

Dual reporter *L. major* strain for *in vitro* and *in vivo* investigations of cutaneous leishmaniasis

Jodie Dixon¹, Katrien Van Bocxlaer¹

¹York Biomedical Research Institute, University of York (UK)

Cutaneous leishmaniasis (CL) is a neglected tropical disease causing a range of skin lesions resulting in life long scarring and thus, discrimination and stigmatisation in poorer communities. Unlike visceral leishmaniasis CL is not fatal and with no human vaccine available, disease control relies on chemotherapeutics; mostly of which are sub-optimal, mounting drug resistance and toxicity. With limited advances in drug research and development (R&D) of CL, better understanding of parasite activity in the host and interactions with compounds is needed to be able to follow infection and accelerate the drug discovery pipeline. Here we justify the development of dual reporter bioluminescent (red-shifted firefly luciferase) and fluorescent (mCherry) virulent strains of *Leishmania*, allowing for a wide range of possibilities for both *in vitro* and *in vivo* investigations.

A 1696bp of PpyRE9h firefly luciferase coding region was PCR amplified from pTRIX2-LucNeon vector, as well as a 755bp mCherry coding region amplified from pGL1894 expression vector. Fragments were inserted into pLEXY-hyg 2.1 (Jenabioscience, Jena, Germany) vector using a High fidelity DNA assembly kit (New England Biolabs). We then engineered a transgenic *L. major friedlin* strain to express the bioluminescent and fluorescent proteins. In an *in vitro* model, parasites infected THP-1 cells to quantify infectivity, to assess activity of relevant drug compounds when added to infected cells. Validation is ongoing, yet this dual reporter may be a promising tool to investigate drug activity both *in vitro* and in experimental models of CL.