I use mathematics to address problems in medicine by developing physiologically structured models to understand the mechanisms underlying treatment resistance. I leverage these models to develop evolutionarily-inspired and clinically-actionable therapeutic strategies through the development of novel analytical techniques. A significant portion of my planned research concerns the search for a functional cure of human immunodeficiency virus type 1 (HIV). My interests involve the evolution of resistance against broadly neutralizing antibodies, treatments that attempt to induce post-treatment control by targeting immune exhaustion, and the establishment of the heterogeneous reservoir of latently infected cells. A further portion of my research concerns the epigenetic regulation of treatment resistance in solid cancers and understanding the evolutionary mechanisms that underpin this phenotypic resistance.

Developing physiologically relevant mathematical models to address these problems will necessarily entail new techniques in the development, simulation, and analysis of structured population models. These include the creation of novel numerical methods to simulate and parametrize the models, the extension of existing numerical bifurcation methods to these infinite dimensional dynamical systems, and the development of analytical approaches to identify key model properties and translate this mathematical understanding to new biological knowledge. Here, I summarize the broad themes of my current research before discussing plans for new directions investigating the evolutionary mechanisms underlying treatment resistance in HIV infection and cancer.

Prior Work

Structured population models are typically infinite dimensional and are non-trivial to develop, simulate, and analyse [15]. As a result, it is common in mathematical biology to use simpler compartmental ordinary differential equation (ODE) models to mimic time delays that arise naturally in structured population models [20, 37]. The well-known linear chain technique shows that these compartmental ODEs –in the specific case where the transit rate between compartments is constant– is equivalent to an Erlang distributed delay differential equation (DDE) [33]. I extended this classical work to include both arbitrary sojourn periods and environmentally-driven variable transit rates by considering the underlying McKendrick type partial differential equation (PDE) for general population renewal [10]. I showed that this PDE is equivalent to a state dependent distributed DDE, that this general setting encompasses the common state dependent discrete DDE, and generalized the linear chain technique to include variable transition rates and concatenated ageing processes [10]. I have recently extended this work to general compartmental models including both delayed arguments and non-linear transit rates [8], and I derived explicit expressions for equilibria and the corresponding characteristic function in a manner that is significantly less computationally demanding than working with the compartmental system.

My work on structured population models bridges the three major categories of blood production models used in quantitative systems pharmacology (QSP): transit compartment ODEs, discrete DDEs and physiologically structured QSP models. These mathematical models, in particular the "gold standard" Quartino model [27], are extensively used in the pharmaceutical sciences to improve support of the blood production system during chemotherapy. By studying the equivalent structured model, I showed that the Quartino model misspecifies the neutrophil precursor maturation stage as an additional proliferative stage [5], and that such misspecifications can be avoided by explicitly modelling the proliferative stage as a physiologically-structured PDE [10]. Drawing upon this work, I developed and validated a model of neutrophil and monocyte production and showed that monocyte concentrations act as an early warning sign for chemotherapy-induced neutropenia [12].

Explicitly including intra- or inter-population heterogeneity via structured poulation models increases their biological relevance [14]. To understand the role of intra-tumour heterogeneity in cancer progression, I developed a distributed DDE mathematical model of tumour growth in the presence of immune pressure

[11]. I completely characterised the importance of tumour-immune interaction by deriving a necessary and sufficient condition for immune-mediated disease control, and showed that decreasing tumour-immune interaction leads to tumour expansion through a transcritical bifurcation. I showed that intra-tumour heterogeneity acts to destabilize the disease-free state. I then extended the model to include oncolytic viral therapy, fit data from late stage melanoma, and utilized the model to perform an *in silico* clinical trial [9]. Using insights gained from my modelling, I developed a clinically actionable combination therapy schedule that significantly improves virtual prognosis while minimizing treatment burden. In recent work by an undergraduate researcher under my supervision, we showed that these physiologically structured models identify the biological processes underlying disease progression by linking the patient-specific intrinsic tumour growth rate with the appropriate combination oncolytic virus treatment strategies [21].

