Project Information

Project Title
Cytokine analysis and lean muscle mass gain in patients on ongoing phase II BILMEK study: analysis of markers of response to chemotherapy and MEK inhibition in advanced biliary cancer.

Amount Requested
50000

Applicant Information

Applicant Name
Mark Doherty

Degree (Select all that apply)
- MD

Academic Level (Select all that apply)
- Post-Doc Fellow

Institution Information

Institution Name
University of Toronto/Princess Margaret Cancer Centre

Institution City
Toronto

Institution State
Ontario

Institution Country
Canada

Email
mark.doherty@uhn.ca

Mentor Information

Mentor Name
Jennifer Knox
### 2017 Research Fellowship Application: Entry # 11021

**Mentor Title**

Associate Professor/Staff Medical Oncologist

**Mentor Institution**

University of Toronto/Princess Margaret Cancer Centre

#### Submission Information

**Scientific Abstract (1500 Max Characters)**

New treatments and strategies for advanced biliary tract cancer (BTC) are urgently needed. BILMEK is an ongoing randomized phase II study in advanced BTC, investigating the addition of the MEK inhibitor selumetinib in two regimens to standard cisplatin/gemcitabine chemotherapy (CisGem). We aim to evaluate two potential biomarkers in patients on this trial, to help identify patient subgroups benefitting from selumetinib: changes in lean muscle mass (LMM), and changes in plasma cytokine signature (CS).

BTC frequently arises in the setting of chronic inflammation, associated with a pro-inflammatory CS. Preclinical studies of selumetinib suggested modulation of interleukin-1 (IL-1), IL-6, and tumor necrosis factor-α (TNF-α). Banked blood from each patient at 1st and 2nd treatment cycles and at disease progression, will be used for analysis of CS (interferon-γ (IFN-γ), IL-1β, 2, 4, 6, 10, and 17, and TNF-α). We aim to analyse the association between changes in CS and treatment response and resistance. Preliminary data with selumetinib suggested an increase in LMM in treated patients, compared with historical controls. We will use quantitative analysis of serial CT scans to measure change in LMM in all patients enrolled on BILMEK. This aims to confirm the previous findings, and to test the association between this change and treatment response. We will also examine the relationship between CS and change in LMM, as pro-inflammatory cytokines are implicated in cancer cachexia.

**Research Summary (1500 Max Characters)**

BILMEK is a randomized phase II study of selumetinib in sequential or continuous combination with CisGem in patients with cholangiocarcinoma or gallbladder cancer. As of September 2016, 75% of a target of 48 patients are accrued.

For CS analysis, 10ml plasma is collected from each patient at cycle 1, cycle 2 and at time of progressive disease (PD), and stored in batch at -70°C. Levels of IFN-γ, IL-1β, 2, 4, 6, 10, 17, and TNF-α will be analyzed using a commercially-available multiplexed electrochemiluminescence (ECL) immunoassay (Meso Scale Discovery, Rockville, MD). Analyses will focus on: differences in mean change in CS between treatment arms, association between changes in CS a PD and survival, and relationship between CS and change in LMM.

For LMM analysis, CT scans performed for response assessment on study will be used. LMM analysis will be performed using Slice-O-Matic software (Tomovision, Montreal, Canada). Total skeletal muscle and adipose tissue will be measured in cm2 at the level of the L3 vertebra. Changes in muscle or adipose tissue will be divided into three categories: muscle loss (≥6cm2), stable muscle mass (+/- 5.9cm2) or muscle gain (≥6cm2), equivalent to a change of ≥1kg in muscle mass. Categories for adipose tissue will use cut-offs of 14.7cm2, equivalent to 1kg change in adipose mass. Differences in mean values for LMM between treatment arms will be tested, and associations will be explored between LMM change and CS, treatment response and survival.

**Lay Summary (1500 Max Characters)**

BILMEK is a randomized phase II study of selumetinib in sequential or continuous combination with CisGem in patients with cholangiocarcinoma or gallbladder cancer. As of September 2016, 75% of a target of 48 patients are accrued.
Cancer of the bile duct (also known as cholangiocarcinoma) is a highly lethal disease, with very few effective treatment options. Chemotherapy can prolong life by a short time and with improved quality of life. However, better treatments are urgently needed, with better ways to predict the benefit of those treatments. A clinical trial of selumetinib (a targeted therapy) in combination with chemotherapy is currently enrolling patients at Princess Margaret Cancer Centre. The proposed project is aimed at identifying which patients truly benefit from selumetinib.

There is a strong relationship between bile duct cancer and inflammation, which can be measured by testing for chemicals called cytokines in blood samples. Some laboratory studies suggest selumetinib may suppress these cytokines, but this has not been studied in humans. Other studies suggest this drug may reverse the loss of weight and muscle mass associated with cancer. We plan to 1) measure the levels of cytokines before and after chemotherapy and selumetinib in patients on our trial, and compare with those treated with chemotherapy alone and 2) measure the gain or loss of muscle mass in these patients using images from their CT scans (used to measure the tumor growth or shrinkage on treatment). We plan to use these analyses to try to predict which patients will benefit from selumetinib in combination with chemotherapy for bile duct cancer, and change the way we choose patients for this targeted therapy.

Specific Aims (2500 Max Characters)

1. To analyze plasma cytokines as predictors of response and resistance to MEK inhibition in combination with chemotherapy for advanced BTC. Selumetinib has preclinical evidence of synergy with gemcitabine-based chemotherapy in BTC, and BILMEK is testing it in biologically-unselected patients. In preclinical studies, selumetinib appeared to modulate levels of pro-inflammatory cytokines, which may represent an alternative mechanism of action, and may also be a marker of response and resistance. Measuring CS following selumetinib and chemotherapy, we aim to explore change in CS as a biomarker for response in BTC. Biomarker discovery could lead to a shift in the clinical development of selumetinib in BTC, and help define subgroups that benefit from this targeted treatment.

