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Two methods for labeling tyrosine kinase inhibitor sorafenib with carbon-11 to obtain a PET tracer for personalized cancer treatment

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Background: Receptor tyrosine kinases play a pivotal role in the signal transduction of vital processes of the cell. Uncontrolled activation of these receptors is often related to tumor formation. Therefore, tyrosine kinases have become important drug targets in the treatment of cancer. Although several TKIs have demonstrated clinical value, most TKIs developed are only effective in a small subset of patients. In our center, PET imaging with radiolabeled targeted drugs is used as quantitative imaging strategy to speed up drug development and to facilitate patient selection. The aim of the present study was to develop radiolabeled [11C]sorafenib, to be used for the selection of patients who might benefit from treatment. Sorafenib was labeled at two positions to obtain [11C]methyl sorafenib or [11C]urea sorafenib. Both compounds are preclinically evaluated for there tumor targeting properties as well as their active metabolites.
Materials and Methods: [11C]methyl sorafenib was synthesized by reacting desmethyl sorafenib with [11C]MeI in a solution of TBAOH in DMF at elevated temperature. For the synthesis of [11C]urea sorafenib, a Rhodium promoted reaction between the corresponding azide and amine was performed at elevated pressure and temperature in the presence of [11C]CO. the crude mixtures were purified by HPLC and the isolated product fraction was formulated by solid phase extraction in ethanol and 2.5% polysorbatum in 0.9% saline solution (1:9 v/v).
Results: [11C]methyl sorafenib was synthesized in a decay corrected yield of 60% and [11C]urea sorafenib was synthesized in a decay corrected yield of 27%. Using analytical HPLC, the radiochemical purity of both products was determined to be higher than 99% and the identity of the product was confirmed by coinjection of the labeled product with reference sorafenib. A metabolite analysis in rats, revealed that the percentage of intact product in blood-plasma samples after 45 minutes are up to 90% for [11C]methyl sorafenib, and 96% for [11C]urea sorafenib, respectively.
Conclusion: Reliable labeling procedures were developed for the synthesis of [11C]methyl sorafenib and [11C]urea sorafenib. Both labeled products were obtained in a high yield and purity. Furthermore, both products have a high metabolic stability in rats and its tumor targeting properties are currently tested in renal cell carcinoma xenografts.

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Predictive assays for targeted therapeutics using image-based high content analysis of patient-derived tumor models

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Background: The role of different cellular components of tumor microenvironment in dictating the sensitivity towards chemotherapy has been used in developing strategies to improve cancer therapy. However, most of the in vitro drug treatment studies use cell lines which lack many major components of the tumor environment. With more drug candidates being introduced for targeted therapies of cancers, it becomes important to test these compounds on a mixture of target positive and negative cell population to demonstrate the selective effect before being introduced into a Phase I clinical trial. Patient derived tumor models are superior to cell lines as representative of the actual tumor microenvironment. However, interpretation of the results from in vitro treatment studies using these cells becomes highly complicated due to the non-homogeneous nature of the cells. Molecular Response has an extensive collection of patient derived tumor cells which can be used to evaluate the therapeutic agents for their targeted action on cell populations. We have developed novel approaches to evaluate the anti-proliferative action of cancer drugs on cell population of specific interest.
Materials and Methods: This study investigated the utility of patient derived tumor cells to investigate selective anti-proliferative effects on cell populations carrying specific mutations. Using qPCR based assays, NSLC and melanoma samples were screened to identify samples with ALK-EML fusion and BRAF-V600E mutation status respectively. Using high content imaging platform, the selective anti-proliferative effects of crizotinib on cell populations with ALK-EML fusion as wells as effect of plx4032 on B-RAF mutated populations were assessed.
Results: We developed a semi-quantitative qPCR based screening strategy for cancer samples (N = 2000) to identify high expression of ALK. The fusion and ALK-EML was confirmed by FISH. PCR assays to detect BRAF mutations were used to screen a collection of melanoma samples (N = 900). Using a novel image-based high content screening assay, the targeted action of crizotinib as well as plx4032 were demonstrated in mutated cell populations in the context of a non-targeted cell population.
Conclusion: The use of patient-derived tumor cells as a tool to develop novel approaches to delineate the effects of therapeutics on targeted cell population has shown great potential in oncology drug development. This novel approach provides a means to evaluate variety of drug candidates used in personalized medicine.

