Slide description for AGEING INTERVENTIONS
(by: Suresh Rattan, University of Aarhus, Denmark)

Slide #1

Title of the presentation Ageing Interventions

Slide #2

A brief introduction of the lecturer/teacher, Dr. Suresh Rattan, giving some information about his educational background, place and country of origin, and academic and professional achievements.

Slide #3

A brief outline of the presentation highlighting the five main points that will be covered in this section. It may be useful to recapitulate some basic principles of the phenomenon of ageing based on which ageing interventions can be discussed.

Slide #4

Examples of the occurrence of ageing at different levels of biological organizations – from body to organs, tissues, cells, intracellular components, and macromolecules. The examples show pictures of young and old trabecular bone; a healthy young and an old Alzheimer diseased human brain; young and old arteries, young and senescent human skin fibroblasts, pattern of young and old telomeres on human chromosomes, and a comparison of DNA damage as measured by the Comet assay in human sperms collected from young and old donors.

Slide #5

The first major principle to emerge from the descriptive studies as described in the Slide #3 is that ageing occurs at all levels of biological organization, but differently. No two individuals, organs, tissues, cell types, and intracellular components including macromolecules age in exactly the same way. This is an extremely important observation that has serious implications with respect to developing potential anti-ageing interventions.

Slide #6

This is a reminder of the general mechanistic principle of ageing as an epigenetic phenomenon, and that it is not a tightly genetically regulated or programmed phenomenon. This slide basically refers to the discussion about the evolution of ageing and longevity, and about the nature of genes involved in the ageing process.

Slide #7

Here the concept of homeodynamics (or the earlier concept of homeostasis) is presented giving examples of various molecular and higher order maintenance and repair systems which are crucial for survival of a biological system. In a normal healthy young individual, there is a large homeodynamic space (a kind of generalised buffering capacity) which allows it to respond, adapt and tolerate stress and other disturbances. Of course, in spite of this large homeodynamic space, there is a zone of vulnerability which can disturb the system and even a young healthy individual can become sick one or more diseases which can even be fatal.

Slide #8 to 11

Starting with the definition of ageing as the failure of maintenance and repair, this set of four slides is an animation depicting ageing as a progressive shrinkage of the homeodynamic space. The main reason for the shrinkage is the occurrence and accumulation of molecular damage which leads to a reduction in the functional and practical space to protect, to counteract and to defend. This generalized shrinkage of the homeodynamic space effectively increases the size of the vulnerability zone and allows for the emergence of clinical manifestation of age-related impairments in the form of various diseases.

Slide #12

This slide reminds that human minds have always been occupied with the ideas of discovering ways of achieving eternal youth and immortality. The 16^{th} century painting by a German painter Lucas Cranach shows the mythical fountain of youth where elderly sick women entering from one side of the pool emerge as healthy young ladies on the other side ready to be received by smart young men and taken away to once again enjoy the pleasures of life. The second picture on this slide is a photograph of the newly installed sculptor on the

Bangkok airport, which depicts the Indian myth of churning of the oceans *(Sagar Manthan)* by gods and devils, leading to the isolation of *Amrit* – the water/nectar of immortality.

Slide #13

An extremely important issue about ageing interventions is how we view ageing in the first place. If ageing is considered as a disease, then it will require strategies to treat the disease by getting rid of all symptoms of ageing, and by reverting the biological system to an age-free youthful condition. However, if ageing is considered as a process that can lead to various diseases, then ageing interventions need to be preventive and modulatory, which can slow down or arrest the process – partially or completely.

Slide #14

This slide introduces that there are two main approaches to ageing interventions: 1. piecemeal remedies; and 2. preventive medicines.

