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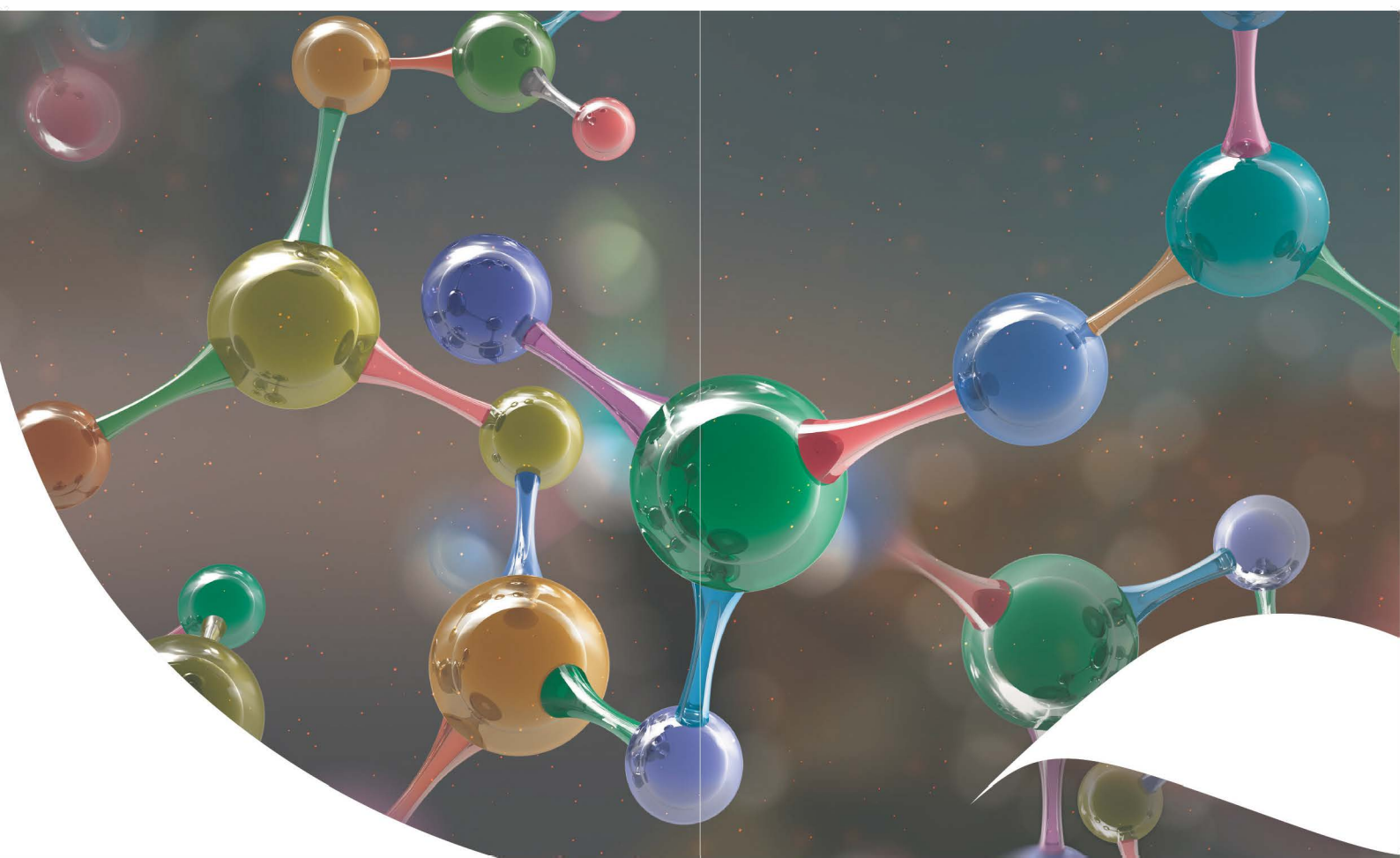
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CAI SYMPOSIUM 2022

Nuclear Medicine and Radiochemistry

Monday 16 May 2022

9:00am — 6:30pm

AIBN Seminar Room & Centre for Advanced Imaging

8:30 - 9:00	*Registration*	*AIBN Seminar Room*
9:00 - 10:00	PLENARY - Prof Rachel Codd Chair: Brett Paterson	AIBN Seminar Room
10:00 - 10:40	*Morning tea*	*Level 2 CAI*
10:40 - 12:00	HDR & Postdoc talks Saikat Ghosh Melyssa Grieve Ting Xiang Lim James Humphries Chair: Amber Prior	AIBN Seminar Room
12:00 - 1:30	*Lunch & Posters*	*Level 5 CAI*
1:30 - 2:00	KEYNOTE - Dr Christian Wichmann Chair: Brett Paterson	AIBN Seminar Room
2:00 - 3:00	HDR & Postdoc talks Nick Fletcher Jessica Van Zuylekom Salma Ahmed Chair: Adrian Almass	
3:00 - 3:30	*Afternoon tea*	*Level 2 CAI*
3:30 - 4:30	HDR & Postdoc talks Rajat Vashistha Vanessa Soh Biswa Prasanna Mishra Chair: Hamed Moradi	AIBN Seminar Room
4:30 - 6:30	Posters, prizes & pizza	Level 5 CAI

PLENARY

**Exploiting Chemical Biology to Expand the Chemical Space of Radiometal Chelators**

Professor Rachel Codd
Professor of Bioinorganic and Medicinal Chemistry
School of Medical Sciences
The University of Sydney

The multi-component nature of an agent destined for use in molecularly targeted nuclear medicine (radiometal-chelator-linker-peptide/antibody) presents both challenges and opportunities for design optimisation. As relevant to the radiometal chelator, the field often defaults to using an 'off-the-shelf' chelator, based on availability, and well understood chemistry. Our group is challenging the known and developing methodological approaches towards accessing new boutique chelators that can be tailored for a given radiometal. This is useful, since high-affinity and high-selectivity chelators will be less prone to in vivo disassociation and trans-chelation phenomena and simultaneously opens new patentable chemical space. Our work has adopted a broad spectrum of methods in chemical biology to generate new radiometal chelators, with a focus on Zr(IV)-89 as an in-development radiometal for immunological positron emission tomography (PET) imaging. We have used metal-templated synthesis to characterise two macrocyclic chelators for Zr(IV) with unusual architectures. We have drawn inspiration from chelators produced by bacteria and used this system as a platform to bioengineer new Zr(IV) chelators with improved water solubility. We have begun pushing further boundaries in synthetic and chemical biology and canvassing the possibility of using enzymes to biosynthesise structurally diverse metal chelators in vitro. Taken together, this body of work aims to move the field beyond standard chelators and to provide both general methods and new chelators with coordination chemistry to enable full benefit from the expanding Radiometal Periodic Table.
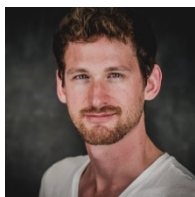
KEYNOTE

Advances in ^{89}Zr- and ^{225}Ac-based radiopharmaceuticals for diagnostic and therapeutic purposes

Dr Christian Wichmann
Tumour Targeting Laboratory
Olivia Newton-John Cancer Research Institute

Zirconium-89 and Actinium-225 are gaining significance as radioisotopes for diagnostic and therapeutic applications in a variety of diseases. Clinical PET bioimaging studies with ^{89}Zr-labelled antibodies are underway and targeted alpha-particle therapy with ^{225}Ac-labelled small molecules, peptides, and antibodies has shown great success in preclinical and clinical settings.

This talk will highlight recent developments in the radiosynthesis of ^{89}Zr- and ^{225}Ac-labelled antibodies and present preclinical evaluation of therapeutic efficacy in a murine glioma model.

ORAL

Design and evaluation of degradable, pH-sensitive camptothecin-loaded polymersomes from RAFT-based diblock polymers

Salma Ahmed[1,2,*], *James Humphries*[1,2,] *Nicholas Fletcher*[1,2], *Craig Bell*[1,2], and *Kristofer Thurecht*[1,2]

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Developing safe and effective nanoscale drug delivery systems for cancer treatment is an ongoing priority for researchers from different scientific backgrounds. The therapeutic performance of these systems, especially when used to improve the solubility of hydrophobic drugs, depends on their shapes, sizes and dynamic behaviour.[1] Herein, we report the synthesis of novel degradable amphiphilic diblock copolymers, by RAFT-terpolymerization of 2-methylene-1,3-dioxepane (MDO), vinyl acetate (VAc) and vinyl bromobutanoate (VBr) monomers. By optimising the length of the hydrophobic blocks, we developed synthetic platforms for the production of pH-sensitive polymersomes (~100 nm), **Figure 1**).[2] We selected the antineoplastic drug, camptothecin, as a model drug to be delivered by our polymeric nanoparticles due to its high toxicity and poor water solubility, which typically restrain its application in chemotherapy. We thoroughly investigated the morphology, stabilitiy, drug encapsulation, release profiles, cellular association and dose-dependent cytotoxicity of polymersomes when tested against MDA-MB-468 breast cancer cells. Our delivery system demonstrated good drug loading efficiency, pH-sensitivity, sustained release rates, and efficient *in vitro* cytotoxicity and cellular uptake. We then assessed the *in vivo* efficacy of polymersomes when used to interrogate BALB/c nude mice bearing xenograft MDA-MB-468 human breast cancer tumours. Polymersomes proved high efficiency in terms of their tumour accumulation, tumor regression, and reduction of side-effects, and have also showed 100% survival rate by the end of the therapeutic study, compared to free drug. Our work confirms the advantages of these nanoparticles to deliver lipophilic and difficult to solubilize drugs.
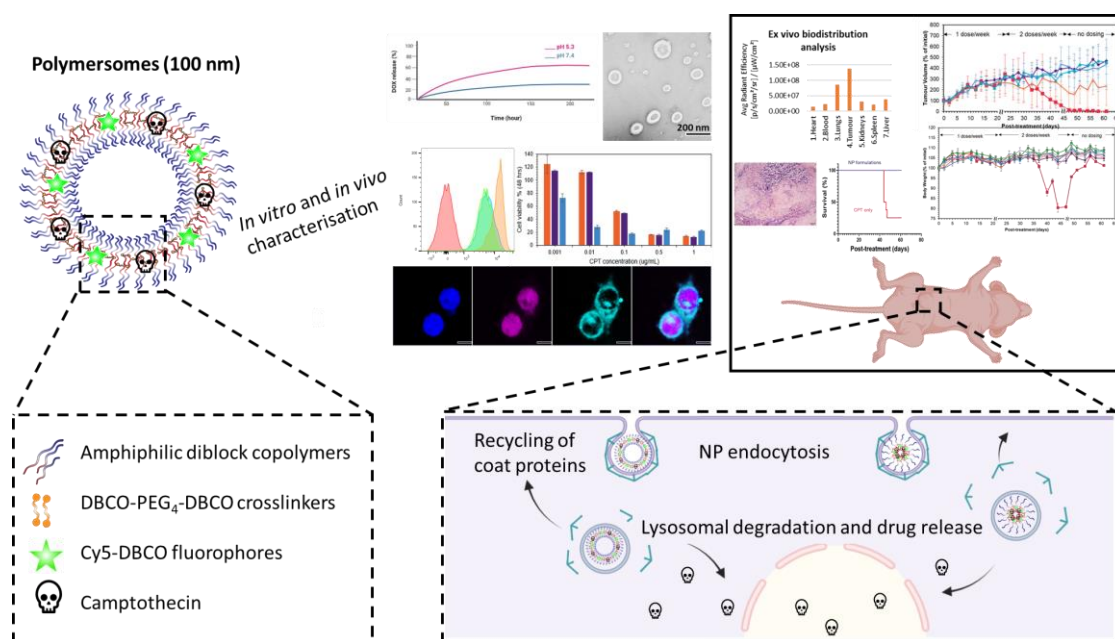
Figure 1. Self-assembled polymersomes and their application as pH-sensitive chemotherapeutic delivery systems for camptothecin.

