

FTIR spectroscopy discriminates very early stage differentiation in living human stem cells

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We have been investigating using vibrational spectroscopy to define lineage commitment in human stem cells, and envisage spectroscopic approaches may be useful for quality control and selection of differentiated stem cells destined for use in regenerative medicine therapies. This is important because cells derived from stem cells used in the clinic will first need to be differentiated to specific differentiation or lineage commitment states for these treatments to be effective. Conversely, the transplantation of undifferentiated stem cells into the human body presents a risk as it may lead the formation of cancerous tumours. Recently, we have been successful in using FTIR spectroscopy to discriminate living, undifferentiated human embryonic stem cells from those differentiated to stages equivalent to the earliest phases of embryonic development. These measurements employed a specialised IR wet chamber and were conducted using the IR beamline at the Australian Synchrotron [1]. Apart for the obvious practical need to analyse live cells in line with the aim to develop a new modality for cell selection for clinical practice, measurements using living cells had advantages compared to those from dried, fixed cells. Significantly, in terms of detecting changes in differentiation state, bands from DNA and RNA were observed in the FTIR spectra of live cells that were not detected at all or were much less prominent in the FTIR spectra from the dried cells [2]. Indeed, changes in bands assigned to nucleic acids, including the carbonyl stretching band from DNA at $\sim 1720\text{ cm}^{-1}$, the anti-symmetric phosphodiester stretching band from DNA and RNA at $\sim 1220\text{ cm}^{-1}$, and stretching bands from C-O groups in DNA and RNA sugars at ~ 1120 and 1050 cm^{-1} , associated with the differentiation of cells from the stem cell progenitors, were observed in average spectra and loaded prominently in Partial Least Squares (PLS) models used to classify the spectra. Large changes in lipid absorbance was also observed as the live stem cells underwent differentiation, in parallel with changes previously observed in the spectra of dried cells [3]. We will discuss these findings and the potential for FTIR spectroscopy as a quality control tool for cell selection in regenerative medicine practice.

References

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