Future directions: structured equations in mathematical biology

A significant obstacle in the use of structured equations in mathematical biology is the lack of appropriate numerical tools to facilitate their study and simulation. In general, numerical methods to simulate distributed DDEs must be developed on a model-by-model basis and are complicated by the evaluation of the integral term, which necessarily induces overlapping in each Runge-Kutta (RK) step. To address this, numerical methods must output a continuous extension of the solution to calculate the intermediate RK steps. The theory of these functionally continuous RK methods is beginning to be developed [3] but has not been explicitly extended to distributed DDEs with infinite delay. Thus, I will develop functionally continuous RK methods to accurately and efficiently simulate distributed DDEs. Numerical techniques for DDEs with infinite delays are quite limited, with most existing work either artificially truncating the delay [22] or imposing artificial equilibrium constraints on the initial data [26]. Accordingly, my work will involve the development of methods to accurately compute these integrals at each intermediate RK step by mapping the semi-infinite convolution integral present in most distributed DDEs to an integral over a finite domain, and the derivation of conditions to ensure that these mappings are both well-defined and preserve the accuracy of the RK scheme.

In collaboration with Francesca Scarabel at the University of Manchester, I will adapt existing pseudospectral techniques to study bifurcations in structured epidemic models [19, 16]. Our preliminary work shows that structuring the infected class by infection age and including waning infectivity both increase biological relevance and cause waves of infection rather than endemic infection. Confirming these bifurcations either analytically or through rigorous numerical bifurcation analysis will require the development of new techniques for these non-local functional equations.

Finally, the equivalence between Erlang distributed DDEs and a system of ODEs is well-known, and implies that the dynamical system encoded by an Erlang distributed DDE is finite dimensional. As the Erlang distribution is a specific case of the gamma distribution, I am interested in quantifying how accurately we can approximate the infinite dimensional dynamics of a general gamma distributed DDE using a finite dimensional model. In first steps towards this goal, I have recently shown that the characteristic function of a generic gamma distributed DDE admits finitely many roots. This suggests that dynamics near equilibria are finite dimensional, and I have derived more accurate finite dimensional approximations of gamma distributed DDEs.

Future directions: treatment resistance and post-treatment control in HIV infection

While antiretroviral therapies (ART) are remarkably effective in controlling viral loads and increasing life expectancy, there is still no functional cure available for individuals with HIV. Broadly neutralizing antibodies (bNAbs) have become increasingly important in the search for a functional cure [35], and a number have recently been tested in HIV positive individuals [7, 23]. While these antibodies induce a transient decrease in viral load in HIV infected individuals [4], existing bNAbs have *not* led to sustained viral control. In particular, viral rebound occurs concurrently with the emergence of antibody resistant

viral strains rather than antibody washout [30, 6, 23]. I developed a series of mathematical models to understand both the viral dynamic response and the evolution of resistance in a recent phase I/II clinical trial of the novel bNAb PGT121. In particular, my work illuminated the mechanisms by which PGT121 led to long-term virological control in two patients. My modelling indicates that treatment-induced competitive release of the resistant population leads to viral rebound and treatment failure.

My modelling indicates that if the development of this resistant population can be avoided through combination therapies, then highly potent bNAbs such as PGT121 may be a promising therapeutic intervention to induce long-term viral suppression in HIV positive individuals. Genetic profiling and *in vitro* neutralization assays of escape virus indicates that sensitivity to other bNAbs is maintained or possibly enhanced following PGT121 therapy. Accordingly, my future research will investigate this population-level resistance by using structured models to explicitly consider the dynamic pathways leading to resistance against bNAb mono- and combination therapies. These models will use the strain specific *in vitro* neutralization efficacy to structure the viral population according to therapy resistance. I will use temporal sequencing data to validate the model predictions. After establishing a model capable of predicting viral response to single and multiple antibody therapy, I will use these models to predict optimal bNAb combinations and scheduling. These strategies will act synergistically to inhibit the development of resistance by maintaining distinct evolutionary selection pressure on resistant viral subspecies.

When ART is initiated during primary HIV infection, roughly 5–15% of patients achieve sustained post-treatment control (PTC) [29]. However, ART has been less effective in establishing PTC in chronically infected patients. Recent work has identified the size and heterogeneity of the reservoir of latently infected cells as a possible determining factor in the establishment of PTC [29]. In a complementary search for a functional cure of HIV, recently developed treatments attempt to activate latently infected cells and mobilize the immune system against these newly activated cells [34]. Thus, by combining ART and latency reversing treatments, it may be possible to control the size of the latent reservoir at cessation of ART therapy and to provide a functional cure. However, this "shock and kill" strategy has not been clinically effective [34].