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ORAL

Engineering a PSMA-targeted antibody for accelerated clearance while maintaining tumour accumulation for targeted radiotherapy

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Prostate-specific membrane antigen **(**PSMA) targeted radiotherapy offers an appealing approach for prostate cancer treatment. Existing small molecule and native antibody carriers have inherent pharmacokinetic and biodistribution properties that limit utility. Small molecules lack the target specificity and level of tumour localization of biologics, and have high renal exposure, whereas antibody-delivered radiation is dose-limited by hematologic toxicities incurred by a long serum half-life.

In this project, rational design processes were used to engineer novel humanized anti-PSMA antibodies with reduced Neonatal Fc Receptor (FcRn) recycling function. This aimed to reduce pharmacokinetic profiles analogous to small molecule tracers, minimizing blood & bone marrow exposure, yet retaining the benefits of enhanced tumor localization, specificity and reduced renal toxicity.

In vitro assays demonstrated successful abrogation of FcRn binding whilst maintaining PSMA affinity comparable to legacy comparator; HuJ591. Constructs efficiently labelled with ^{64}Cu and in vivo preclinical assessments (Fig1) demonstrated tumor accumulation comparable to HuJ591 (Fig1B) with a pronounced reduction in circulation time; completely clearing from circulation within 3 days (Fig1C). This pharmacokinetic modulation resulted in a >4-fold higher Tumour:Blood ratio at 48h post-injection for engineered constructs
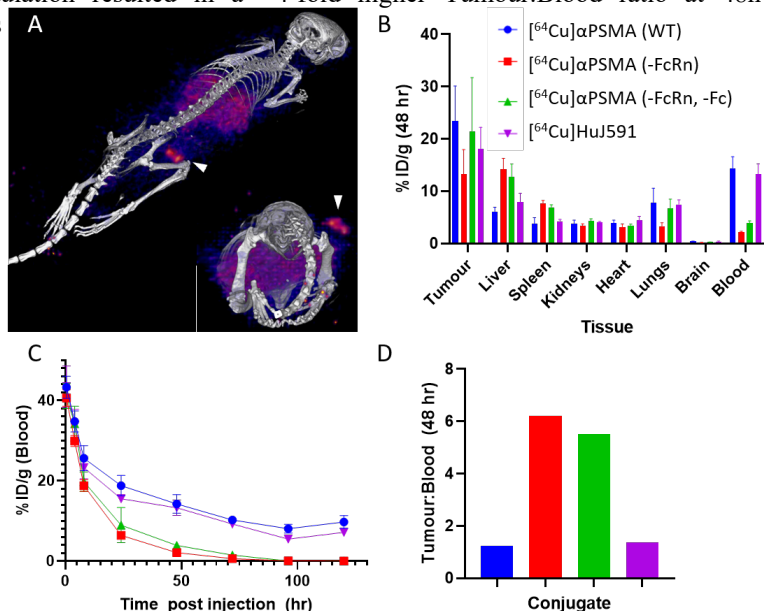
Figure 1. Preclinical PET imaging (A) of [^{64}Cu]-labelled engineered construct in LNCaP xenograft model. *Ex vivo* biodistribution (B), blood profile (C) and Tumour:Blood ratios (D) show modulated pharmacokinetics of engineered constructs compared to HuJ591.

Engineered PSMA targeting antibodies with abrogated FcRn binding were produced and demonstrated successful shortening of circulation time whilst maintaining tumour accumulation. Such carriers with modulated pharmacokinetics are appealing for radiotherapy interventions with reduced residence time and associated side-effects.

ORAL

Novel antibody theranostics for Glypican-1 over-expressing solid tumors

Saikat Ghosh[1,2,3]*, Pie Huda*[2,3]*, Nicholas Fletcher*[2,3]*, Zachary Houston*[2,3]*, Christopher Howard*[2]*, Yanling Lu*[3]*, Maria Lund*[4]*, Douglas Campbell*[4]*, Bradley Walsh*[1,4]*, Kristofer Thurecht*[1,2,3]

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[4]Glytherix Ltd., Macquarie Park, Sydney, New South Wales, Australia

Glioblastoma (GBM) is recognized as the most aggressive form of brain cancer. Despite advanced treatment strategies like surgery, radiation and chemotherapy, the average survival period of patients with GBM is less than 15 months, and this statistic has not improved in the last 30 years. Unfortunately, stage-4 GBM is considered an untreatable disease due to its ability to rapidly grow undetected, subsequently spreading to nearby areas of the brain. Existing GBM treatment strategies like surgery involves invasive measures that compromise the integrity of the brain, while radiation therapy affects healthy cells in patients causing unwanted side-effects. Even diagnosis in the form of tumor biopsies is a risky procedure. We hypothesize that these challenges could be mitigated through 'radioimmunotherapy' (use of novel antibody-based agents carrying therapeutic and/or diagnostic radioisotopes), to complement the existing strategies.

My research (in collaboration with industry partner Glytherix, Ltd., Australia) focuses on developing therapeutic and diagnostic agents which would selectively target GBM cells, sparing the healthy cells of the body. Such specificity is achieved through the use of antibodies, proteins which are an integral part of our body's natural defence system. In my research, I use a particular antibody engineered to bind GBM cancer cells. This antibody was further modified to carry radioactive elements suitable for tumor imaging applications or with cancer cell-killing properties. These 'targeted drug' formats could improve the efficiency of treating GBM by restricting the therapeutic radiation dose to the tumor mass, thereby reducing side-effects. Moreover, early detection of tumors could be achieved through non-invasive imaging modalities. These agents are currently being tested in pre-clinical GBM tumour models to determine their safety and effectiveness using PET/CT imaging.
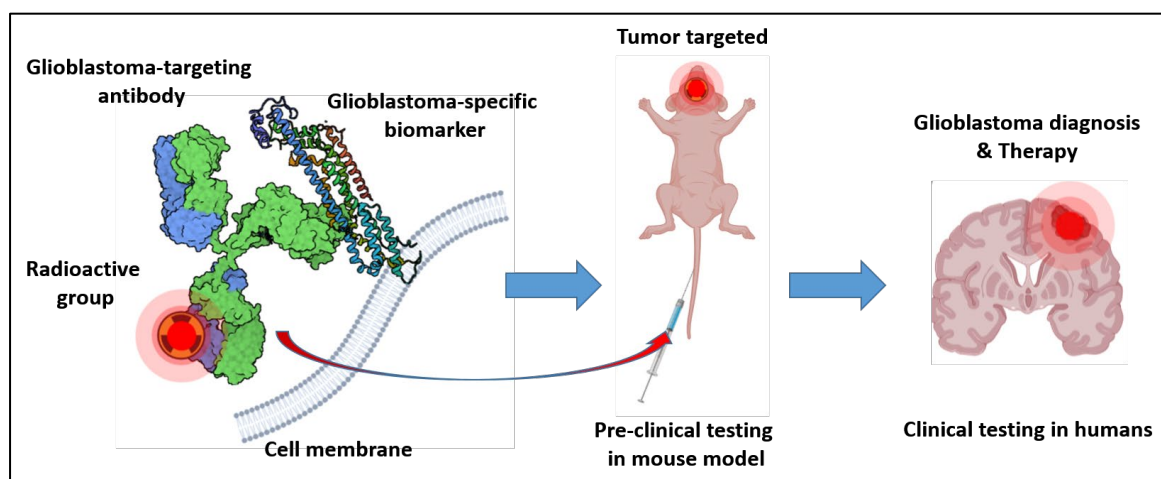
Figure 1: Novel antibody-based radio-immunotherapy for glioblastoma diagnosis and treatment

A versatile bis(thiosemicarbazone) macrocyclic chelator for use in radiopharmaceuticals

*Melyssa L. Grieve[1], Patrick R. W. K. Davey[1], Craig M. Forsyth[1], Brett M. Paterson [2]*

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Developing radiopharmaceuticals for imaging and/or therapeutic applications using radiometals requires chelators that can generate stable and inert complexes. Chelators that can provide a suitable coordination environment for a range of radiometals with different applications are particularly useful.[1] Thiosemicarbazone functional groups are versatile N,S donors that can coordinate metal ions as neutral or anionic ligands, with the resulting complexes displaying diverse coordination chemistry. N-heterocyclic thiosemicarbazones have been investigated for their pharmacological properties, which have shown that the metal complexes can display bioactivities which differ from those of either the ligand or the metal ion.[2,3]

A new bis(thiosemicarbazone) macrocyclic chelator has been synthesised that has versatile coordination chemistry, chelating a wide range of metal ions.[4] The versatility of the ligand results in both 6- and 8-coordinate complexes depending on the metal ion. The first-row transition metals Mn^{2+}, Co^{2+} and Zn^{2+} generate 6-coordinate complexes with distorted octahedral geometry in the solid state (Figure 1).[4] Density functional theory calculations indicated that the relative energies of the diastereomers are within 10 kJ mol^{-1}. Magnetic susceptibility of the complexes indicated that both the Mn^{2+} and Co^{2+} ions are high-spin. The In^{3+} ion generates complexes with both 6- and 8-coordination numbers under certain conditions as seen by ^{1}H NMR. The ligand was radiolabelled with the positron emitting isotope gallium-68 which produced a single species in high radiochemical purity (>95%) at 90 °C for 10 min. As a result, this chelator has demonstrated the potential for use in a variety of radiopharmaceutical applications.

