Project Details	
Project Code	MRCPHS25Br Lloyd-Lewis
Title	Under pressure: Investigating the role of tissue density and mechanics in breast cancer development
Research Theme	Population Health Sciences
Summary	A high breast mammographic density is a major risk factor for breast cancer, yet the underlying molecular mechanisms remain unclear. In this interdisciplinary project, the student will combine advanced methods in genetic epidemiology with lab-based cell biology techniques (e.g. confocal imaging, flow cytometry, 'omics) in patient-derived breast organoids and in vivo models to investigate how increased tissue density and stiffness predisposes the breast to tumourigenesis.
Description	**IMPORTANCE** Breast cancer is the most common cancer in women, with over 4000 new cases diagnosed monthly. As incidence rates continue to rise globally, there is an urgent need to identify new, modifiable risk factors for breast cancer prevention and early detection. A high breast mammographic density (MD) is one of the strongest risk factors for breast cancer, conferring a x4-6 increase in risk. Our recent work implied that early-life adiposity decreases breast cancer risk largely through reducing breast MD in adulthood. This finding is exciting as it suggests that adult MD is modifiable during the pubertal growth period, which might provide opportunities to intervene during adolescence to reduce lifetime MD and associated breast cancer risk. The mechanistic molecular pathways linking breast MD to cancer however remain poorly understood. Addressing this gap in knowledge is critically important as progress in this area has the potential to transform precision cancer prevention approaches in women with elevated breast MD. High MD breast is associated with increased tissue stiffness. While a stiffened extra-cellular matrix (ECM) is implicated in accelerated mammary (breast) tumour progression, how tissue stiffness contributes to tumour predisposition remains unknown. Accumulating evidence, including our exciting preliminary data, suggests that tissue stiffness regulates mammary stem cell activity and differentiation dynamics. As re-acquiring stem cell fate is considered an early step in breast cancer, this project will investigate the hypothesis that increased breast MD perturbs normal mammary stem cell activity and differentiation dynamics, and that this places the breast epithelium in a high-risk, pre-malignant state. **RESEARCH TRAINING** To address this hypothesis, the project will use an interdisciplinary approach that combines methods in genetic epidemiology, bioinformatics and laboratory-based techniques in molecular, cell and stem cell biology. The student will therefore acquire a versatile and highly sought-after skill set during their PhD. Aim 1. Determine the genes and traits associated with high breast MD that are causally associated with breast cancer Training in genetic epidemiological techniques such Mendelian Randomisation (MR) at the world leading MRC Integrative Epidemiology Unit at the University of Bristol will enable the student to identify genes associated with high breast MD and to test their causal association with

	breast cancer. In addition to using the largest genetic datasets of breast MD and breast cancer available, they will also map the contribution of common genetic variation to breast density as detected by MRI in young (age 20-22 years) nulliparous women available from the Avon Longitudinal Study of Parents and Children (ALSPAC) birth cohort. This unique dataset includes detailed metabolic, hormone and body fat mass measurements taken at 8, 16, 18 and 25 years of age for each individual, which can be used to identify novel traits associated with breast MD. Aim 2. Determine whether increased tissue stiffness alters mammary epithelial stem cell properties and molecular profile Training in laboratory methods will allow the student to determine how perturbing ECM stiffness affects mammary epithelial stem cell activity, differentiation dynamics and gene expression profile. The student will address this in mammary gland tissues in vivo using available genetic lineage tracing mouse models, and ex vivo using patient-derived breast organoid cultures. The student will gain skills in experimental cell and molecular biology techniques including 3D organoid culture, fluorescence confocal and light-sheet microscopy, flow cytometry, immunohistochemistry, RNA-sequencing, as well as in image processing and bioinformatic analysis of RNA-seq datasets. Aim 3. Determine whether identified mechanically regulated genes are causally associated with breast cancer Following training received in earlier aims, the student will steer their project to prioritise tractable genes and signalling networks of interest to them for further investigation. Data collected will also inform further genetic epidemiological analysis. For example, whether mechanically regulated genes identified in Aim 2 are causally associated with breast cancer can be investigated using MR and validated in breast cancer patient datasets (e.g. The Cancer Genome Atlas). Functional investigation of validated hits can subsequently be performed in laboratory in vivo and in vitro models using the same techniques applied in Aim 2.
Supervisory Team	
Lead Supervisor	
Name	Dr Bethan Lloyd-Lewis
Affiliation	Bristol
College/Faculty	Health and Life Sciences
Department/School	Cellular and Molecular Medicine
Email Address	bethan.lloyd-lewis@bristol.ac.uk
Co-Supervisor 1	
Name	Dr Rebecca Richmond
Affiliation	Bristol
College/Faculty	Health and Life Sciences
Department/School	Bristol Medical School
Co-Supervisor 2	
Name	Professor Richard Clarkson
Affiliation	Cardiff
College/Faculty	
Department/School	School of Biosciences
Co-Supervisor 3	
Name	Dr Marina Vabistsevits

Affiliation	Exeter
College/Faculty	
Department/School	Exeter Medical School