I will use mathematical modelling to determine the physiological characteristics of patients most likely to achieve PTC. My modelling will focus on quantifying the roles of immune exhaustion and latent reservoir heterogeneity in establishing PTC. I will extend the mathematical model from [13] to include heterogeneity in the latently infected cell population and examine the role of anti-exhaustion immunotherapies on establishment of PTC. By using clinical and experimental data, I will parametrize the resulting mathematical model and use analytical and numerical techniques to identify possible strategies of therapeutic intervention. These therapeutic interventions, such as immune checkpoint inhibitors or latency reversing agents, will effectively force the infection into the basin of attraction of the low viral load set point corresponding to PTC. Accordingly, characterisation and prediction of the qualitative change in viral dynamics following treatment is particularly amenable to the language of dynamical systems. Thus, by using a mix of analytical and numerical bifurcation techniques, I will identify the physiological processes most likely to determine whether a given patient will exhibit PTC.

Future directions: evolutionary therapy in cancer

Despite advances in targeted and precision medicine, most patients with advanced cancer will experience drug resistance, treatment failure and, ultimately, disease recurrence [28, 25]. This resistance, long thought to result from genetic heterogeneity and evolution, is increasingly understood as the result of non-mutational evolutionary adaptations to therapy [24, 2]. Recent studies have indicated that epigenetic regulation of genetically identical non-small cell lung cancer cells induces a reversible drug-tolerant phenotype that expands during cytotoxic chemotherapy [32, 18]. This phenotypic heterogeneity is modulated by a number of complex physiological factors, including the current state of the tumour and tumour microenvironment

as well as the life history of the parental cell [31, 36]. Accordingly, as systems-level experiments of these complex and non-linear interactions are currently intractable, standard experimental techniques are unable to probe the physiological determinants of non-genetic heterogeneity. Thus, biologically-relevant and physiologically-structured mathematical models are integral to our understanding of how therapeutic selection pressure shapes the epigenetic evolution of malignant tumours. Consequently, it is presently unclear how to adapt evolutionarily inspired therapies [17] to address reversible non-genetic mechanisms that change on a faster timescale than Darwinian evolution.

My research will address the systemic determinants of epigenetic resistance through the development of mechanistic mathematical models of tumour evolution in response to therapy. Unlike existing models, these models will take the form of physiologically structured PDEs that are explicitly designed to study intra-clonal population heterogeneity, and that will serve as the basis of the development of evolutionarily inspired therapies that explicitly address non-mutational resistance. These mathematical models will facilitate understanding of how therapeutic selection shapes the epigenetic evolution of malignant tumours. With this understanding in hand, I will use techniques from dynamical systems, pharmacometrics, and numerical analysis to develop rational and clinically-actionable treatment schedules to steer this epigenetic evolution towards desired clinical outcomes. This rational use of drug-interventions will be particularly important when considering large heterogeneous tumours where single drug therapies are unlikely to be successful. There, evolutionary steering may involve drug cycling to maintain a drug-sensitive population of cells and thus limit treatment failure or treatment strategies designed to induce collateral sensitivity wherein resistance to the first therapy increases sensitivity to subsequent therapeutics [1]. This work will benefit from my existing collaboration with Sandy Anderson, the head of the Integrated Mathematical Oncology department at the Moffitt Cancer Center.

Outlook

My research will make important contributions to the theory and application of structured equations in mathematical biology. As structured population models permit the explicit study of heterogeneity in disease progression, my work will make original contributions by developing mathematical methodologies with real clinical implications. Answering these questions will require the development of novel mathematical models, techniques for their simulation and analysis, and will benefit from detailed collaboration with scientists in a number of fields. Consequently, my work offers numerous opportunities for trainees from the undergraduate to post-doctoral level in fields ranging from QSP, to numerical analysis, dynamical systems, mathematical modelling, and mathematical analysis. Further, my trainees will benefit from my existing collaborations and experience with industrial applications of mathematics. Taken together, my research will develop new mathematical models to understand the physiological mechanisms underlying disease and use these models to develop evolutionarily-inspired and clinically-actionable therapeutic strategies to improve clinical outcomes.

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