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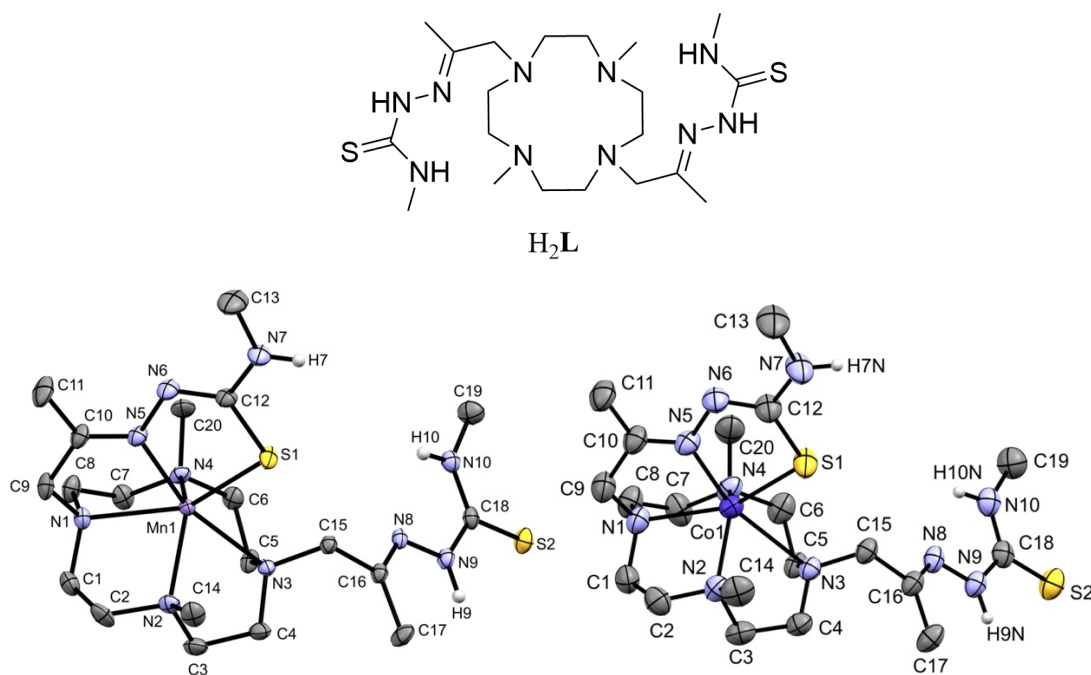
Figure 1. The structure of the bis(thiosemicarbazone) macrocyclic chelator (top), the x-ray crystallographic structure with Mn^{+2} (bottom left) and Co^{2+} (bottom right).

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ORAL

Understanding acquired immunity to polymer nanomedicines – management strategies and potential benefits

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The antibody mediated clearance of synthetic polymers presents a number of unmet challenges to the field of nanomedicine. Despite being widely reported, usually in response to some nanomedicines featuring the synthetic polymer poly(ethylene glycol) (PEG) in both preclinical and clinical settings[1], strategies to address this phenomenon have been sparingly investigated. It has long been thought that the development of synthetic materials with similar properties to PEG (but different chemical identities) can be used to overcome these obstacles, however it is currently unknown if this is the case in practice as current studies have utilised models that do not accurately reflect how nanomedicines are employed in the clinic.

In this talk a rational, materials-driven approach to overcoming acquired immunity against synthetic polymers will be demonstrated, revealing that altering the hydrophilic polymer identity used in a model nanomedicine is capable of restoring a naïve equivalent biodistribution in immunologically-relevant animal models, as assessed by a combination of ^{89}Zr PET-CT and cold immunoassay formats (**Figure 1**).

A secondary challenge surrounding the development of PEG alternatives is the lack of clear data surrounding their interaction with components of the adaptive immune system, and lack of studies investigating their ability to induce humoral antibody responses. To this end, a series of novel anti-polymer monoclonal antibodies have been successfully isolated, and pertinent information such as their typical binding kinetics and specificity will be discussed in order to further inform on the potential clinical applicability of these materials as PEG alternatives.
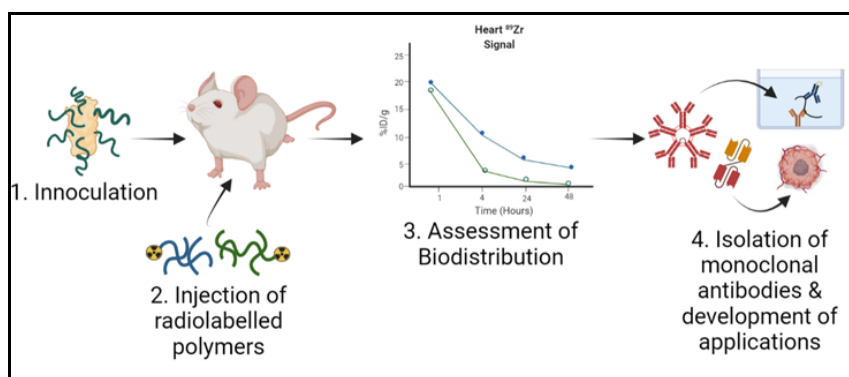
Figure 1. Schematic overview of investigation of antibody mediated clearance of different polymer systems and the development of new applications from specific anti-polymer monoclonal antibodies.

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ORAL

Novel Aryl Hydrocarbon Receptor PET Imaging Agents for Glioma

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Glioma is a deadly disease with high mortality rates; there has been little improvement in the 5-year survival rate over the past 30 years. The aryl hydrocarbon receptor (AhR) is a transcription factor that regulates a myriad of genes involved in immune response modulation and tumorigenesis. The AhR is considered as an attractive drug target and studies have shown that its activation by small molecules can impact innate and adaptive immunity and prevent AhR-mediated tumor promotion in several cancer types. In human gliomas, the enzymes tryptophan-2,3-dioxygenase (TDO) and indoleamine 2,3-dioxygenase (IDO) constitutively generate kynurenine, leading to kynurenine-AhR suppression of anti-tumour immune responses and reduced activity leads to increased immunosuppressive regulatory T-cells and improves patient prognosis. As we approach a paradigm shift towards individualised medicine, positron emission tomography (PET) imaging tracers are an important method of identifying therapeutic molecular targets. To date, there are no PET tracers specific towards glioma-specific antigens or receptors. Herein we report the synthesis of AhR targeting radioligands _via_ the copper-mediated radiofluorination of small molecules containing benzimidazoisoquinolines and indole scaffolds. Manual radiolabelling experiments with the corresponding boronate ester precursors and $Cu(OTf)_2py_4$ shows that this method of radiofluorination is sufficiently robust to fluorinate electron-rich heterocyclic substrates like benzimidazoisoquinolines (RCC $39.9\pm6.7\%$) and is also able to tolerate unprotected indoles (RCC $54.2\pm27.9\%$) in manual labelling experiments. Automation of the radiosynthesis afforded (6-[^{18}F]fluoro-1_H_-indol-3-yl)(6-(trifluoromethyl)pyridin-2-yl)methanone (**[^{18}F]TX19**) with RCY of $9.06\pm0.9\%$ (non-decay-corrected) and molar activity of 60.0 ± 17.7 GBq/µmol at the end of synthesis (n=5). Further work needs to be done to automate the benzimidazoisoquinoline tracers and validate these AhR targeting radiotrafcers _in vitro_ and _in vivo_.

ORAL

The isolated chicken ASIC1a thumb domain (ATD-c1a) retains the structure and ligand binding properties of the full length ASIC1a

Biswa Prasanna Mishra[1], Ben Cristofori-Armstrong[1], Elena-Laura Budusan[2], Xinying Jia[1], Yanni Chin[1], Lachlan Rash[2], Mehdi Mobli[1]

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Acid-sensing ion channels (ASICs) are potential novel therapeutic targets in a range of pathologies including ischemic stroke, spinal cord injury, chronic pain, multiple sclerosis, rheumatoid arthritis, and migraine.[1] Nonetheless, there are no drugs available on the market that target these channels selectively. A hurdle in the development of selective ASIC drugs is the lack of adequate and reliable structural data of these pH sensitive ion channels in their multiple conformational and protonation states. All ASIC structural data available to date are from a single isoform (1a) derived using X-ray crystallography and electron microscopy. This study proposes to study the thumb domain of the ASIC channel in isolation using solution state techniques. The ASIC thumb domain (ATD) is a cysteine-rich domain, and its two α-helices are part of the "acidic pocket" that collapses upon extracellular acidification (Fig.1a).[2] It is also known to be an important ligand binding domain, acting as the receptor site for four highly potent and selective venom peptide toxins PcTx1, MitTx, and mambalgins-1 and 2.[3-6] Here, we present the first structure of the isolated thumb domain of the chicken ASIC1a (ATD-c1a) in solution determined by nuclear magnetic resonance (NMR) spectroscopy (Fig. 1b). Using a combination of NMR and ITC measurements, we characterise the structural and thermodynamic details of ligand binding of the ATD using endogenous and exogenous ligands. Ligand binding is performed across a range of pH values. This allows us to provide atomic resolution information on the pK_a of protonatable residues, identifying residues that are important for the pH sensitivity of ligand binding. This information will help researchers screen for, design and optimise ASIC modulator as novel therapeutic lead compounds.
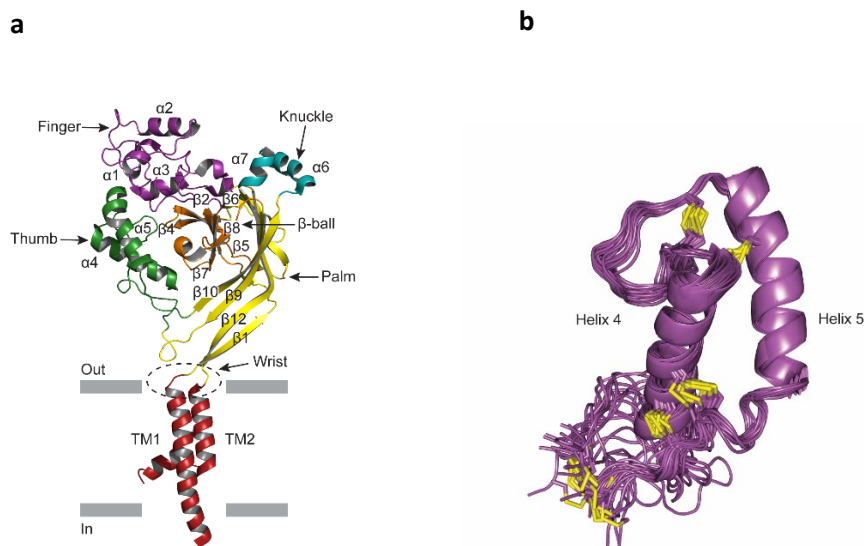
Fig. 1 (a). Location of the thumb domain in a ASIC1a channel monomer, **(b).** Ensemble of 20 structures of ATD-c1a solved using solution NMR spectroscopy. The disulfide bond connectivities have showed in yellow.

