SRF VACATION SCHOLARSHIP REPORT 2022

The form below should be completed by the student, then forwarded to the supervisor for approval and submission to srf@conferencecollective.co.uk within 8 weeks of completing the project.
Please submit the form as a PDF document and save it as: First name, Last name and 'VS'.

A maximum of one figure (with legend of less than 100 words) may be appended if required.

Please note: excerpts from this form may be published on the SRF website, so please ensure content is suitable for website publication, and does not compromise future dissemination of data in peer-reviewed papers etc. The SRF reserves the right to edit responses to ensure suitability for publication on the website, newsletter or in promotional material.

Student's Name:	Hanning (Julia) Li	**Student's Institution/University:**	University of Edinburgh
Degree Title and year of study:	Reproductive Biology (Hons) Year 3		
Supervisor's Name:	Norah Spears Heather Flanagan	**Supervisor's Department and Institution:**	University of Edinburgh
Project Title:	TGF-β1 stimulates embryo attachment in an in vitro model system		

Briefly outline the background and aims of the project *(max 200 words)*

While in normal pregnancy the embryo implants in the uterus, in tubal ectopic pregnancy (tEP) the embryo implants in the Fallopian tube. tEP can cause severe health consequences, however the mechanism behind tEP remains unclear. Epithelial to mesenchymal transition (EMT) is a cell transition process whereby epithelial cells gain mesenchymal characteristics: epithelial cells start to migrate, have increased proliferation, and lose cell adherence and polarity. The protein expression changes that occur in epithelial cells undergoing EMT and in Fallopian tube epithelial cells in tEP are similar, suggesting that EMT in Fallopian tube epithelial cells may lead to tEP. This project aimed to explore how the EMT-inducer transforming growth factor beta 1 (TGF-β1) affects embryo attachment to Fallopian tube and endometrial epithelial cells. This was investigated using a co-culture model where murine embryos were cultured with monolayers of a Fallopian tube epithelial cell line to model tEP: monolayers of an endometrial epithelial cell line were used to model normal implantation control. The monolayers were treated with TGF-β1 before the addition of embryos and the embryo attachment level on monolayers was determined and compared against non-TGF-β1 treated monolayers. Additionally, the effect of TGF-β1 on EMT marker TWIST1 expression was explored using immunohistochemistry.

Did the project change from that proposed in the application? If so, what changes were made and why? *(max 100 words)*

No changes were made to the project.

What were the main results/findings of the project? *(max 300 words)*

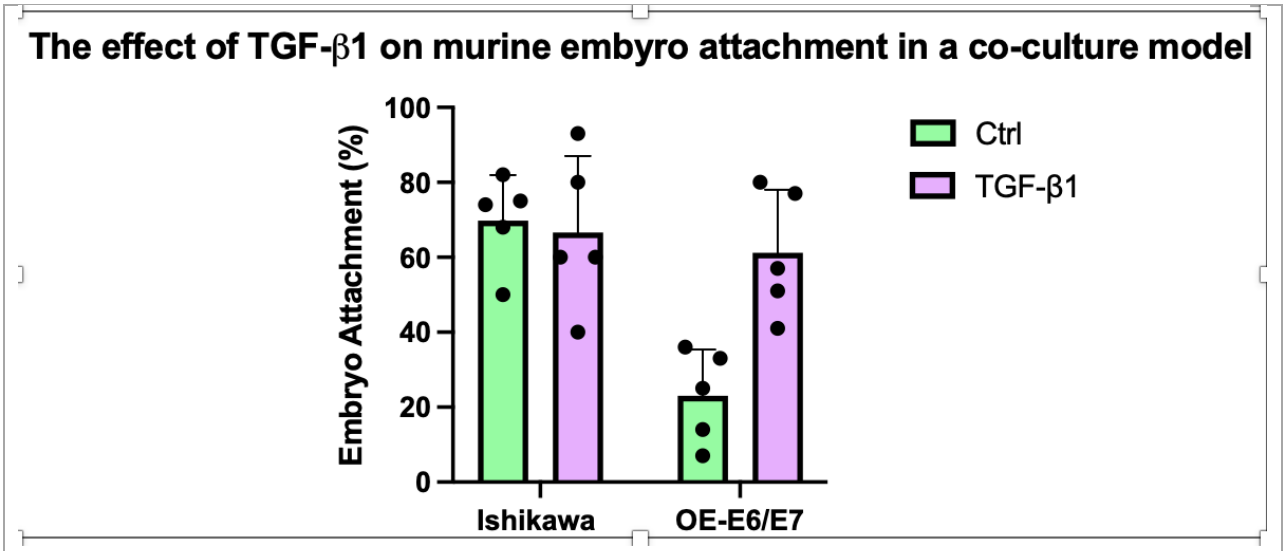

Figure 1: The effect of TGF-β1 *on murine embryo attachment level in co-cultures. Ishikawa = endometrial epithelial cell line, OE-E6/E7 = Fallopian tube epithelial cell line. Each data point represents results from one run. A total of 5 co-culture runs were conducted. In each co-culture run, 2 co-culture wells were prepared per treatment, with 10 embryos in each well.*

