Progress Report on BAOMS Research Grant

**Title**

Investigating the potential of statins to prevent and treat Anti-Resorptive agent induced Osteonecrosis of the Jaw (ARONJ).

Principle investigator: Mr Ibraz Siddique

**Method**

Completion of In-vivo study in the 5TGM1 murine model of myeloma

Groups of 6‐8 week old male mice have been injected intravenously with Phosphate Buffered Solution (PBS) or 2x106 5TGM1-eGFP cells.

Tumour induced mice have been treated with PBS, zoledronic acid (125μg/kg subcutaneously once a week) and/or simvastatin (10 mg/kg daily).

Mice have been sacrificed at the first signs of illness. Tumour burden has been analysed in femoral bone marrow flushes by flow cytometry and by histological analysis of tibial sections stained with anti-CD138 by colony counting.

Bone disease has been assessed by micro‐CT scanning.

OsteoMeasure has been used to count the numbers of tartrate resistant acid phosphatase (TRAP) positive osteoclasts and osteoblasts on cortico‐endosteal bone surfaces. Levels of bone serum markers (TRAP5b expressed by osteoclasts and pro-collagen 1 N-terminal peptide) have been measured to assess systemic bone disease by ELISA.

**Study Groups (n=8/group)**

Group 1: Naive

Group 2: Tumour + PBS

Group 3: Tumour + zoledronic acid

Group 4: Tumour + statin

Group 5: Tumour + combination

**Stage 1:** **Completed**

Dose titration in tumour free mice.

Dose-response of statin and bisphosphonate monotherapy in mice.

Effects have been assessed by Micro-CT Histology.

Milestone 1 (completed): Estimation of lowest dose for simvastatin and bisphosphonate monotherapy to induce measurable responses.

**Stage 2: Completed**

Evaluation of statin and bisphosphonate combination therapy in tumour free mice.

Minimum effective doses from Stage 1 has guided doses for Stage 2. Untreated and monotherapy animals from Stage 1 have acted as controls. Effects have been assessed as per Stage 1.

Milestone 2 (awaiting data analysis): Determination of whether statins alter the effect of bisphosphonate on bone remodelling in a tumour free model.

**Stage 3: Completed**

Groups of mice injected with vehicle or 2x106 5TGM1-eGFP cells intravenously. Following establishment of tumour mice were treated with vehicle, bisphosphonate and/or statin (doses guided by results from Stage 1 and 2). Mice were sacrificed at first signs of illness. Assessment methods as per Stage 1: MicroCT, histology and serology. CD138 immunohistochemistry was used to show myeloma cells in bone marrow. Electrophoretic measurement of serum paraprotein and flow cytometric estimation of myeloma cell count in bone marrow flushes

Milestone 3 (awaiting data analysis): Determination of laboratory and behavioural effects of bisphosphonates on bone metabolism in a myeloma model.

 Financial Itinerary



Current status

In vivo study complete. Awaiting data analysis.