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ORAL

New ^{64}Cu-labeled CXCR4-targeting ligands for theranostic applications

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Theranostics is a stratified approach to medicine which ultimately aims to improve patient outcomes. Targeted Copper Theranostics (TCTs) using copper radionuclides, ^{64}Cu ($t_{1/2}$ = 12.7 hours, β^+ = 17.4%) for imaging and ^{67}Cu ($t_{1/2}$ = 61.9 h, β^- = 100%, E_{max} = 562 keV) for therapy, offer significant advantages in the development of next generation theranostics. The sarcophagine (SAR) chelator is an ideal choice for developing theranostic agents as they form extremely stable copper complexes *in vivo*. To demonstrate the utility of the ^{64}Cu/^{67}Cu radionuclide pair in oncology theranostics, two 64Cu-labeled CXCR4-targeting SAR constructs, [^{64}Cu]Cu-SAR-CXCR4 and its PEGylated analogue [^{64}Cu]Cu-SAR-PEG-CXCR4, were developed for preclinical PET imaging of CXCR4, and can be translated into ^{67}Cu-labeled therapeutics.

Two constructs containing the high affinity CXCR4-targeting pentixather scaffold and the SAR-derived chelator, MeCOSar, were synthesised. Cell binding assays were performed in SK-BR-3 adenocarcinoma cells. The metabolic and transmetalation stabilities of the tracers were evaluated in mouse serum and excess EDTA respectively, and lipophilicity determined via shake-flask assays. Small-animal PET-CT and biodistribution studies at different time-points were performed in xenografts of Daudi lymphoma in mice.

The radiotracers were efficiently radiolabeled with high radiochemical yields (>95%) and purities (>98%), and demonstrated high metabolic and transmetalation stabilities. [^{64}Cu]Cu-SAR-CXCR4 displayed a more efficient CXCR4-binding compared to the PEGylated tracer *in vitro*. PET-CT imaging and biodistribution studies showed tumour targeting by both radiotracers over 24 h p.i.. [^{64}Cu]Cu-SAR-CXCR4 demonstrated superior tumour retention over 24 hours, leading to comparatively higher tumour-to-organ ratios at all time-points. The higher non-specific accumulation of [^{64}Cu]Cu-SAR-CXCR4 in the liver and spleen compared to [^{64}Cu]Cu-SAR-PEG-CXCR4 could be attributed to its enhanced lipophilicity.

In conclusion, We have successfully prepared two CXCR4-targeting radiotracers showing efficient radiolabeling with ^{64}Cu and high stabilities in serum and with EDTA challenge. The compounds showed tumor targeting and retention resulting in high resolution PET-CT images, and may make promising tracers for future application in endoradiotherapy with ^{67}Cu.

ORAL

Modeling resistance to PRRT in NET

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Neuroendocrine tumour (NET) is a rare and complex cancer that is poorly responsive to conventional treatments.[1] Peptide Receptor Radionuclide Therapy (PRRT) using Lu-177 DOTA-octreotate (LuTate) is a targeted treatment for NET, which delivers high doses of radiation directly to tumours via somatostatin receptor type 2 (SSTR2), thus exhibiting minimal off-target effects.[2] PRRT it is now a routine treatment for NET, with patients generally showing a good initial response, although it is rarely curative.[2] While some patients are inherently resistant to PRRT, more commonly patients develop resistance over multiple cycles of therapy, thereby reducing effectiveness and durability.[3] Developed resistance is most frequently attributed to de-differentiation of the tumour and subsequent loss of SSTR2. However, there is a subset of patients that develop resistance without SSRT2 loss,[3] which we hypothesise is attributed to enhanced DNA damage repair pathways, a more general radiation resistance phenotype.
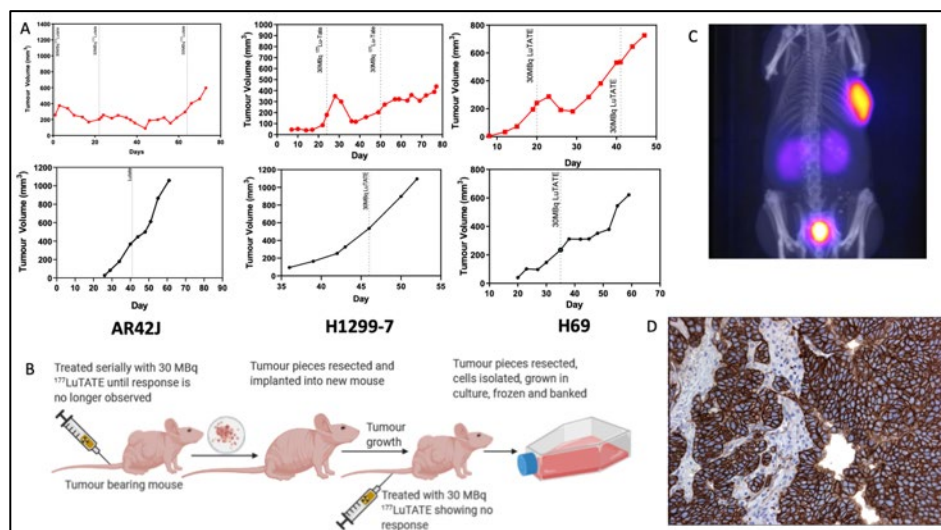
Figure 1. a) NETs were serially treated with 30 MBq of LuTate, reimplanted, and re-treated to confirm resistance. b) Tumour pieces were then resected, cells isolated, and grown in culture to generate a resistant cell line. Retention of SSTR2 expression in LuTate-resistant xenografts was confirmed by c) [68]GaTate PET imaging and d) IHC.

Whilst multiple animal NET models exist that mimic inherent resistance to PRRT, none model developed resistance without target loss. We describe a method of generating models of 'developed resistance' in three NET cell lines, and show SSTR2 expression is retained via in vivo imaging and immunohistochemistry (IHC).

NET bearing mice of three different xenographs (AR42J, H1299-7 and H69) were treated with 30 MBq of LuTate causing regression in tumour growth. When each tumour started to show regrowth after this first treatment, they were retreated with another 30 MBq of LuTate. If the tumour continued to grow after this second challenge, they were harvested and reimplanted into a new host. Resistance was confirmed when a third 30 MBq LuTate challenge demonstrated no regression (**Fig.1a**). The resulting tumours were resected, cells isolated, and grown in culture to establish a cell line described as 'LuTate-resistant' (**Fig.1b**). To confirm SSTR2 expression, these 'LuTate-resistant' lines were subcutaneously inoculated into new mice and either imaged in vivo with [68]GaTATE PET (**Fig.1c**) or characterised with immunohistochemistry (IHC) (**Fig.1d**).

Our SSTR2 expressing models of developed resistance mimic the described resistance phenotype seen in a subset of patients, and we are now utilising them to investigate mechanisms of resistance in vivo, and in turn assessing treatment options for overcoming this resistance in patients.

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ORAL

# PET arterial input function estimation using machine learning

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Dynamic positron emission tomography (PET) scans are often converted to parametric images measuring tracer exchange between tissue compartments. The kinetic parameters aid in clinical diagnosis, therapy monitoring and treatment planning across a multitude of diseases and disorders. An estimate of the arterial input function (AIF) is necessary to be able to predict kinetic parameters. Conventionally, the AIF is estimated using invasive blood sampling for the plasma tracer concentration, and existing non-invasive methods, including image derived input functions (using different imaging modalities) and population based AIFs, have shortcomings [1]. This gap provides an opportunity of exploring the estimation of the subject specific AIF from PET scans alone using machine learning. The dPETSTEP simulator and the Feng function (i.e., AIF) was used to create dynamic PET images based on a ground truth AIF [2]. A total of 28 dynamic images over a 60 minute period were simulated [3]. The machine learning pipeline in Figure 1 was used to recover the AIF. Specifically, dynamic PET images were assumed to be a Gaussian mixture of tissue time activity and AIF. A patch of size 16×16 was chosen arbitrarily, see Figure 1, and encoded via the variational autoencoder (VAE). Encoded patches were used as input to the long-short term memory (LSTM) components followed by the application of a mixture density network (MDN). Lastly, parameters for each time step (the mean - μ, standard deviation – σ and weight - π) formed by the mixture of tissue activity (μ_t, σ_t, and π_t) and the AIF (μ_a, σ_a, and π_a) were estimated. The weights (π_a) over the time steps provide the AIF estimate. In Figure 2 examples of the estimated AIF are compared against the actual AIF. The average of relative root-mean-squared error percentage between the actual and estimated AIF's for five different simulated subjects were 4.57 %, which is acceptable for accurate estimation of parametric mapping. The study was conducted using simulated data demonstrating the applicability of machine learning methods for accurate AIF estimation. The method may find utility in predicting the AIF non-invasively from clinical PET data.
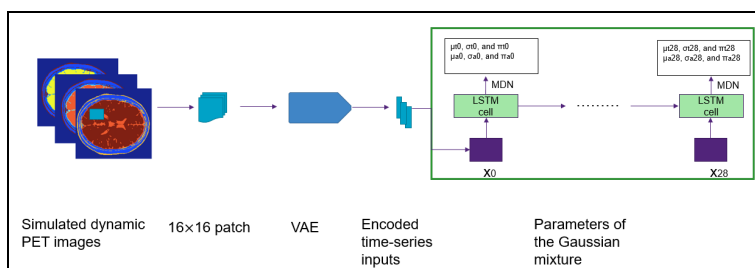
Figure 1. Pipeline for direct estimation of AIF.
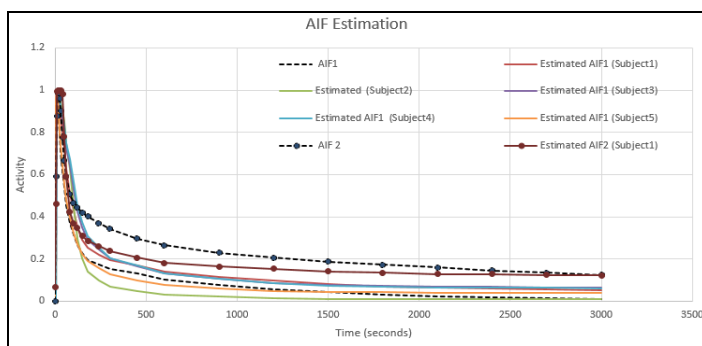
Figure 2. Plot for actual and estimated AIF