Fallopian tube epithelial cell monolayers were used to model tubal ectopic pregnancy (tEP) and endometrial epithelial cell monolayers were used to model normal intrauterine implantation: both cell lines were treated with TGF-β1 to induce epithelial to mesenchymal transition. The main results of this project were obtained by determining embryo attachment levels on TGF-β1 treated monolayers and comparing against embryo attachment levels on non-TGF-β1 treated monolayers.

The findings demonstrate the following: in non-TGF-β1 treated monolayers, the embryo attachment in the Fallopian tube cell line had an average of 23.0%, this was significantly lower than embryo attachment in the endometrial cell line which had an average embryo attachment level of 70.0% ($p<0.05$). This difference between embryo attachment in the two cell lines was as expected, since previous work done by the project supervisor demonstrated similar results. With TGF-β1 treatment, embryo attachment significantly increased in the Fallopian tube cell line. Specifically, embryo attachment increased from an average of 23.0% to 61.0% ($p<0.005$): this is similar to the attachment level seen in the endometrial cell line. On the other hand, TGF-β1 treatment did not lead to a significant change in embryo attachment in the endometrial cell line.

Additionally, fluorescent immunohistochemistry experiments were performed to investigate the level of TWIST1 (an EMT marker protein) expression in TGF-β1 treated and non-TGF-β1 treated co-cultures. There was no significant difference in the level of TWIST1 fluorescence between different treatment groups. Fluorescent immunohistochemistry experiments were also performed to investigate and compare the expression level of TWIST1, Oviductal Glycoprotein 1 (OVGP1) and Oestrogen receptor beta (ER-B) in TGF-β1 treated and non-TGF-β1 treated monolayers of the Fallopian tube cell line and the endometrial epithelial cell line i.e. without the presence of embryos. Results indicate that TGF-β1 exposure did not significantly change the level of TWIST1, OVGP1 and ER-B fluorescence level.

What do you conclude from your findings? *(max 150 words)*

Overall, this project used an in vitro co-culture model where murine embryos were cultured with monolayers of immortalised cell lines of human Fallopian tube epithelial origin and human endometrial epithelial origin to model tubal ectopic pregnancy and normal implantation, respectively. To investigate whether epithelial to mesenchymal transition may lead to tubal ectopic pregnancy, epithelial to mesenchymal transition was induced by the addition of TGF-β1.

This project demonstrated that inducing epithelial to mesenchymal transition using TGF-β1 significantly increased murine embryo attachment to Fallopian tube epithelial cell monolayers. The findings support the idea that epithelial to mesenchymal transition in fallopian tube epithelial cells may be involved in the underlying mechanism of tubal ectopic pregnancy in women. Investigating whether patients who suffer from tEP have higher levels of EMT in their Fallopian tube epithelial cells could be a potential topic for future research.

How has this experience influenced your thinking regarding your future/ongoing studies, and/or career choice? *(max 150 words)*

This summer research opportunity gave me an insight to what it is like to work as a researcher, and it has been a positive and enjoyable experience. This is a good indication that working in research is suitable for me and this encourages me to continue my education after my undergraduate degree in order to pursue a career in the research field in the future.

At the moment, I am planning to do a master's degree after completing my Honours degree next year. Previously, I have been considering both taught MSc degrees and research-based MSc degrees, however since I highly enjoyed working in a research lab during this summer project, this experience helped me to decide to focus on applying for research-based MSc programmes during the next application season.

Please use the space below to add any other comments/thoughts about the SRF Vacation Scholarship *(max 100 words)*

Student: I'm very thankful for SRF for funding my project, I had a wonderful and rewarding time at the lab. The instructions for the SRF application and report are clear and easy to follow.

Supervisor: Julia got on very well throughout her vacation scholarship, and it was a pleasure to have her in the lab. She worked hard, was careful in the laboratory, and put thought into the meaning of the results that she obtained. I am very pleased to see that she has managed to submit an abstract on the work to Fertility 2023, well deserved.