Slide #15

Piecemeal remedies mean that different parts of the ageing system can be repaired or corrected one at a time according to the severity of the problem. For example, there can be a replacement of complete organs by transplantation (such as the heart, the kidneys etc), or partial replacement (for example the facial skin, the lungs, and the liver). Since the replacement of one organ may or may not affect the rate of progression of ageing in other organs, and also in the replaced organ, this will require repeated replacements. Another kind of replacement of organs is using organs and body parts made from more durable materials such as titanium. This kind of replacement can also be either once for all or done repeatedly according to the general ageing in the rest of the body. The third piecemeal remedy is the use of stem cells. However, this also depends on the ageing of the rest of the body, and stem cells may or may not perform optimally at the site of the damaged tissue. Therefore, stem cell therapies will also be needed to be done repeatedly ion accordance with the altered systems around the target.

Slide #16

Introducing a formula for immortality and eternal youth. $E = GMC^2$ is a conceptual "formula" to make a point that in order to achieve immortality and eternal life (E), at least three factors are required to be modulated. These factors will be discussed in the following slides. It is obvious that this formula is a "play of symbols" from the famous Einsteinian equation representing relationship between energy and mass!!

Slide #17

According to the equation $E = GMC^2$, the first variable to be controlled is G, genes. This is because genes are surely the biological basis of information for life. Since the evolution has come to select genes in accordance with its requirement for survival only up to reproductively successful age, human technology is required to redesign the genes for longer survival and for anti-ageing. Some of the major genetic pathways that require significant improvement are those involved in the maintenance, repair and management of molecular damage, such as macromoleular repair genes, macromolecular turnover genes, anti-oxidant and anti-glycation genes, etc. The use of the phrase "intelligent redesigning" is a pun on creationist and design theories. Furthermore, our ability to intelligently redesign at the gene level also raises the question of whether we should take the next step on enhancing the bodies, for example by having a second heart, an extra eye on the back, or by re-acquiring wings etc.

Slide #18

This is the list of present day problems and limitations in manipulating genes, both in terms of technical limitations and gaps in our knowledge. Of about 20,000 genes in human cells, perhaps less than 100 may be sufficient with respect to ageing and longevity. Therefore, it looks like that sooner or later, all these technological issues will be resolved and we will be able to manipulate genes (G in the equation), thus making one of the three requirements fulfilled for achieving eternal youth and immortality!!

Slide #19

This represents the second component of the equation as M = milieu or the environment. This is because genes on their own have no value or function unless they are provided with

an appropriate milieu to be able to get transcribed and translated. However, what constitutes milieu is an issue that is several orders of magnitude more complex to resolve than that in the case of genes and their interactions. This slide presents a list of various levels at which milieu may be considered, and it ranges from the micro intracellular milieu to higher order components involving the domains of psychology and sociology.

Slide #20

From this slide onwards, there are examples of various attempts being made to manipulate and to modulate the internal and external environmental conditions as ageing interventions. One of the most popular, and least rigorous, is the use of cosmetics. A multibillion-dollar cosmetics industry thrives throughout the world promoting products which, legally, are not even supposed to do anything. Although there is a lot of untested and hyped products sold in the market, during the last fifteen years or so, several potentially powerful modulators have become available. That is why, the term cosmeceutical tends to be used more frequently for such formulations in which certain active ingredients may have near-pharmaceutical effects and mode of action. In almost all these cases, the effects most commonly documented are for immediate effects on photo-damaged skin, with little or no information with respect to their long term preventive or other effects. Some muscle-paralysing toxins, such as botox and its derivatives, are also being used for their immediate smoothening effects on photo-damaged skin.

Slide #21

Modulation of the milieu from within is one of the most popular anti-ageing strategies. Generally, these approaches make use of the biogerontological research done on model systems and on humans which has shown a plethora of age-related alterations in various biochemicals, metabolites, gene products, enzymes and hormones. The basic logic behind this approach is that if there is something whose levels decrease with age (such as various hormones and enzymes), then providing the same from the outside in whatever form will compensate that loss, and will have anti-ageing effects. This slide lists various categories of natural and synthetic compounds used as nutritional supplements, functional foods, and pseudo-medicines.