2. To examine change in LMM in patients treated with MEK inhibition with chemotherapy for advanced BTC, explore relationships between LMM change and CS, and analyze the association between this effect, tumor response and survival. Despite better understanding of underlying causes of cancer cachexia, uncertainty persists. IL-1β and IL-6 have been associated with the inflammatory state that drives cachexia, and as noted above may be modulated by selumetinib. Treatments that prevent or reverse loss of LMM have proven elusive, aside from the development of ghrelin agonists. Gain in LMM was reported in a small study of selumetinib. Confirming this effect in a randomized trial and correlating LMM with treatment response and survival may aid biomarker discovery. In addition, correlation with CS may help explain the underlying mechanism, and could lead to further study of selumetinib as a treatment for cachexia.

Background and Significance (3000 Max Characters)
Chemotherapy in patients with advanced BTC can prolong survival and improve quality of life, with the current standard of care being the CisGem combination. Chronic inflammation plays a large role in the development of BTC, as evidenced by several of its risk factors: gallstone disease, chronic hepatitis and primary sclerosing cholangitis. Additionally, in patients with established BTC, elevation of the neutrophil/lymphocyte ratio (a signature of inflammation) is frequently observed and has been associated with poor survival. Interleukin-6 (IL-6) is a pro-inflammatory cytokine, and seems to play a significant role in biliary carcinogenesis. IL-1β and IL-6 are also associated with cancer cachexia, a frequent problem in patients with advanced cancer.

To date, no molecularly targeted agents have been approved in the treatment of BTC. Studies of the molecular biology of BTC have reported frequent mutations in the KRAS gene, as well as in other components of the mitogen-activated protein kinase (MEK) pathway. Inhibition of MEK can arrest signaling through this pathway, and specific MEK inhibitors are in clinical development. Selumetinib (AZD 6244, AstraZeneca, London, UK) is an allosteric inhibitor of MEK1/2 phosphorylation of ERK. As a single agent, it demonstrated moderate clinical activity in advanced BTC as well as manageable toxicity. In preclinical studies performed at Princess Margaret Cancer Centre, there was evidence of a sequence-dependent effect when selumetinib was combined with gemcitabine in BTC primary xenografts. When gemcitabine was administered 48 hours after stopping selumetinib, there was evidence of synchronised S phase cell cycle re-entry, and marked tumour growth inhibition compared to continuous dosing of the two agents, or either agent alone. In combination with the clinical data regarding CisGem chemotherapy, these results formed the basis for the BILMEK trial.

As a single agent, selumetinib was noted to increase skeletal muscle mass with 84% of patients gaining lean muscle, compared with 17% of historical controls on standard chemotherapy. This effect has not been confirmed in the setting of a randomized trial. It is postulated that this phenomenon may be mediated through direct inhibition of IL-6 production, and some preclinical data suggest that selumetinib can suppress TNF-α, IL-1 and IL-6 although conclusive evidence is yet to be presented. Other studies have also suggested IL-6 overexpression and activation of the STAT3 pathway play a role in selumetinib resistance. Given this background, we propose analysis of these two potential biomarkers of response to selumetinib to aid treatment selection for patients and to develop strategies to overcome treatment resistance.

Project Timeline (2500 Max Characters)

The BILMEK trial opened to accrual in March 2015 at Princess Margaret Cancer Centre in Toronto, ON, Canada and Cross Cancer Institute in Calgary, AB, Canada. It has enrolled 75% of a target 48 patients to date, and completion of accrual is expected by March 2017. CT scans for measurement of LMM will be performed at study enrollment and at week 10 on study treatment. With this schedule, the analysis of LMM can be undertaken approximately 10 weeks after enrollment of the final study patient. Plasma for cytokine analysis is drawn at study entry, cycle 1 day 1, cycle 2 day 1 and at time of disease progression. With this schedule, batched analysis of PCS is planned for Q3 2017. The principal applicant's fellowship extends until Q4 2017 allowing completion of the trial and this proposed study, and 25% of the applicant's workload is related to the BILMEK trial. We expect to submit the results of these analyses for presentation at the American Society of Clinical Oncology Annual Meeting 2018, followed by publication in a peer-reviewed journal.

Literature Cited

Budget

Impact or Collab Award

I want to apply for an Impact Award (up to $50,000)

Impact Applicant Name

Mark Doherty

Impact App Title

Clinical Research Fellow in Medical Oncology

Impact App Institution

University of Toronto/Princess Margaret Cancer Centre

Impact App Personnel 1 Name
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### Impact Equipment List

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<td>Slice-O-Matic Software license and support contract.</td>
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<td>ECL platform usage cost for analysis of cytokine samples.</td>
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### Impact Supplies List

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<td>ECL cytokine panels, Cryovials, pipettes, pipette tips, EDTA tubes</td>
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### Impact Travel List

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<td>Attendance at Annual American Society of Clinical Oncology meeting to present results of data.</td>
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<td>Shipping of samples for cytokine analysis and CT images to Princess Margaret Cancer Centre, storage of cytokine samples at -70°C.</td>
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<td>Laboratory technician for ECL assays.</td>
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<td>Bio-statistician fee to analyze cytokine and imaging data.</td>
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**Impact Total Requested**

50000