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INTRODUCTORY LECTURE: SOME INSIGHTS ABOUT AGEING FROM A SHARK AND AN OYSTER

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The subject of the 32nd Old Herborn Seminar is on the impact of ageing on the microbiome. In this brief introductory lecture I would like to explore the subject of ageing. What underlies the ageing process? In general, ageing is a process that occurs as the regenerative potential of the organism diminishes. As a consequence, the functions of vital organs gradually fail. Why does this happen? Is there an ageing "clock" in the human body? Are we composed of cells that are genetically programmed to count the hours until their senescence is reached? Or is the rate of ageing controlled by a "master clock"? Can we slow down the ageing clock?

Perhaps if we examine some long-lived animals we might discern some hints about the ageing mechanism. One of the oldest known animals is a clam collected off the coast of Iceland (*Scourse* et al., 2006). Based on the "rings" of the shell of this animal, it was estimated to be about 500 years old, and called the "Ming Clam" to highlight that it began its life during the Chinese Ming dynasty. Other clams caught in that area were similar in age, suggesting that the Ming was not an unusual outlier. Clearly these individuals survived predation. As extraordinary they survived infection and malignancy, despite (or because of?) the absence of an adaptive immune system. This animal teaches us that a robust adaptive immune system is not necessarily required for longevity.

The Greenland shark was recently discovered to be the oldest living vertebrate, with the maximum age estimated to be 600 yrs. (*Nielsen* et al., 2016). The ages of the animals were determined by radiocarbon dating of the lens of the eye, which is formed during embryogenesis. Prior studies had shown that the Greenland shark reached sexual maturity at about 200 years. The ageing clock of this animal appears to run about 10-fold more slowly than the human's. This animal teaches us that tissue renewal and organ regeneration, including reproductive capacity, can be sustained in a vertebrate for hundreds of years.

In 1961 Hayflick and Moorehead proposed that an animal's longevity had its basis at the cellular level (*Hayflick* and *Moorehead*, 1961). They demonstrated that the fibroblasts from short lived animals would undergo fewer divisions in cell culture before reaching senescence compared with the cells of longer-lived animals. It was later suggested that the number of cellular divisions a cell was capable of sustaining was dependent upon the integrity of the chromosomal telomeres, and the expression of telomerase. With each successive division, the telomeres of the stem cells shortened, until chromosomal replication was compromised.

Subsequent studies failed to validate the relationship between the longevity of a species and the number of divisions to senescence. A rather telling study asked whether the fibroblasts

grown from a biopsy taken from a 90+ year old human would undergo senescence after fewer divisions *in vitro* than those from younger individuals (*Maier* et al., 2007). The results demonstrated that the fibroblasts from the nonagenarians behaved very much like young cells with respect to the maximal number of replications. These observations suggest that the replicative capacity of a stem cell is influenced by factors contributed by the *in vivo* setting. What anti-senescence factors are present *in vivo*, but absent *in vitro*? Might a gradual decrease in "anti-senescence "hormones" be responsible for ageing?

Caloric restriction has been shown to increase longevity in flies, worms, fish and mice (*Balasubramanian* et al., 2017), so better understanding of the physiological consequences of caloric restriction could provide some insight into the processes influencing longevity: IGF-1, insulin, and growth hormone concentrations fall in serum (*Bartke* and *Westbrook*, 2012; *Cady* and *Sadagurski*, 2017); plasma ketone bodies increase, providing the brain with a highly efficient source of energy, reducing its dependence on glucose; and most curious, reduced inflammation within the hypothalamus (*Cady* and *Sadagurski*, 2017).

A recent study suggests that progressive hypothalamic inflammation might be a key factor in systemic ageing (*Zhang* et al., 2013). In this study, a lentivirus expressing GFP driven by an NF-κB response element was injected to either the hypothalamus or the cortex of mice. As the animals aged, the intensity of GFP+ cells increased in the hypothalamus, but not in the cortex, demonstrating that the hypothalamus experiences an inflammatory milieu in contrast to the cortex. The hypothalamus has a central role in homeostatic regulation of metabolism, vital functions (temperature, blood pressure, etc), and growth, so inflammation could impact negatively on function. Indeed, in this study numerous hypothalamic peptides were examined to determine which, if any, changed in the older animals, and GnRH, which decreased, was identified as a candidate. Surprisingly, treatment of the older animals with parenteral GnRH stimulated neurogenesis in the brain. Most surprising GnRH treatment increased muscle strength, skin thickness, and cognitive functions, in a sense, restoring a more youthful phenotype.

So what controls ageing? Does inflammation within the hypothalamus drive generalized ageing of our bodies? Is it all that simple?

LITERATURE

Balasubramanian, P., Howell, P.R., and Anderson, R.M.: Aging and Caloric Restriction Research: A Biological Perspective With Translational Potential. EBioMedicine 21: 37-44 (2017)

Bartke, A. and Westbrook, R.: Metabolic characteristics of long-lived mice. Front. Genet. 3, 288 (2012).

Cady, G. and Sadagurski, M.: Targeting neuroinflammation - a potential for anti-aging interventions. Aging (Albany NY) 9, 1951-1952 (2017).

Hayflick, L. and Moorhead, P.S.: The serial cultivation of human diploid cell strains. Exp. Cell Res. 25, 585-621 (1961).

Maier, A.B., le Cessie, S., de Koning-Treurniet, C., Blom, J., Westendorp, R.G., and van Heemst, D.: Persistence of high-replicative capacity in cultured fibroblasts from nonagenarians. Aging Cell 6, 27-33 (2007).

Nielsen, J, Hedeholm, R.B., Heinemeier, J., Bushnell, P.G., Christiansen, J.S., Olsen, J.,

Ramsey, C.B., Brill, R.W., Simon, M., Steffensen, K.F., and Steeffensen, J.E: Eye lens radiocarbon reveals centuries of longevity in the Greenland shark (Somniosus microcephalus). Science 353, 702-704 (2016).

Scourse, J., Richardson, C, Forsythe, G., Harris, I., Heinemeier, J., Fraser, N., Briffa, K., and Jones, P.: First cross-matched floating chronology from the marine fossil record: Data from growth lines of the long-lived bivalve mollusc Arctica islandica. The Holocene 16, 967-974 (2006).

Zhang, G., Li, J., Purkayastha, S., Tang, Y., Zhang, H., Yin, Y., Li, B., Liu, G., and Cai, D.: Hypothalamic programming of systemic ageing involving IKK-β, NF-κB and GnRH. Nature 497, 211-216 (2013).