# SRF Vacation Scholarship report 2018

The form below should be completed by the student, then forwarded to the supervisor for approval and submission to [srf@conferencecollective.co.uk](mailto:srf@conferencecollective.co.uk) within 8 weeks of completing the project. Please submit the form as a Word document.

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.

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| **Student’s Name:** | Georgia May | **Student’s Institution/University:** | University of Sheffield |
| **Degree Title and year of study:** | Biomedical Science (Masters stream)  2nd year of study at time of scholarship acceptance | |  |
| **Supervisor’s Name:** | Alireza Fazeli  Lisa Thurston | **Supervisor’s Department and Institution:** | Academic unit of reproductive and developmental medicine,  Department of Oncology and metabolism, University of Sheffield |
| **Project Title:** | Investigating the role of extracellular vesicles as mediators of gamete-oviductal communication in the female reproductive tract | | |

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| **Briefly outline the background and aims of the project** *(max 200 words)* |
| Correct communication between oviduct cells and gametes is vital for establishing a successful pregnancy, and although this communication has been described in past studies, it is not yet understood how this is mediated (Fazeli and Holt, 2016).  Extracellular vesicles (EVs) form a fundamental aspect of communication mechanisms within a variety of physiological systems, and this may occur due to their transport of microRNAs (miRNAs) (Yanez-Mo et al., 2015). miRNAs impact cellular processes primarily through actions on protein translocation of genes, and mediating signal cascades responsible for differential splicing (Lewis et al., 2005). Communication between gametes and the female reproductive tract occurs in conjunction to an upregulation of proteins within the reproductive tract epithelia, because of which, EV mediated miRNAs may be considered as a potential method for this cross talk (Fazeli et al., 2004; Georgiou et al., 2007).  The aims of the project were as follows:  Objective 1: To characterize the size, concentration and electrical surface properties of EVs secreted by boar spermatozoa and porcine oviductal epithelial cells (POECs).  Objective 2: To determine whether co-incubation of boar spermatozoa with POECs effects EV production and/or characteristics.  Objective 3: To determine the miRNA content of EVs secreted by spermatozoa and POECs.  Citations:  Fazeli, A., Holt W.V., 2016. Cross talk during periconception period. Theriogenology 86, 438-442.  Fazeli A., Affara N.A., Hubank M., Holt W.V., 2004. Sperm-induced modification of the oviductal gene expression profile after natural insemination in mice. Biology of reproduction 71, 60-5.  Georgiou A.S., Snijders A.P., Sostaric E., 2007. Modulation of the oviductal environment by gametes. Journal of proteome research 6, 4656-66.  Lewis B.P., Burge C.B., Bartel D.P., 2005. Conserved seed pairing, often flanked by adenosines, indicates that thousands of human genes are microRNA targets. Cell 120, 15-20.  Yanez-Mo, M., Siljander P.R., Andreu Z., Zavec A.B., Borras F.E., Buzas E.I., Buzas K., Casal E., Cappello F., Carvalho J., Van der Grein S.G., Vasconcelos M.H., Wauben M.H., De Wever O., 2015. Biological properties of extracellular vesicles and their physiological functions. Journal of Extracellular Vesicles 4, 27066. |
| **Did the project change from that proposed in the application? If so, what changes were made and why?** *(max 100 words)* |
| My proposed aims were surrounding the changes in EV characteristics over the porcine oestrus cycle however, due to exciting developments in the laboratory focused around the miRNA content of EVs, I instead focused on characterizing the miRNA profiles of EVs produced by porcine oviductal epithelial cells (POECs) in the presence of sperm. |
| **What were the main results/findings of the project?** *(max 300 words)* |
| As accepted for presentation at Fertility 2019, this study investigated how the physical characteristics and miRNA content of EVs secreted by POECs alter in the presence/absence of spermatozoa.  Experimental groups (POECs-only, spermatozoa-only, POECs plus spermatozoa) were cultured in EV-depleted M199 for 24 hours, from which the EVs were then isolated using size exclusion chromatography. EV size, concentration and zeta potential were evaluated by nanoparticle tracking analysis. EV miRNA was also extracted and sequenced for bioinformatics analysis.  POECs secreted EVs 143.04 (±2.17) nm in size with a mean zeta potential of -25.69 (±0.8) mV. In contrast, sperm produced a distinct population of EVs determined by size (165.38 ± 4.73 nm) and zeta potential (-22.80 ± 0.56mV). There was no change in the number of EVs produced between the POEC alone, and POECs with sperm experimental groups. However, the larger sized sperm-EVs were totally diminished in POEC and sperm co-cultures, indicating a movement of EVs between sperm and POECs.  Eighty-one EV-mediated miRNAs were detected by sequence analysis, of which many were unique to specific experimental groups: POEC-only (13 miRNAs), sperm-only (1 miRNA), POEC plus spermatozoa (5 miRNAs). |
| **What do you conclude from your findings?** *(max 150 words)* |
| Bioinformatics analysis suggests that these miRNAs are involved in innate immune responses, cell proliferation and cellular migration. BLAST analysis showed that immune system related processes were targeted by both common and uniquely expressed miRNAs. miRNAs isolated from POEC and sperm co culture were found to target vesicle-mediated transport and in vivo embryonic development.  Clarification of the role of EVs in transporting miRNAs, and their influence on spermatozoa-oviductal communication will help us to develop new treatments for infertility, and aid the success rates of in vitro fertilization technologies. |
| **How has this experience influenced your thinking regarding your future/ongoing studies, and/or career choice?** *(max 150 words)* |
| This experience has provided me with a valuable insight into how medical research is undertaken, something that my studies have not been able to provide. From attending, and presenting in, the fortnightly laboratory meetings, I have gained a strong knowledge of the working environment within hospital laboratories. It has also provided a platform from which I have been able to attend conferences such as the Annual COST Action Meeting on In vitro 3‐D total cell guidance and fitness, Croatia, 2018, and Fertility 2019, Birmingham, UK.  In addition it has provided me with greater background and practical knowledge of many techniques and equipment such as: cell culture, size exclusion chromatography, protein extraction, northern/western blotting, primer design, cDNA synthesis, qPCR and relative PCR. I believe that this is beneficial not only for ensuring my learning is thorough, but also that I will be able to carry these skills into my future careers. |
| **Please use the space below to add any other comments/thoughts about the SRF Vacation Scholarship** *(max 100 words)* |
| ***Student:*** *I would like to take the opportunity to thank both SRF and my supervisors for enabling me to not only make the most of a brilliant opportunity, but also for the beneficial impact I believe that this has had on my confidence and knowledge around medical research.*  ***Supervisor:*** |