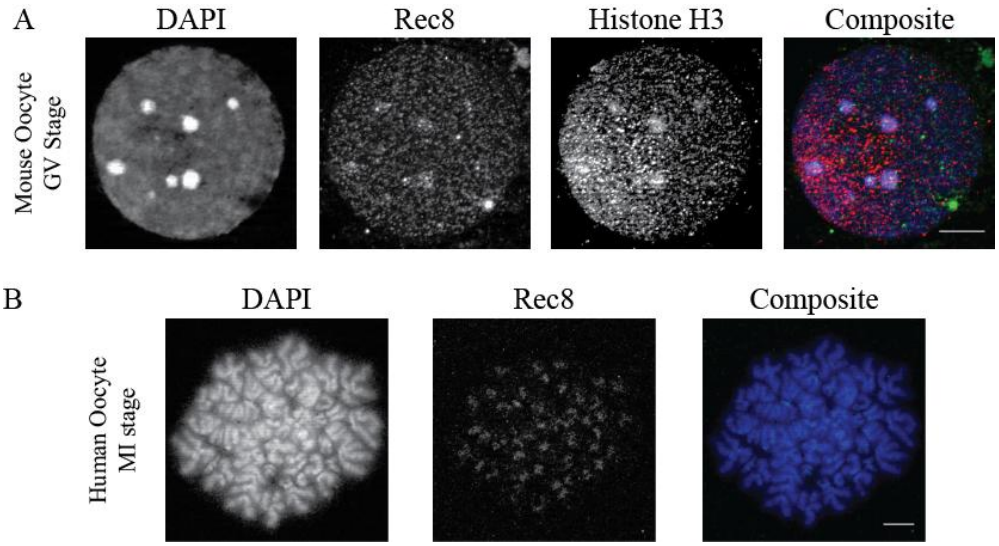
SRF Vacation Scholarship Report

The increasing trend for women in the developed world to delay pregnancy to an older age has led to an escalation in the frequency of infertility, miscarriage and birth defects. Understanding the cause of these problems could help prevent these issues from continuing to become increasingly prevalent. The underlying cause of age-related decline in female reproductive function has been shown to be the result of an increased incidence of chromosome segregation errors during meiosis I. Cohesin is a key factor in preserving chromosome architecture and ensuring adequate chromosome segregation; it is therefore central to the whole problem. Its role is to maintain cohesion between sister chromatids and is essential to stabilising bivalent chromosome structure established between maternal and paternal homologs undergoing reciprocal recombination.

Evidence suggests that the cohesin complex is dissociated from oocyte chromosomes during female ageing. This is accompanied by the destabilisation of chiasmata and the loss of tight association between sister centromeres required for stable biorientation of homologous chromosomes. The identification of cohesin loss as a cause for aneuploidy and loss of fertility is merely the first step towards improving fertility in ageing females. The next step must be to determine when and how cohesin loss occurs, and whether the depletion of cohesin is related to the depletion of the germ cell pool. Determining any marked differences in cohesin levels between young and aged females at different stages of oogenesis will allow us to ascertain any particular point where cohesin loss is accentuated in the reproductive lifespan.

Rec8 forms part of a tripartite ring of cohesin, that when cleaved causes the dissociation from DNA and therefore loss of cohesion. My project aimed to determine whether a suitable chromosomal marker can be identified to normalise Rec8 levels for quantitative immunofluorescence. For this purpose I used an antibody to Histone H3 and found that it gave a clear chromosome-associated signal (Fig 1A). Further work is required to determine whether chromosome-associated Histone H3 levels remain constant during female ageing. In preliminary experiments, I also found that Rec8 can be detected on chromosomes of human oocytes (Fig 1B). Ongoing work will determine whether this changes during female ageing.



The images are taken at 65x in zeiss Apotome II microscope with scale bar 10μm.

A) Shows the chromosome spread of Rec8-myc mouse oocyte at GV stage with optimization of Histone H3 antibody as an arm marker to normalize the measurements of Rec8.
B) Shows human oocyte at MI stage with optimization of human-antiRec8 antibody in chromosome spread.