Nanoprecipitated solid drug nanoparticles of poorly soluble niclosamide and *in-vivo* demonstration of long-acting delivery

Catherine Unsworth[1], Alison Savage[1], James Hobson[1], Andrew Dwyer[1], Steve Rannard[1]

[1]Centre of Excellence for Long-acting Therapeutics, Department of Chemistry, University of Liverpool, Liverpool, L7 8TX, UK

Background: Niclosamide (NCL) is a cheap, broad-spectrum anthelmintic drug with reported activity against certain viral respiratory infections, including SARS-CoV-2, several forms of cancer, Parkinson's disease, and some bacterial infections. NCL is practically insoluble in water, resulting in very low bioavailability. As a result, the current oral formulation is unsuited to provision of the high systemic drug concentrations required for therapeutic activity, a hurdle which could be overcome through parenteral administration which may also offer long-acting delivery. Solid drug nanoparticle (SDN) synthesis techniques offer the opportunity to create nanoparticles made of the drug itself, allowing for higher loading, increased therapeutic concentrations and lower excipient content, reducing side effects.

Methods: We have demonstrated scalable formation of NCL SDNs through nanoprecipitation in the presence of biocompatible excipients to produce a semi-crystalline material suitable for injectable administration. NCL dissolved in a water-miscible solvent system is added rapidly to an aqueous phase, leading to supersaturation and nucleation. Stabilising excipients limit growth of NCL particles and a sonication phase breaks down any aggregates. Precipitation is immediately followed by spray drying to remove solvents and produce a free-flowing, dispersible powder. Optimisation of the nanoprecipitation procedure was driven by feedback from observation and measurement of target characteristics: stability of the redispersed material in aqueous media suitable for injection, SDN particle size - hydrodynamic diameter, Dz – and dispersity (PDI) measured by dynamic light scattering and "syringability" – the concentration of NCL in dispersion which can be passed through a syringe and needle of a gauge suitable for injection. HPLC was used to determine concentration of NCL in SDN powder.

Results: Optimised SDN powders consisted of particles with Dz in the range of 550-600 nm, PDI in the range of 0.168-0.179, syringable concentrations of up to 450 $mg.mL^{-1}$ with respect to NCL and powder stability of several months. Adjustment of NCL solvent system, NCL loading within the formulation, excipient choice and aqueous volume in the nanoprecipitation were found to positively impact syringable concentrations. Confirmation of targeted drug loading (50% w/w) in the powder was achieved by gradient HPLC analysis both immediately following synthesis and after several months in storage, suggesting no degradation of the drug. In-vivo study of the long-acting nature of NCL delivery through intramuscular injection in Sprague Dawley rats was used to establish the pharmacokinetic (PK) release profile and demonstrated sustained NCL plasma concentrations over 28 days. However, a high initial release (Cmax) was observed.

Conclusions: We have developed a formulation of NCL shown to have characteristics and an *in-vivo* PK profile suitable for injectable, long-acting delivery. Improvement to the formulation and delivery volume to control the initial burst release could increase the period in which NCL is at therapeutic concentrations in systemic circulation and reduce the likelihood of any potential toxic side effects. Post-production gamma irradiation sterilisation and accelerated storage stability trials are in process and initial indications suggest long-term stability of the material as well as no distinguishable degradation of NCL after sterilisation. The application of NCL SDNs to treatment (bacterial infection) and prophylaxis (post-surgery thrombosis) is under investigation investigated.