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POSTER

Development of novel ^{18}F-PET imaging probes for mTOR signaling in Focal cortical Dysplasia type II

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The journey to the familiar cortex presents a large window of vulnerability to disease. Now termed malformations of cortical development (MCDs), these diseases are a heterogeneous group of disorders resulting in abnormalities in brain size, neuronal organisation and gyrification[1]. Focal cortical dysplasia (FCD) is one such disorder and causes disorganization of the cerebral cortex as a consequence of somatic mutations in genes governing neuronal development[2]. This manifests as lesions with architectural abnormalities, namely cortical dyslamination or the presence of abnormal cell morphology within the cortex. Being a heterogeneous disorder, FCD presents a varied pathology giving rise to various subtypes (types I-III) which exist as distinct genetic entities[3]. FCD is often drug resistant and surgical resection is limited by the variability of lesion localisation across subtypes. Consequently, the pathological features of FCD can evade detection by radiological techniques, thus hampering imaging prospects in presurgical stages[4].

In recent years, the role of the mammalian target of rapamycin (mTOR) pathway has emerged as a key player in cortical development. As such, it is understood that aberrant mTOR activity is linked to developmental malformations and epileptogenesis. Constitutive mTOR activity is common feature in a subset of MCDs, namely tuberous sclerosis complex (TSC), hemimegaencephaly (HME) and FCD-II[5]. With limitations in sensitivity for extratemporal epileptic foci using current PET imaging tools, we consider mTOR as a potentially new imaging target for diagnosing FCD-II and propose the development of a novel ^{18}F-PET imaging probe targeting hyperactive mTOR in the brain.

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POSTER

Protein structure analysis by EPR spectroscopy using GD^{3+} spin labelling

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Protein is an integral part of life and understanding the protein structure has numerous uses in pharmaceutical industry. In a crowded cellular environment, most proteins function at low concentrations, which can impact their structure and dynamics. The proposed project aims to develop new EPR methodologies for studying protein structure and dynamics at low concentrations and under in-cell circumstances. The methods to be used are paramagnetic labelling of proteins, EPR spectroscopy to measure distance between the Gd^{3+} labels followed by developing new computational algorithms to interpret distance distribution data from EPR spectroscopy for protein structure modelling.

The first step of the project is synthesis of Gd^{3+} tags. Gd^{3+}, being one of the best paramagnetic labels, is used for spin labelling because it performs best at W-band and is highly sensitive compared to nitroxide labels which are limited to Q band. Gd^{3+} tags are conjugated with the non-canonical amino acids(ncAAs) that will be genetically encoded at specific sites of the protein. The ncAAs to be used are Cyanopyridylalanines that will be genetically encoded by using aminoacyl-tRNA synthetases. The nitrile-aminothiol (NAT) click reaction between the aminothiol group of Gd^{3+} tags and nitrile group of Cyanopyridylalanines will used for site specific labelling in response to an amber stop codon.

NOTE: I will be poster presenting just the synthesis of Gd^{3+} tags along with the 3D-model of the unnatural amino acids used for site specific tagging.

POSTER

Targeted Treatment of Brain Tumours Using a Multi-drug Loaded, Relapse-Resistant Polymeric Theranostic

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Glioblastoma (GBM) is the most malignant of brain cancers, with a median survival of less than 15 months from diagnosis. Treatment is complicated by GBM cancer cells invading extensively into surrounding brain tissue and having a high rate of proliferation. Current standard clinical intervention is surgery combined with chemo-(Temozolomide, TMZ) and radiotherapy. However, resistance to TMZ is prevelant due to high expression of DNA repair protein O^6 methylguanine DNA methyltransferase (MGMT), which can partially restore the activity of TMZ alkylated genes and induce tumour recurrence.[1] The combination of the MGMT inhibitor, O^6 Benzylguanine (O^6BG) with alkylation drugs in resistant GBM cell lines has shown benefit in tumour bearing animal models.[2,3] However, the therapeutic effect of TMZ with O^6BG in clinical trials was less pronounced, mainly due to the reduced dosage of TMZ used as a result of severe side effects for this combination treatment.[4,5] Hence, an effective drug delivery system that can improve the targeted release of TMZ and O^6BG at the nidus and maintain the dosage of TMZ in treatment could increase the therapeutic effect of this drug combination to TMZ-resistant patients.

This research aims to generate such an effective platform by linking TMZ and O^6BG to a hyperbranched polymeric backbone functionalized with bispecific antibodies (αPEG-αEphA2 BsAb) that bind to the tumour-associated antigen, Ephrin receptor A2 (EphA2) to increase specific tumour accumulation (**Fig. 1**). This hybrid platform facilitates targeted drug delivery through covalently linking TMZ to the polymer backbone, and utilizing an acid-sensitive bond for site-selective O^6BG release. In addition, the engineered system includes a dye (Cy5) for monitoring of the delivery system in both cell and animal studies, forming a synergistic theranostic platform for GBM treatment. Studies on resistant human GBM cell lines showed the improved accumulation and therapeutic efficacy of the antibody conjugated dual-drug platforms. Biodistribution and tumour accumulation of this nanomedicine in preclinical GBM models will also be presented.
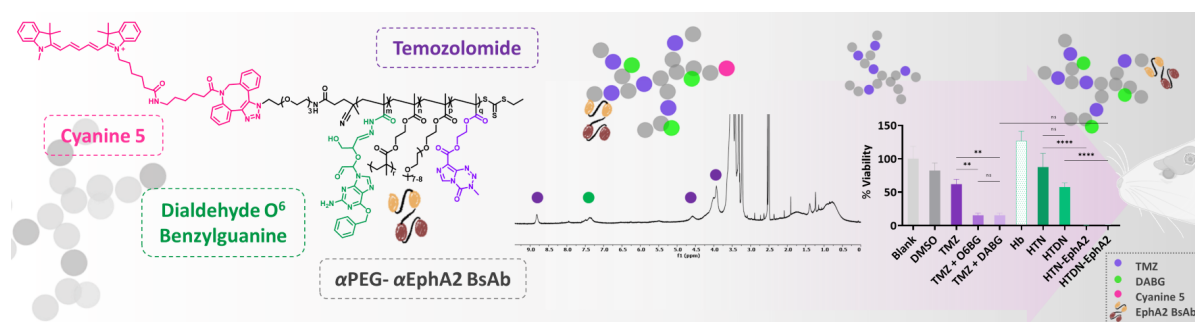

Figure 1.

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POSTER

Unsupervised learning for epileptogenic lesion detection on 7T brain MRI in patients with focal epilepsy

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Introduction: In drug resistant epilepsy, seizure freedom can only be obtained through seizure onset tissue resection. Qualitative radiological images and clinical findings are used to define regions of epileptic foci, such as in focal cortical dysplasia. Delineation using current approaches can be incomplete, warranting the development of automated methods of lesion identification using machine learning approaches. Data have been acquired using epilepsy series protocol on 7T Siemens MRI scanners. The acquired data consists of two study groups healthy control and patients with focal epilepsy. This study proposed a machine learning framework to identify the subtle epileptogenic lesions for MRI-negative patients. The proposed method takes a patient brain MRI and produces a tissue map or score map identifying locations of dysplasia.

Methods: We have proposed a deep learning approach-based anomaly detection framework for epileptogenic lesion detection on brain MRI. The proposed framework consists of two basic building blocks, unsupervised learning-based feature representation and anomaly detection. The first block or feature extraction block of the proposed framework will extract a latent representation of the patch as a feature representing the centre voxel of the given 2D patch. In the second block or anomaly detection block, we will train one class classifier(oc-SVM) model using the features extracted from all the healthy control subjects for a particular voxel. Afterward, extracted features from all the patient data are used for testing the oc-SVM model to check whether the voxel is normal or abnormal. If the trained model predicts a voxel as an outlier indicates that the voxel is abnormal. After post-processing the predicted values from the oc-SVM model, the final lesion map will be generated. The proposed model was trained and tested on clinically widely used MP2RAGE and FLAIR sequences and all the images are co-registered to MNI 1mm template. We have used 62 healthy control subjects and five patients' images to train and test the model, respectively. We have evaluated our proposed model performance by comparing the results with the ground truth MRI. The expert neurologist and radiologist define the ground truth.

Results: The performance of the proposed model is reported in Figure-1. The first row of the image displays the focal epilepsy patients images with actual lesions and the second row displays the overlapped images of the model and patient image to correlate the identified and actual lesions. We have tested the proposed model on five epilepsy patients' images. The epileptogenic lesions in these test images have a variety of sizes and properties. Our model identified the lesion for four patients' P04, P09, P10, and P07 images and could not detect it for one patient's P05 image. In the visual inspection of the P05 image, we can observe that the lesion is significantly subtle.
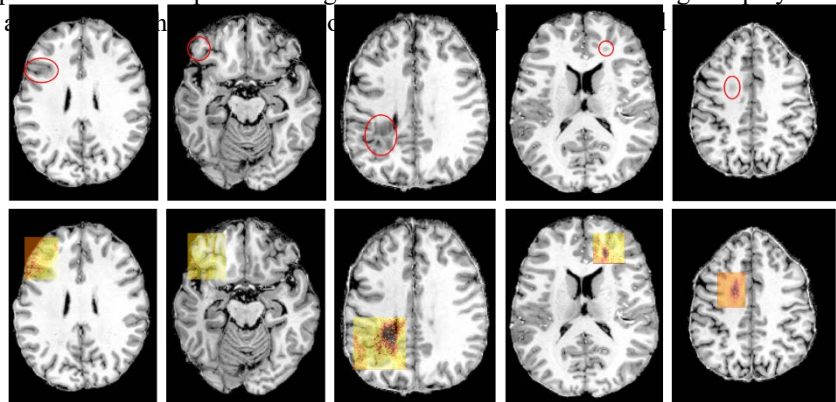

Figure 1 : Output of the proposed model for epileptogenic lesion detection. From left to right, the patient is P04, P05, P09, P10, and P07.

