### Research theme
Respiratory Disease

### Supervisor
Dr Tomás Carroll

### Web page
[https://research1.rcsi.ie/pi/gmcelvaney/team.asp](https://research1.rcsi.ie/pi/gmcelvaney/team.asp)

### Co-supervisor
Professor N G McElvaney

### Web page
[https://research1.rcsi.ie/pi/kHennelly/index.asp](https://research1.rcsi.ie/pi/kHennelly/index.asp)

### Title of PhD project
The impact of alpha-1 antitrypsin deficiency heterozygosity on immune function.

### Brief project description
Cigarette smoking is the principal environmental risk factor for the development of chronic obstructive pulmonary disease (COPD); however, the pronounced variability in the development of chronic airflow obstruction among smokers suggests that other genetic and environmental factors are important in COPD pathogenesis. Severe alpha-1 antitrypsin deficiency (AATD) is a proven genetic risk factor for COPD but is found in only 1-2% of COPD subjects. AATD is a hereditary disorder characterized by low circulating levels of the antiprotease alpha-1 antitrypsin (AAT) which renders lung tissue susceptible to proteolytic degradation. The pulmonary disease which develops is characterized by neutrophil-dominated airway inflammation and elevated intra-pulmonary protease levels. The gene encoding AAT is the SERPINA1 gene and the two most common SERPINA1 mutations associated with AATD are the Z and S mutations, however, the vast majority of AATD individuals with emphysema are ZZ homozygotes. Crucially, there is a lack of clarity pertaining to MZ and SZ heterozygotes and their risk of COPD. This is a vital clinical and public health question, as the global burden of heterozygote AATD is under-recognised. For example there are over 170,000 MZ and 10,000 SZ individuals in Ireland alone.

We have previously investigated circulating monocytes isolated from clinically stable asymptomatic ZZ individuals and shown exaggerated cytokine responses and evidence for endoplasmic reticulum (ER) stress compared to MM individuals. This was the first study to show that Z AAT protein accumulation induces ER stress in peripheral blood monocytes. However, the findings raise several important questions, particularly what other functions of monocytes are impaired by Z AAT expression? In addition, we have preliminary data demonstrating that elements of ER stress pathways are up-regulated in ZZ neutrophils. This raises the possibility that ER stress pathways are activated in heterozygotes, and we hypothesize that Z AAT expression in MZ and SZ monocytes and neutrophils is responsible.

The central hypothesis of this research project is that MZ and SZ heterozygotes are at increased risk of developing COPD through immune dysfunction. We will recruit healthy non-smoking MZ and SZ individuals with normal lung function to correct for disease-related inflammation and compare them to healthy MM individuals. We will explore the functional effects of heterozygosity on circulating monocyte and neutrophil function, and explore a role for ER stress. Overall, we aim to determine whether intermediate AAT deficiency is associated with increased risk of COPD and outline mechanisms behind this predisposition.

### Project Outcomes:
In addition to generating important new information which could explain the predisposition to COPD in AATD heterozygotes, the findings in this study may also have profound implications for individuals with severe AAT deficiency and in AAT-replete COPD subjects.

### Ethical approval:
Ethical approval for this study to be carried out has been obtained from the Beaumont Hospital.
Hospital Ethics committee.

**IP/commercialisation potential:**
Currently, the only specific therapy for severe alpha-1 antitrypsin deficiency (ZZ) is intravenous augmentation therapy. The McElvaney lab is at present leading a multi-centre study evaluating the clinical effect of this therapy. The knowledge acquired from the proposed project may lead to recommendation of intravenous alpha-1 antitrypsin (AAT) augmentation therapy in SZ and/or MZ heterozygotes.

**Key distinguishing points:** e.g. multidisciplinary approach; training and access to specific facilities; access to specific meetings/conferences/learned society meetings.

This project forms part of the broad research strategy of the McElvaney group and the mechanisms and pathways studied within this project are directly applicable to not only alpha-1 antitrypsin deficiency (AATD), but to other inflammatory lung diseases such as cystic fibrosis and chronic obstructive pulmonary disease (COPD), as well as conformational disorders such as Alzheimer's disease in which pathological protein misfolding occurs. In addition, this project has a significant translational aspect. It involves biological samples and will take place on a hospital campus with easy access to clinical samples and patient data. The McElvaney lab is situated adjacent to the RCSI Clinical Research Centre, where several clinical trials are already underway in AATD and cystic fibrosis. Importantly, the National Alpha-1 Antitrypsin Deficiency Targeted Detection Programme is based in RCSI Education & Research Centre at Beaumont Hospital. This programme has so far identified over 120 ZZ, 120 SZ and 1200 MZ alpha-1 antitrypsin deficient individuals as part of an ongoing screening programme. Beaumont Hospital is the site of the national referral centre for AATD. As the national referral centre we provide a rapid access weekly AATD clinic for newly-diagnosed ZZ, SZ, and MZ individuals. The clinic is coordinated by a dedicated AATD clinical research nurse and AATD individuals are seen by a multidisciplinary team of doctors, nurses, and physiotherapists with international best practice standards of care followed. This affords us is the real possibility of bench to bedside translational research, and also easy access to clinical samples (blood, sputum, lavage etc.) from AATD patients. The ready availability of large, well-characterised populations of ZZ, SZ and MZ individuals enhances the feasibility of this project.

The student will be provided the opportunity to present his/her work at national and international conferences such as the Irish Thoracic Society annual meeting, the European Respiratory Society and American Thoracic Society annual conferences.

**Undergraduate disciplines relevant:** Biology, Biochemistry, Microbiology, Biotechnology, Genetics, Biomedical Science.

**References:**

**Skills & techniques that the student will learn from the project**
During this project the student will develop the skills to work collaboratively as part of a research team and also independently. The student will acquire techniques such as monocyte and neutrophil isolation, cell culture, RNA isolation, cDNA synthesis, real-time quantitative RT-PCR, Western blotting, ELISA, the analysis of superoxide production by cytochrome c assay, the analysis of chemotaxis by Boyden chamber assay, bacterial killing.
assay, and confocal laser scanning microscopy.