Biomarkers of Ageing

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UK SPINE Knowledge Exchange

- UK SPINE Knowledge Exchange is a collaboration between:
  - University of Dundee Drug Discovery Unit
  - Medicines Discovery Catapult
  - University of Birmingham
  - University of Oxford
  - Francis Crick Institute

Aims of the UK SPINE KE

- To bring together multi-disciplinary expertise from:
  - Higher education institutes
  - Various industries
  - Charities
  - NHS partnerships
- To improve therapeutics for older people and reduce the impact of age-related multimorbidity

Ageing and Multimorbidity

- Biological age is a measure of physiological health, as opposed to the chronological age in years
- Various factors can accelerate an individual's biological ageing, including sedentary lifestyle, illness and severe trauma
- Multimorbidity is known to increase with age and is therefore one of the key challenges facing health researchers today
- It is estimated that one seventh of the population will be aged over 75 by 2040 and multimorbidity will become an increasingly pressing issue, with implications for patient quality of life as well as health and care systems

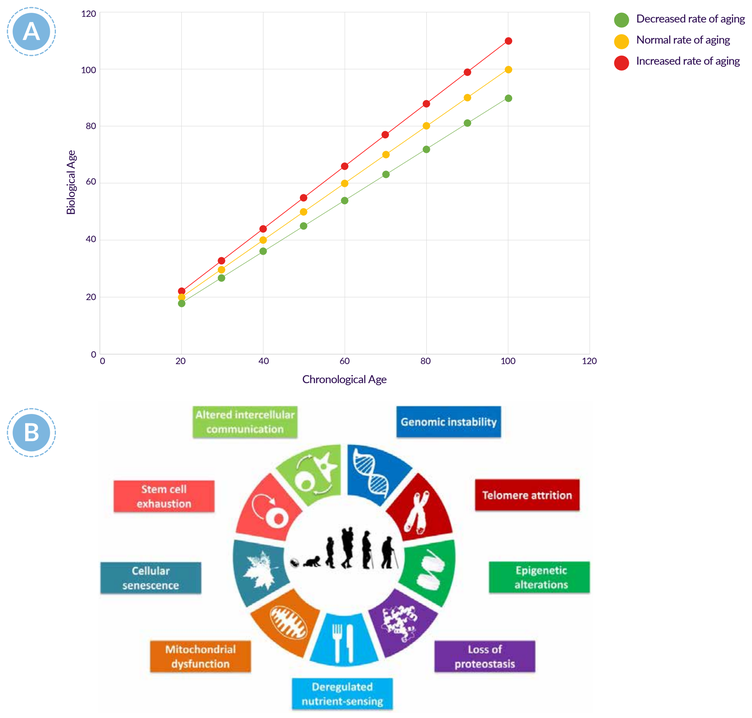


Figure 1: Chronological age vs. biological age (A) and the nine hallmarks of ageing in humans (B)[1].

Pathways Associated with Ageing

- Many pathways have been identified as associated with ageing, including PI3K and mTOR signalling, autophagy and cell senescence; however the mechanisms underlying ageing are still poorly understood
- Elucidation of these mechanisms will help us to better understand the ageing process and provide potential drug targets to prevent or treat age-associated multimorbidity, as well as other age-associated diseases such as arthritis and Alzheimer's disease