Conclusion: The proposed model exhibits significant performance in subtle lesion detection. The proposed model identified the lesion for four patients out of five. Currently, the model has executed only a part of the brain instead of the whole brain to avoid the enormous computation time. This work leads to future investigations on the influence of different MRI contrasts as input to the machine learning framework and how genomics data can be utilized in subtle lesion detection for MRI-negative focal epilepsy patients.

POSTER

The effect of avidity on nanobody-based formats for theranostics

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Biological medicines have received significant attention in the last several decades due to their versatility and effectiveness across a variety of diagnostic and treatment applications. Like other antibody fragments, Nbs can be engineered to be multi-specific and multivalent to improve their binding capabilities through functional affinity (avidity). Together with their small mass and rapid clearance, the high specificity of Nbs is a particularly attractive feature for diagnostic imaging and radioimmunotherapy in oncology. There is a frequent assumption that engineering antibodies or fragments (e.g. Nbs) in this way will be synonymous with greater diagnostic or therapeutic efficacy, but some studies have shown that this is not the case *in vivo*. In these studies, reduced tumour accumulation and retention was observed as the complexity and size of targeting ligands increase, however there is no conclusive evidence to suggest this. Broadly, this thesis therefore aims to elucidate the effect of increasing avidity through engineered Nb formats and how it affects the biodistribution and penetration into solid tumours. This will be achieved using Nbs developed against clinically relevant target molecules, HER2 and HER3 which are selectively over-expressed in various solid cancers due to gene amplification and/or microenvironment adaptation. HER2-3 dimers send the most potent oncogenic signals to the cell, making them ideal targets for cancer immunotherapeutics. In the clinical setting, monotherapy with mAbs are not effective because of the redundancy within the EGFR family leading to drug resistance, and HER2-3 combination therapy is highly toxic. Using Nbs to target an external payload to these receptors, rather than trying to inhibit their signaling, may allow us to take advantage of their tumour-specific over-expression without the toxicity of classical molecular inhibitors.

Figure 1. Low affinity nanobodies with high dissociation rates exhibit higher tumour penetration compared to nanobodies that are high affinity with low dissociation rates.
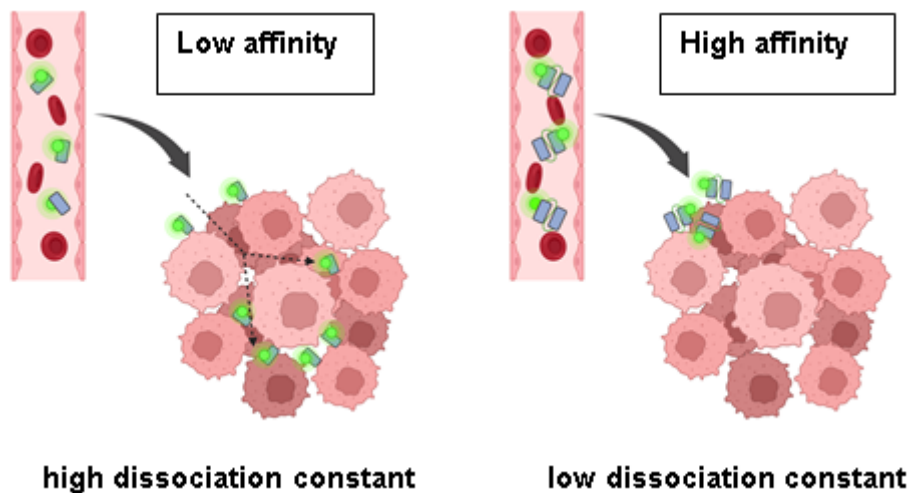
POSTER

A new tool for facilitating structural characterisation of peptides containing non-canonical amino acid (ncAA) by standard NMR methods

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Peptides containing ncAAs encompass a large pool of biologically active molecules including a number of important drugs. The significant structural and functional diversity found in these peptides has presented challenges to their modelling using standard protein and peptide NMR structure determination software, including the popular CYANA, XPLOR, and ARIA packages. In these cases, the presence of amino acids outside the genetic code requires additional input files not readily available. We have expanded the publicly available Automated Topology Builder (ATB, atb.uq.edu.au) to generate input files for standard NMR packages[1]. We have focused particularly on CYANA, a torsion angle dynamics software package, which has the most complex input requirements[2]. Using this tool vastly expands the utility of NMR structural characterisation of peptides and proteins that fall outside the 22 natural amino acids. In addition, this method will also reliably generate distance restraints required for the cross-linking of sidechains, which are often present in ncAA-containing peptides. With this streamlined approach we hope to improve the utility of NMR structural characterisation for the analysis of complex peptides, thereby improving our understanding of this large class of biologically active proteins and peptides.

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POSTER

Development of polymeric micelles-based nanoparticles for monitoring cytokine release *in vivo* via DNA aptamer

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Breast cancer (BC) is the most frequent malignancy among women worldwide, mortality rates have remained high over the past two decades. Current treatments such as surgery, chemotherapy, radiotherapy, and (neo)adjuvant combination therapy are available now and have yielded results, quality of care can be improved by reducing adverse effects exhibited in each treatment strategy. For example, patients undergoing radiotherapy (RT) can experience radiotherapy-induced fatigue (RIF) during treatments. Some investigations showed an association between fatigue and circulating inflammatory cytokine levels. Interestingly, proinflammatory cytokine Interferon-gamma (IFN-γ) secretion has been reported to increase in the serum of BC patients, while other research indicates the IFN-γ expression was not affected by RT. This divergent result is caused by the transient, dynamic cytokine secretion process with a limited quantity, and common detection methods such as ELISA, are less sensitive, conducted ex vivo and measure the process indirectly. Thus, to develop an *in vivo* delivery platform for monitoring IFN-γ release dynamically is necessary to overcome these limitations.

Polymeric nanoparticles are promising delivery vehicles with the advantages of easy modification and facile preparation. Therefore, a polymeric micelles-based delivery platform conjugated with anti-PEG/anti-TAG72 bispecific antibody and IFN-γ specific DNA aptamer on the surface, is favourable to monitor the cytokine release through fluorescence-based signal transduction. To achieve this, conjugated micelles are first be delivered to breast cancer cell surface. Then the second molecule containing a Lu^{177} radioisotope is injected and allowed to bind with nanoparticles at the tumor sites via iEDDA (Tetrazine and trans–cyclooctene–tetrazine) click chemistry. Finally, the in vivo dynamic biodistribution is revealed through imaging techniques such as Positron emission tomography (PET-CT) or Single-photon emission computed tomography (SPECT). Overall, the primary purpose of this project is to establish a novel method for detecting radiotherapy-stimulated cytokine modulation in vivo to evaluate the effect of delivery and provide a better understanding of the underlying mechanism for fatigue during radiotherapy at the biomolecular level. (Please see the following scheme)
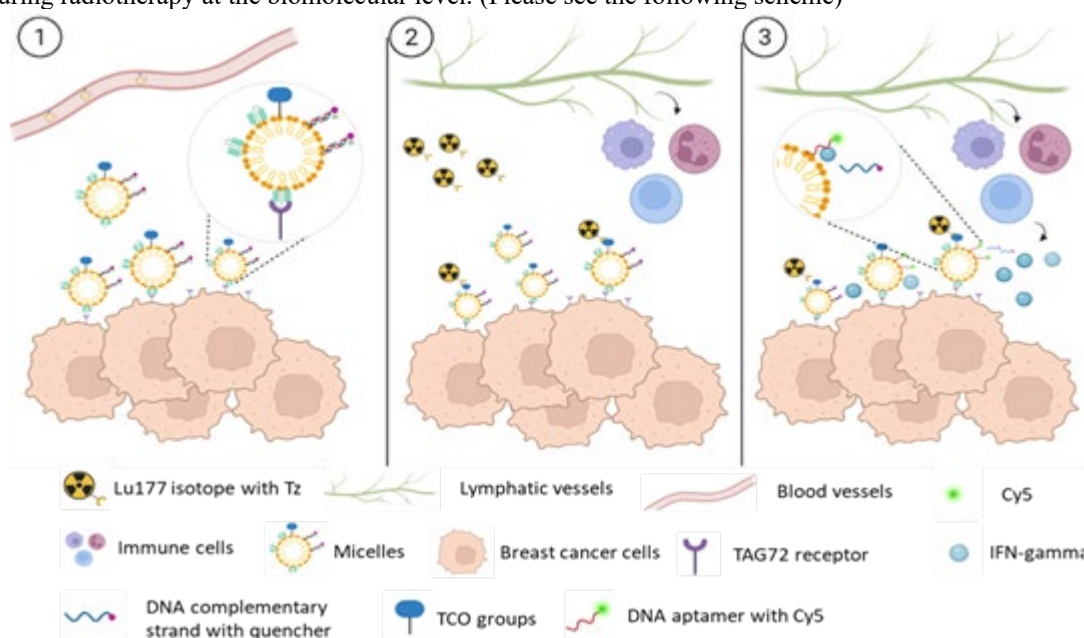
Scheme 1. The scheme of the project, by which shown in 1, polymeric micelles are injected into mice firstly via blood vessel, bispecific antibody make the vehicle target to the tumor surface and bind with TAG72 receptor. Then in 2 a second molecule Lu^{177} isotope chelated to a tetrazine functionalized DOTA is injected, immune cells via lymphatic vessels are activated and release IFN-γ. Theoretically the two molecules will bind together and the released IFN-γ will react with Cy5 modified DNA aptamer due to high affinity, which shown in 3.