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Molecular imaging demonstrates GLPG0187, a small-molecule integrin antagonist, binds to RGD-integrin receptors in vivo and is efficacious in tumor and metastasis models

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Background: GLPG0187, an integrin antagonist with nanomolar affinity for RGD-integrin receptors, shows potent anti-tumor activity in vivo. In a mouse model of human breast cancer bone metastasis, the combination of GLPG0187 with standard-of-care anti-resorptive (zoledronate) and chemotherapeutic (paclitaxel) agents shows superior efficacy when compared to each treatment taken separately providing further evidence that GLPG0187 could be an effective therapeutic for the treatment of tumors and metastases. The primary objective of this study was to evaluate whether GLPG0187 reaches target RGD-integrin receptors in vivo to identify an imaging biomarker suitable for patient selection. We also compared doses of GLPG0187 that compete for receptor binding with doses that are pharmacologically relevant.
Materials and Methods: Molecular imaging was performed by fluorescent tomography using Integrisense680and by μPET using 18F probes. Mice were bearing established human melanoma A375 xenografts and human MDA-MB-231/B02 breast cancer bone metastases.
Results: In the bone metastasis model, GLPG0187 administered at the pharmacologically efficacious dose of 30 mg/kg, p.o., b.i.d., displaced up to 70% of the binding of Integrisense680, thereby demonstrating its ability to bind RGD-integrin receptors in vivo. In melanoma xenografts, higher doses of GLPG0187 were needed to reach significant effects, both in receptor binding and efficacy studies. An assessment using [18F]-RGD and [18F]-FDG in melanoma xenografts indicated probe displacement and therapeutic efficacy, respectively, by GLPG0187.
Conclusion: These molecular imaging experiments show that GLPG0187 binds to target RGD-integrin receptors in tumors and metastases in vivo. Recent successful developments in oncology have relied on careful patient selection based on target expression. These preclinical data support the use of PET probes to select patients bearing RGD-integrin positive tumors in GLPG0187 clinical trials.

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Gene expression profiles obtained from mouse hair, xenograft tumor and ex vivo human scalp hair to determine the effects of drug response following treatment with an EGFR inhibitor

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Background: Plucked human scalp hair represents an ideal surrogate sentinel tissue to enable non-invasive monitoring of drug response, both in clinical trials and also as a discovery platform ex vivo. Congruence of patterns of transcriptome activity show high concordance to that of primary target tissue. EGFR is often over expressed or deregulated in a variety of solid tumours and EGFR inhibitors are increasingly part of the therapeutic treatment of advanced lung, head-and-neck and colorectal carcinoma. Desirable non-invasive transcriptional biomarkers of EGFR inhibition demonstrate target engagement and define a PK/PD relationship, as well as enabling the monitoring of a well tolerated dose schedule with maximal biological effect.
Materials and Methods: In this study, we initially focused on a preclinical mouse model in which we analysed gene expression profiles in plucked hair following treatment with erlotnib. These biomarkers were then explored further in an erlotinib dose escalation study in mouse hair and tumour samples collected from a preclinical lung xenograft model. Epistem has also developed an ex vivo human hair culture platform in which we are able to model pharmacodynamic consequences of small molecules and biotherapeutics. We deployed our proprietary platform here to investigate the relationship of pre-clinical markers of erlotinib exposure in vivo to those discovered ex vivo in human hair treated with the erlotinib.
Results: We were able to demonstrate from the xenograft study, a panel of transcriptional markers that exhibited a similar response to drug treatment in both tumour and hair. The differentially expressed genes identified in both mouse and human hair samples were biologically relevant. The panels identified included genes in the EGFR signaling pathway or are known to be associated with this pathway. We also used our ex vivo human hair culture platform to further explore the relevance of pre-clinical markers of