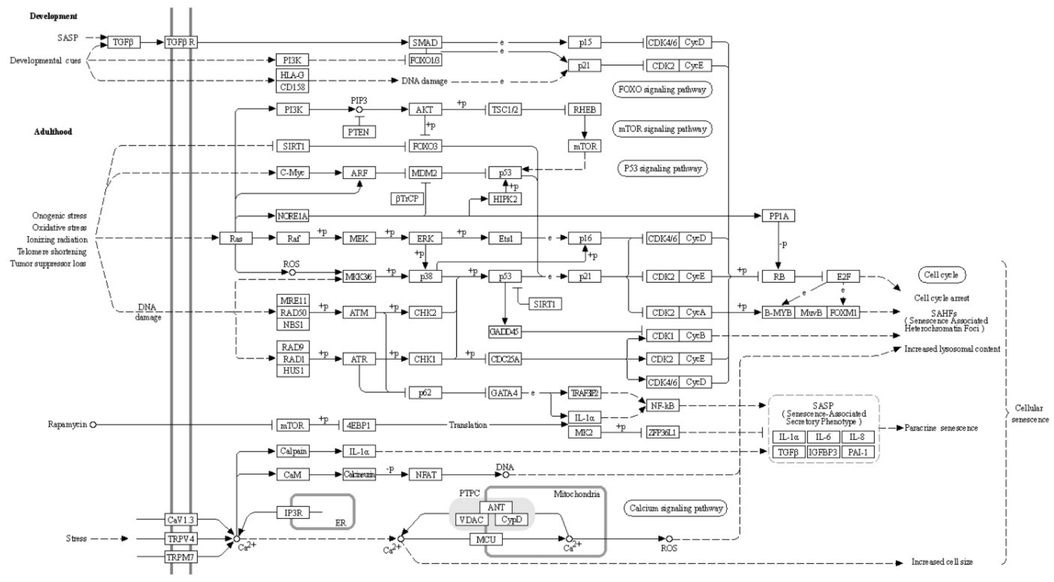


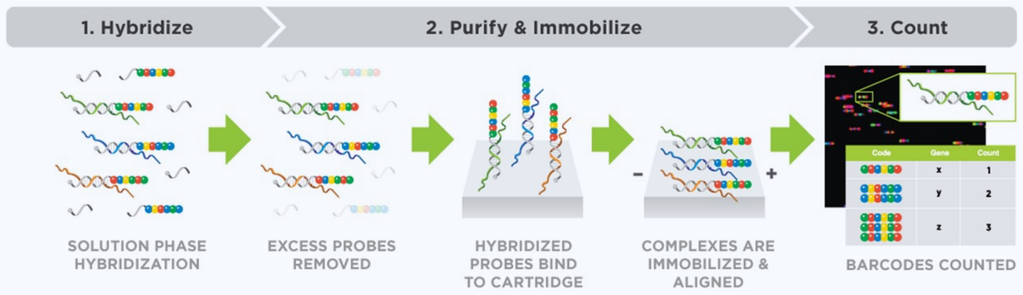
Figure 2: KEGG pathway map of the cellular senescence pathway in humans.

Project Overview

- The aim of this study was to identify an RNA biomarker signature of ageing within clinical plasma samples collected from individuals aged below 45 and above 65
- Patient medical history was used to exclude individuals suffering from chronic conditions (such as cancer, diabetes and autoimmune diseases) from the study

RNA extracted from plasma → RT-qPCR testing → Nanostring nCounter testing

- Following RNA extraction, RT-qPCR testing was performed on the eluates to determine the level of RNA degradation within the sample. These results were then used to select samples for testing on the Nanostring nCounter instrument
- The Nanostring nCounter platform is able to measure the RNA count of up to 800 genes within each sample using a CodeSet
- A custom CodeSet of 409 genes associated with ageing was designed for use in this study, as well as 20 housekeeping genes used to normalise between samples



Results

- Samples were clustered according to the age of the patient, with some pathways upregulated and others downregulated in patients over 65 years (n=37) compared with those aged under 45 years (n=38)
- Some samples collected from patients aged over 65 more closely resembled those of patients aged below 45 and vice versa; this may be due various reasons such as lifestyle choices
- Of the 409 genes within the CodeSet, 112 were seen to be differentially expressed between patients aged below 45 and over 65 ($p \leq 0.01$), however the Log_2Fold change was between -1 and 1 for many of these genes

A

Adjusted p-value	Total Genes	Log_2Fold Change	Total Genes
<0.001	75	≤-1	15
		-1<x<1	55
		≥1	5
<0.01	112	≤-1	17
		-1<x<1	90
		≥1	5
<0.05	133	≤-1	17
		-1<x<1	111
		≥1	5

B

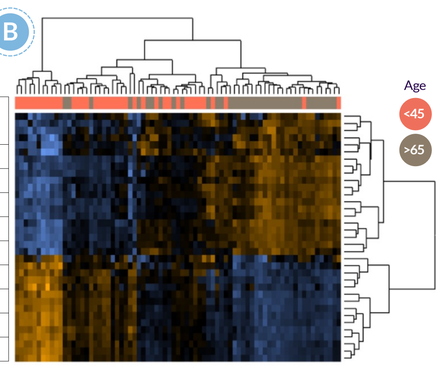


Figure 3: Differentially expressed genes (A) and heatmap of molecular pathways between patients <45 and >65 years old (B).

Next Steps

- This data will be used to generate an RNA biomarker signature of ageing, which could be used to predict an individual's biological age
- The RNA biomarker signature could be validated using a new cohort of clinical samples and measuring its effectiveness at stratifying individuals according to age
- This signature could then be tested in patients suffering with chronic diseases or severe trauma, to investigate the pathways underlying the increased rate of ageing observed in these patients
- Differentially expressed genes identified within this study will be investigated as potential therapeutic targets for prevention / treatment of age-related diseases

Acknowledgements
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References

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