POSTER

Targeted delivery of immune-boosting peptides using polymeric nanoparticles to manage lung cancer

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Lung cancer is one of four leading cancers causing a high death rate globally. Approximately 85% of lung cancers are non-small-cell lung cancers (NSCLCs), and the average 5-year survival is 15-20%. The dominant factors contributing to the high death rate for NSCLCs are: 1) Advanced stage diagnosis with metastatic disease for most patients, 2) Heterogenicity and low antigenicity of NSCLCs. Thus, novel therapeutic options for long-term treatment of NSCLCs are necessary for patients who have poor prognosis and immune-therapeutic outcomes. InterK Peptide Therapeutics have developed peptides with immune-boosting functions, re-invigorating exhausted T cells and potentially serving as a novel therapeutic agent against human lung cancer. The ability to produce a site-specific response will be the major hurdle to translate these therapeutic peptides for clinical use. Rapid growth in nanotechnology provides an opportunity to improve the pharmacokinetic and pharmacodynamic behaviour of drugs due to their physical and chemical characteristics. Thus, nanocarriers are the ideal platform for the targeted delivery of InterK peptide and increasing their concentration at the tumour site. This project investigates the targeted delivery of InterK's novel immune-boosting peptides to the tumour microenvironm   bution and tumour suppre
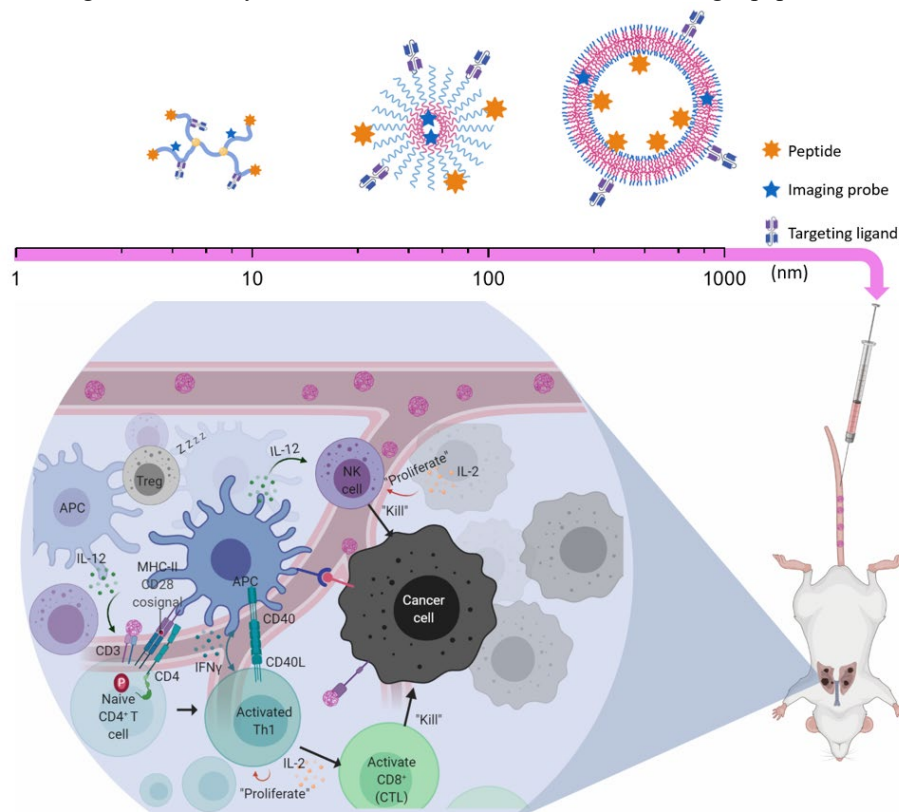

Figure 1 Schematic design of nanocarriers (NPs and cells are not drawn to scale)

POSTER

Novel Bis(thiosemicarbazonato) Technetium-99m Nitrido Complexes for Prostate Cancer Imaging

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Prostate cancer is the second most commonly diagnosed cancer and the fifth-highest cause of cancer death in men worldwide.[1] Recently, nuclear imaging has been investigated to improve the diagnostic accuracy and to allow for earlier detection of prostate cancer.[2] This can be achieved by attachment of a radionuclide to a molecule that targets a receptor overexpressed in prostate cancer cells, known as the prostate specific membrane antigen (PSMA).[3] In this work, a series of bis(thiosemicarbazone) (BTSC) chelators were synthesised and attached to PSMA binding motifs using both solution-phase and solid-phase chemistry. The linker was altered to explore the hydrophilic/lipophilic balance of the chelators and the addition of amino acids to increase binding of the radiopharmaceuticals to the PSMA channel. The chelators were radiolabelled with the technetium-99m nitrido core, $[^{99m}Tc][TcN]^{2+}$, which has previously shown promising results with BTSC chelators.[4] The radiolabelling occurred in a simple one-pot synthesis at 85°C for 10 min and all chelators achieved a high radiochemical purity (> 99%). The hydrophilic/lipophilic balance was explored by reverse-phase HPLC and distribution coefficient (logD) that showed all complexes were hydrophilic with negative logD values. Preliminary *in vitro* stability studies, undertaken in the presence of a competing ligand (cysteine) and in human serum at 37°C, were very promising indicating that the complex was kinetically inert under both conditions over 24 h. This suggests the potential for these radiopharmaceuticals to be used as imaging agents for prostate cancer and further *in vivo* and *in vitro* studies will be undertaken to explore this.
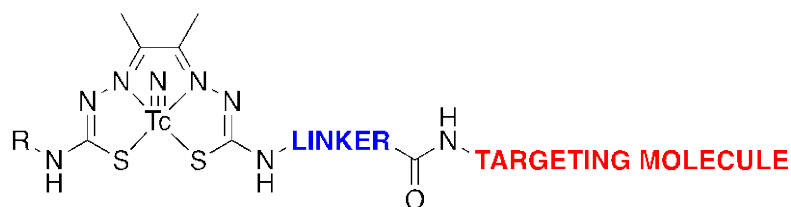
Figure 1. General structure of $[^{99m}Tc][TcN(BTSC)]$ for prostate cancer imaging.

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POSTER

Looks like a spider but acts like a scorpion — A bivalent remipede toxin promotes calcium release via ryanodine receptor activation

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Abstract

Multivalent ligands of ion channels have proven to be both very rare and highly valuable in yielding unique insights into channel structure and pharmacology. Here, we describe a bivalent peptide from the venom of Xibalanus tulumensis, an arthropod from the enigmatic class Remipedia, that causes persistent calcium release by activation of ion channels involved in muscle contraction. The high-resolution solution structure of Xibalbin3-Xt3a reveals a tandem repeat arrangement of inhibitor-cysteine knot (ICK) domains previously only found in spider venoms. The individual repeats of Xt3a share sequence similarity with a family of scorpion toxins that target ryanodine receptors (RyR). Single-channel electrophysiology and quantification of released Ca^{2+} stores within skinned muscle fibers confirm Xt3a as the first bivalent RyR modulator. Our results reveal convergent evolution of RyR targeting in remipede and scorpion venoms, while the tandem-ICK repeat architecture is the first evidence of this evolutionary innovation outside spiders.

POSTER

Dynamic FDG-PET shortened acquisition protocols determined using machine learning

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Introduction: PET kinetic modelling methods were developed several decades ago. Despite the contribution of additional information relevant to disease diagnosis, prognosis and treatment planning they are yet to be used widely in the clinic. The low uptake is the long 60-minute acquisition time required to estimate kinetic parameters from dynamic PET (dPET) images. Shortened dPET protocols may facilitate clinical adoption of kinetic modelling. In ^{18}F-FDG PET the time activity curve (TAC) generated for a volume of interest (VOI) or a voxel is used to estimate tissue specific parameters using the two tissue compartment model, namely the influx rate (K_1, ml/g/min), rate constants (k_2, k_3, 1/min) and net influx rate ($K_i = K_1 k_3 / (k_2 + k_3)$, ml/g/min). Studies have investigated how well kinetic parameters can be fitted in shortened acquisition windows using the conventional non-linear least squares (NLSS) and machine learning based approaches. However, it remains unclear which parts of the TAC can be omitted to still achieve accurate estimates of kinetic parameters. Using supervised machine learning methods, we investigated the minimum TAC sampling requirements for accurate kinetic parameter estimation.

Methods: An in-house simulation environment in MATLAB® was created to produce synthetic data for ^{18}F-FDG dPET. Four thousand TACs were generated based on the two-tissue compartment model with irreversible binding. Individual dPET TACs were simulated for 60 min (4×30s, 8×60s, 10×120s and 6×300s frames). Parameters K_1, k_2 and k_3 were selected based on values from previously reported ranges. The blood fraction in tissue, V_b, was randomly chosen in [0.02, 0.05]. Noise was introduced to each TAC using a previously described approach. Testing was performed on a digital dPET brain phantom with realistic grey-white matter kinetic parameters. Neighbourhood Component Analysis (NCA) was used to establish the sensitivity of kinetic parameter estimation to data collection time. Different TAC durations were also evaluated by systematically increasing time intervals up to 60 min. Gaussian Process Regression (GPR) appended by an autoencoder was employed to build the kinetic modelling framework. Individual TACs and the arterial input function formed the feature vectors, and corresponding kinetic parameters, K_1, k_2, k_3 and K_i were the labelled data. Goodness of fit was assessed using the R^2 value, and error in the kinetic parameter estimates was recorded.

Results: The result shows that the highest index feature weights for K_1 and k_2 occur at early time points, i.e., 0 to 5 min, and k_3 estimation requires the first 8 minutes and last 15 minutes of a 60 min imaging window. Our NCA results confirmed that the initial imaging window, 0 to 8 min, contained critical information for estimating all kinetic parameters. While the final minutes of the imaging window are essential for estimating k_3, and less critical for k_2 and K_i, and not necessary for K_1. The results confirm that the double imaging protocol [0-8 min and 55-60 min] and a single imaging window protocol [0-24 min] provide similar accuracy in estimating the kinetic parameters ($R^2 > \sim 0.90$) as the standard dPET protocol.

Conclusion: Results provided suggest that the proposed machine learning kinetic modelling framework, including GPRs and autoencoders, provides robust kinetic parameter estimates in shortened imaging windows identified using NCA.

