes is shown to be 37.6°C at the time of observation, when the air temperature was 28.6°C measured by calibration thermometer. (See www.flir.co.uk for details of FLIR thermal analysis software.)

savannah elephant [10]. A thermocouple implanted in the pinna main artery of a free-ranging, 4-ton bull African savannah elephant recorded around 40°C for blood flowing during a bout of activity, shooting up briefly to a high of 44°C when the apparently distressed animal ran up a slope (I. Douglas-Hamilton, DPhil thesis, University of Oxford, 1972). Blood temperatures were around 36°C when the animal was resting or drinking, which is an elephant's way of cooling down.

Germline versus soma selection

All things considered, it appears that the elephant's testes may experience temperatures dangerously high for mammalian sperm production, even under normal body temperatures. High temperature metabolism tends to be coupled with cellular oxidative stress, which increases the probability of mutations. Such mutations could be gene duplications, including multiplications of the *TP53* gene. On chromosomes with damaged DNA, p53 helps stabilise telomeres and thus secures correct crossover [11], critical for healthy cell divisions [12]. Considering its varied actions during cell divisions, in elephant evolution the p53 protein could have played several key roles in securing stable spermatogenesis [8] where millions of cell divisions happen in rapid succession during mating periods (and hot contests) in order to provide the millions of spermatozoa inhabiting each ejaculate.

The importance of keeping the testicles at functional temperature is shown by the whale, another outsized and long-lived mammal. Cetaceans also have in-body testicles, although in this case presumably not as an ancestral genetic constraint but as an adaptation to life in cold water. Here testicular temperature is controlled by the rete mirabile, an abdominal network of heat-exchanging arteries and veins comparable with (and perhaps evolved from) scrotal cooling. Note that whales only have a single set of *TP53* genes but with molecular pathways apparently directly linked to cancer [13].

The suggested hypothesis outlines a testable opinion (Box 1). It proposes that elephants initially evolved multiple copies of *TP53* not to fight cancer but to protect sperm production in testicles that were increasingly temperature challenged as the animals grew in size during their evolutionary history. Sperm selection is a powerful driver in evolution and a bull's reproductive success will depend critically on the quality of his spermatozoa, first by one sperm being successful in beating the competition to fertilise an egg and after that by the quality of the genes handed down to the zygote and developing offspring. Indeed, heat stress in spermatogenesis

Box 1. The hot testicle hypothesis

The hot testicle hypothesis outlines a testable scenario that is not in conflict with research into p53 as an anticancer agent but suggests additional avenues of research. The hypothesis focuses on elephants and links temperature, germline, *TP53* retrogenes, and p53 protein isoforms. Tests would start with addressing the temperature range of elephant testicles and their links to body core temperatures. This topic has serious implications if climate change increases temperatures in the elephant's natural range, with the potential of endangering whole populations. Next, one might examine how other taxa, related or not, with ascrotal testicles, compare concerning testes temperature and p53 isoforms. Finally, for cancer researchers, probably the most important question would focus on the mechanism by which the elephant's various *TP53* retrogenes and p53 protein isoforms are able to guard, or not, the soma. Such studies would probably rely on genetic profiling of tissue samples and cell lines. Elephants are not as easily studied as mice. But as a natural 'experiment' evolved over millennia, they provide us with a unique research opportunity to drill deep into the genetic and molecular fundamentals of 'Guardian of the Genome' genes and proteins.

may well be a great opportunity for jumping genes to foster adaptations [14]. Yet elephants are also rather good (it seems) at either avoiding or fighting cancers and it remains to be seen whether and how this ability is linked to the presence of many and diverse p53 isoform proteins encoded in their 20 copies of *TP53* genes.

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Declaration of interests

There are no conflicts of interest.

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References

1. Caulin, A.F. and Maley, C.C. (2011) Peto's paradox: evolution's prescription for cancer prevention. *Trends Ecol. Evol.* 26, 175–182
2. Lane, D. and Levine, A. (2010) p53 research: the past thirty years and the next thirty years. *Cold Spring Harb. Perspect. Biol.* 2, a000893
3. Kastenhuber, E.R. and Lowe, S.W. (2017) Putting p53 in context. *Cell* 170, 1062–1078
4. Sulak, M. *et al.* (2016) TP53 copy number expansion is associated with the evolution of increased body size and an enhanced DNA damage response in elephants. *eLife* 5, e11994
5. Abegglen, L.M. *et al.* (2015) Potential mechanisms for cancer resistance in elephants and comparative cellular response to DNA damage in humans. *JAMA* 314, 1850–1860
6. Padariya *et al.* (2022) The elephant evolved p53 isoforms that escape MDM2-mediated repression and cancer. *Mol. Biol. Evol.* 39, msac149
7. Sharma, V. *et al.* (2018) Loss of *RXFP2* and *INSL3* genes in Afrotheria shows that testicular descent is the ancestral condition in placental mammals. *PLoS Biol.* 16, e2005293
8. Robinson, B.R. *et al.* (2023) Testicular heat stress, a historical perspective and two postulates for why male germ cells are heat sensitive. *Biol. Rev.* 98, 603–622
9. Hirano, K. *et al.* (2022) Temperature sensitivity of DNA double-strand break repair underpins heat-induced meiotic failure in mouse spermatogenesis. *Commun. Biol.* 5, 504
10. Domínguez-Olivaa, A. *et al.* (2022) Anatomical, physiological, and behavioral mechanisms of thermoregulation in elephants. *J. Anim. Behav. Biometeorol.* 10, 2233
11. Tutton, S. and Lieberman, P.M. (2016) A role for p53 in telomere protection. *Mol. Cell Oncol.* 4, e1143078
12. Marcet-Ortega, M. *et al.* (2022) p53 controls meiotic prophase progression and crossover formation. *Int. J. Mol. Sci.* 23, 9818
13. Tollis, M. *et al.* (2019) Return to the sea, get huge, beat cancer: an analysis of cetacean genomes including an assembly for the humpback whale (*Megaptera novaeangliae*). *Mol. Biol. Evol.* 36, 1746–1763
14. Bhalla, N. (2020) Meiosis: is spermatogenesis stress an opportunity for evolutionary innovation? *Curr. Biol.* 30, 1471–1473