Slide #22

This slide draws attention to the several important points with respect to the problems and limitations regarding the use of numerous anti-ageing products discussed in the previous slide. Although there may be some temporary beneficial effects of some of these products in the case of severe deficiencies and other clinical problems, their abilities to modulate the rate and progress of ageing remain to be shown.

Slide #23

This slide gives a list of various categories of stimulators of the intracellular environmental components which may be the potential targets for anti-ageing interventions. Some of these stimulators may be able to maintain the youthful milieu of the cells, and others may be able to improve the milieu than the original conditions.

Slide #24

Another kind of ageing intervention requires the development of inhibitors of various damaging pathways. The major categories in this case are the inhibitors of inflammation, of molecular cross links, and of unspecific degradatory pathways.

Slide #25

Unlike previous approaches of ageing intervention in which one target or one pathways is dealt with one-at-a-time, a novel approach for ageing intervention is to deal with the overall homeodynamic ability of biological systems with a view to strengthen it from within. The basis for this approach is the fact that repetitive mild stress or challenge is the fundamental impetus behind homeodynamics or homeostasis. This phenomenon is known as “hormesis”, in which those physical, chemical and biological which are harmful, toxic or even lethal at certain levels, have physiology-strengthening effects at low levels. The paradigm for hormesis is physical exercise, which in spite of its well known effects in producing damaging free radicals, acids, and other cellular damage, is well known to have health promoting beneficial effects if it is performed at a moderate level.

Slide #26

Here the three types of dose response curves are shown to highlight the differences between the frequently used "linear threshold curve" and "linear no-threshold curve" in pharmacology, toxicology and radiation biology, and the U-shaped or J-shaped hormetic curves which show that at low levels, the effects of potentially toxic and harmful conditions can be reverse and useful.

Slides #27 to 31

In this series of slides, examples are given from experimental studies performed since 1998 in the labs of the teacher of this section (Suresh Rattan) on determining the hormetic effects of repeated mild heat shock on human skin fibroblasts and keratinocytes. These effects include anti-ageing, enhancement of stress tolerance, activation of proteasome, stimulation of wound healing and enhancement of angiogenesis by endothelial cells. Complete data for these studies can be seen in the following research papers published by our group:

Berge, U., Kristensen, P. and Rattan, S.I.S. Hormetic modulation of differentiation of normal human epidermal keratinocytes undergoing replicative senescence in vitro. *Experimental Gerontology, 2008 in press.*

Rattan, S.I.S., Sejersen, H, Fernandes, R.A. and Luo, W. Stress mediated hormetic modulation of wound healing and angiogenesis in human cells *Annals of the New York Academy of Sciences, 1119, 112-121, 2007.*

Rattan, S.I.S. and Ali, R.E. Hormetic prevention of molecular damage during cellular aging of human skin fibroblasts and keratinocytes. *Annals of the New York Academy of Sciences, 1100: 424-430, 2007.*

Nielsen, E.R., Eskildsen-Helmond, Y.E.G. and rattan, S.I.S. MAP kinases and heat shock-induced hormesis in human fibroblasts during serial passaging in vitro. *Annals of the New York Academy of Sciences, 1067: 343-348, 2006.*

Rattan, S.I.S., Gonzalez-Dosal, R., Nielsen, E.R., Kraft, D.C., Weibel, J. and Kahns, S. Slowing down aging from within: mechanistic aspects of anti-aging hormetic effects of mild heat stress on human cells. *Acta Biochimica Polonica, 51: 481-492, 2004.*

Beedholm, R., Clark, B.F.C. and Rattan, S.I.S. Mild heat stress stimulates 20S proteasome and its 11S activator in human fibroblasts undergoing aging in vitro. *Cell Stress and Chaperones, 9, 49-57, 2004.*