POSTER

Understanding nanoparticle accumulation in a complex *in vitro* tumour-on-a-chip model

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Microfluidic devices for cell biology research have revolutionized the way we can analyse nanoparticles (NPs) and novel drugs over the past 20 years.[1] These devices have shown great promise in being highly tunable, high throughput systems that can be used for a wide variety of biological experiments to better understand NP interactions on the cellular level.[2] In my work, I have designed a polydimethylsiloxane (PDMS) microfluidic device with the capability to culture multicellular tumour spheroids (MCTS) that can be imaged and analysed in real time to gain detailed understanding of NP interactions at the tumour site.[2] These MCTS are designed to incorporate cancer (SKOV-3), fibroblast (NIH/3T3), and macrophage (RAW264.7) cell lines to create a complex 3D model that better replicates the tumour microenvironment. Flow rate and sheer stresses can be appropriately scaled in this tumour-on-a-chip model, allowing for extravasation and accumulation of NPs to be understood through real-time imaging in a physiologically relevant environment. This work will look at a library of NPs with various surface chemistries and characteristics to gain insight into what properties allow greater accumulation and penetration into the tumour site. Through this research, we hope to show how a complex *in vitro* dynamic model such as the tumour-on-a-chip can better recapitulate the tumour microenvironment and therefore enhance our understanding of biological interactions with NPs.
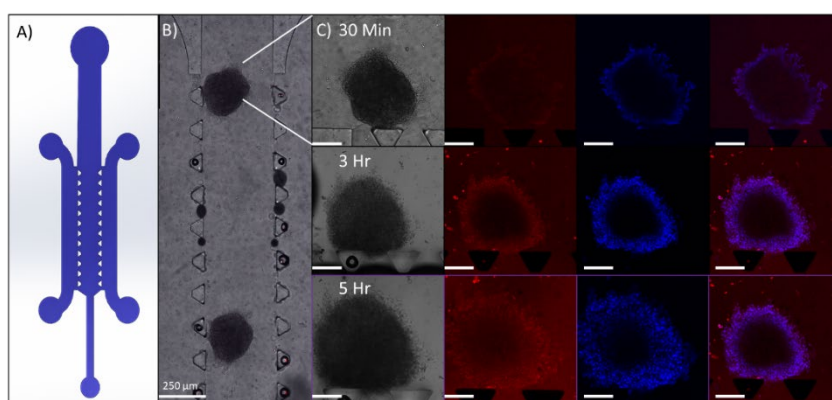
Figure 1: Tumour-on-a-chip microfluidic device design created with CAD software (A). Tile scan image of tumour spheroids present in the microfluidic device after being incubated overnight (B). NP uptake into tumour spheroid at various timepoints (C) with NP tagged with Cy5 shown in red and Hoechst stain of live cells in blue. Images taken using a Leica SP8 confocal microscope at 10x magnification. Scale bar denotes 200 μm.

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POSTER

Biologics as Peptide Delivery Vehicles for Immunotherapy

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Lung cancer is the leading cause of cancer-related deaths in the world. Conventional treatments for non-small cell lung cancers (NSCLCs) include surgery and chemotherapy but these treatments are associated with undesirable side effects. InterK Peptide Therapeutics have developed immune-boosting peptides as an alternative treatment strategy. Currently, the accumulation of these peptides in the tumour microenvironment is limited as they undergo rapid renal clearance. This talk will discuss the use of antibody fragments specific to various tumour and immune cell surface receptors for targeting the tumour microenvironment. These antibody fragments have been linked to InterK's peptides, creating biologics which have a longer blood circulation time. As a result, the antibody fragments can guide InterK's peptides to the tumour microenvironment and increase their site-specific accumulation. The peptides then boost the immune system by re-invigorating exhausted T cells and mounting an immune response to suppress lung cancer. Validation of these constructs have been undertaken in mouse models. InterK's peptides are a promising new class of immunotherapeutics that could serve NSCLC patients who do not respond to conventional therapies. The use of a delivery platform, such as the biologics in this project, are beneficial to ensuring these therapeutic peptides are directed to the tumour microenvironment. With a decrease in the likelihood of undesirable side effects, the quality of life for patients suffering from NSCLCs is likely to improve. While this project focusses on NSCLC treatment, the findings from this study can be applied to other types of cancer.

POSTER

Cross-modality image estimation in molecular imaging

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Abstract

Multi-modality medical images are integral to clinical decision making. Acquisition of images using multiple modalities has distinct disadvantages because multiple scans increase cost and radiation dose and delay clinical workflow. One potential solution is cross-modality image estimation, which involves the synthesis of medical images between different imaging modalities.

Cross-modality image estimation is beneficial for molecular imaging in different ways. The first application is estimation of CT from MRI. The superior soft tissue contrast and avoidance of radiation exposure has led to MRI replacing CT in many clinical applications. However, CT images are still required for radiotherapy treatment dose calculation and for PET attenuation correction as part of image reconstruction (Leynes et al. 2018). The second need is for the estimation of PET from MRI. Instances arise in the clinical setting, for example in Alzheimer's disease, where patients who have undergone MRI scans were not able to complete a PET scan (Li et al. 2014). Here, MRI can be used to estimate ^{18}F-FDG-PET images, as it is known that the ^{18}F-FDG-PET tracer is taken up by the brain in general, resulting in PET soft tissue anatomical contrast. The final notable gap is where the low-resolution PET provides anatomical and functional mismatch and blurring between the modalities. In this situation a high-resolution PET image can be estimated from low count data.

Machine learning methods have widely been investigated for cross-modality medical image estimation. Image estimations have been performed using convolutional neural networks (CNNs), requiring large datasets to project highly complex information across modalities (Mamoshina et al. 2016). The generative adversarial network (GAN) is an extension of CNNs, where two competing networks are trained simultaneously, a so-called generator and discriminator. A conditional GAN (cGAN) additionally constrains the output by placing learning conditions on the generator or the discriminator to help constrain image estimates (Dar et al. 2019).

Cross-modality image estimation methods have received increasing attention in the field of medical imaging. Today, cGANs appear to have superseded CNNs and conventional GANs for cross-modality image estimation. Though, there are still few considerations to develop the deep learning framework for image estimation. First, to ensure spatial consistency is retained between the input and output images, the learning process requires constraints, either through registration, landmarks, or novel loss functions. Second, the training data to achieve the desired results in deep learning, should be large which is typically challenging due to existing small sample size. Finally, the limitation of computational resources should be compensated by choice of input to the network. These findings now provide opportunities to pursue cGAN based approaches for cross-modality medical image estimation involving SPECT.

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POSTER

Submillimeter, Sub-minute Quantitative Susceptibility Mapping using a Multi-Shot 3D-EPI with 2D CAIPIRINHA Acceleration

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Quantitative Susceptibility Mapping (QSM) uses phase information to produce maps of magnetic susceptibility that give insight into tissue composition and structural organization. In number of clinical applications where submillimetre voxel size and large imaging volumes are beneficial, scan times using the traditional 3D Gradient Echo (3D-GRE) sequence can be as long as 8 to 15 minutes.[1,2,3] In this work, we implemented a multi-shot 3D-EPI sequence combined with 2D-CAIPIRINHA acceleration to achieve high-quality susceptibility maps, with minimal blurring and distortions, at 0.80 mm and 0.65 mm isotropic resolutions in 58 and 87 seconds, respectively.

All images were acquired on a MAGNETOM Prisma 3T MR scanner (Siemens Healthcare, Erlangen, Germany) on software version syngo.MR E11 with a 64-channel head/neck coil. The prototype 3D-EPI sequence uses a binomial water excitation RF pulse was used with TBW = 24, FA = 16º, FOV = 250 mm, EPI Factor = 13, BW = 395 Hz/pixel, TE/TR = 25/31 ms. A multi-echo 3D-GRE sequence with FA = 15º, FOV=250 mm, BW = 210 Hz/Px, TE/TR = (5,10, 15, 20)/31 ms, was acquired for comparison. Data were processed offline with the QSMxT pipeline[4], using TGV-QSM[5].

Figure 1 shows susceptibility maps for the 0.65 mm resolution 3D-GRE and 3D-EPI scans. In this case, an EPI factor of 13 (13 phase-encoding lines per shot) was a good compromise between acquisition time and a reduction in EPI artefacts, and no B0–related distortion correction needed to be performed. The 3D-GRE QSM is visibly smoother, with higher SNR than the 3D-EPI as a result of the multi-echo acquisition and lower receiver bandwidth. However, the contrast of the maps and the ability to differentiate between different anatomical structures are comparable across the sequences and increasing accelerations. The susceptibility distributions in tissue regions are comparable across the acquisitions, and are comparable to values previously reported in the literature.[5,6] Future work will focus on evaluating the reduced sensitivity to motion of the 3D-EPI sequence.
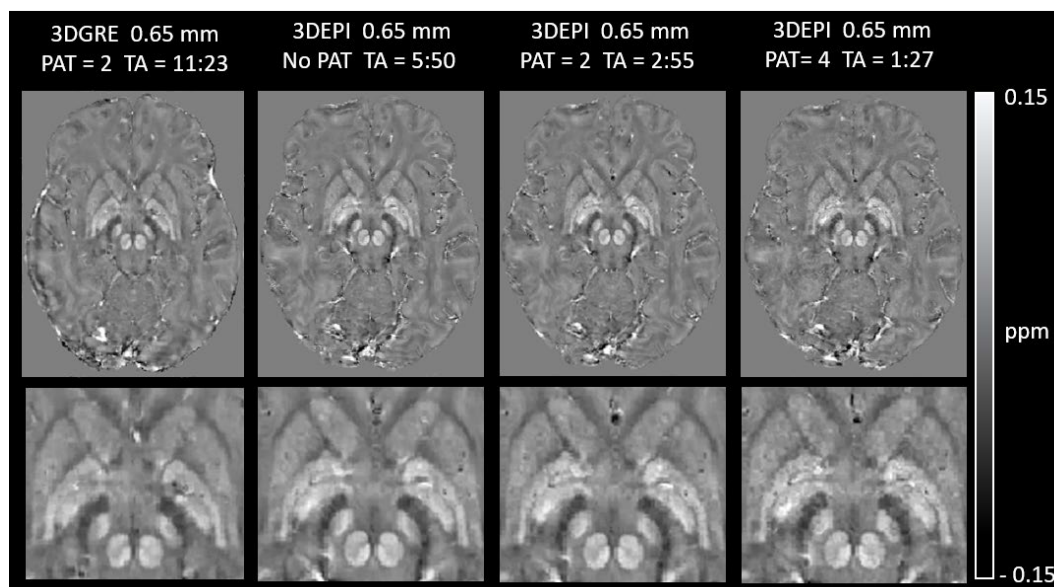
Figure 1. *Top Row: Acquisition times (TA) and susceptibility maps for the 0.65 mm isotropic resolution 3D-GRE acquisition (left column) and 3D-EPI acquisitions (right three columns) with different acceleration factors (PAT). Enlarged regions for each map are shown in the bottom two rows.*

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