Rattan, S.I.S., Eskildsen-Helmond, Y. and Beedholm, R. Molecular mechanisms of anti-aging hormetic effects of mild heat stress on human cells. *Non-Linearity in Biology, Toxicology and Medicine, 2: 1-12, 2004.*

Hercus, M., Loeschcke, V. and Rattan, S.I.S. Extension of longevity of Drosophila by repeated mild heat stress exposures. *Biogerontology, 4, 149-156, 2003.*

Fonager, J., Beedholm, R., Clark, B.F.C. and Rattan, S.I.S. Mild stress-induced stimulation of heat shock protein synthesis and improved functional ability of human fibroblasts undergoing aging in vitro. *Experimental Gerontology, 37: 1223-1228, 2002.*

Verbeke, P., Deries, M., Clark, B.F.C. and Rattan, S.I.S. Hormetic action of mild heat stress decreases the inducibility of protein oxidation and glycoxidation in human fibroblasts. *Biogerontology, 3, 117-120, 2002.*

Verbeke, P., Clark, B.F.C. and Rattan, S.I.S. Reduced levels of oxidized and glycoxidized proteins in human fibroblasts exposed to repeated mild heat shock during serial passaging in vitro. *Free Radicals in Biology and Medicine, 31, 1593-1602, 2001.*

Verbeke, P., Clark, B.F.C. and Rattan, S.I.S. Modulating cellular aging in vitro: hormetic effects of repeated mild heat stress on protein oxidation and glycation. *Experimental Gerontology, 35, 787-794, 2000.*

Rattan, S.I.S. Repeated mild heat shock delays ageing in cultured human skin fibroblasts. Biochemistry and Molecular Biology International, 45, 753-759, 1998.

Slide #32

Based on research on hormesis, a new category "hormetin" has been proposed for natural and synthetic compounds which induce stress responses in the cells, and may be hormetically beneficial at low doses. There can be further sub-division of hormetin categories, such as those which directly induce the heat shock response, or those which enhance / co-induce the heat shock response, those which induce antioxidant responses, those which induce DNA repair responses, and others yet to be identified.

Slides #33-34

An example of natural hormetin is curcumin, a component of a widely used food spice Turmeric, which is known to have anti-cancer and anti-inflammatory effects at high doses. At lower doses, curcumin is now reported to have hormetic effects by inducing stress responses, and has beneficial biological effects such as improved wound healing and enhanced proteasome activities.

Slide #35

Another example of a natural hormetin from food sources is a polyphenol rosmarinic acid isolated from the herb sage. This hormetin also induces intracellular stress response, and has anti-ageing biological effects.

Slide #36

There are several other stresses being tested in a wide variety of experimental model systems. This slide shows the front cover of the first book published on this topic, which compiles review articles discussing the effects of various stresses being tested for their effects on ageing and longevity. The complete reference for this book is:

Le Bourg, E. and Rattan, S.I.S. (editors) *Mild Stress and Healthy Aging: applying hormesis in aging research and interventions;* Springer Publishers, Dordrecht, The Netherlands, 2008.

Slide #37

This slide recapitulates the pictorial representation of ageing as a progressive shrinkage of the homeodynamic space, which can be slowed down or prevented through hormesis. This picture also shows that hormesis can slow down the rate of ageing and the rate of increase of the size of the vulnerability zone, thus preventing or delaying the onset of age-related diseases. In this way, even if the lifespan is not extended, the health-span is extended significantly.

Slide #38

This slide gives a list of age-related diseases which can be the potential targets for hormesis induced prevention through specific organs and tissues.

Slides #39-40

This slide comes back to the conceptual equation E = GMC2 and reminds that G (genes) and M (milieu) are open to interventions, but the third factor C (chance) or stochasticity in all complex interacting systems makes it virtually impossible to achieve eternal youth and immortality.

Slide #41

This final slide gives the contact address of the teacher Dr. Suresh Rattan, Department of Molecular Biology, University of Aarhus, Denmark (Email: rattan@mb.au.dk)

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