IRISH MEDICINES BOARD
NON-TECHNICAL PROJECT SUMMARY

NON-TECHNICAL PROJECT SUMMARY DETAILS
Project reference number: V012/2013Q3
Project title: Research investigation into the neurodevelopmental and behavioural effects of methamphetamine exposure during pregnancy and lactation in rats
Duration of the project work (months): 36 months
Project keywords: Methamphetamine; pregnancy; lactation; development; behaviour; rats
Purpose of the project under Article 5 of Directive 2010/63/EU: Translational and applied research; human nervous and mental disorders
Project objectives, scientific unknowns or scientific or clinical needs being addressed: Clinical evidence shows an increase in the use of methamphetamine, a drug of abuse, in pregnant women. It is vital that enhanced understanding of the risks associated with taking this drug during pregnancy or breastfeeding, and the potential adverse effects exposure can have on the children of mothers involved are investigated. This study will examine whether methamphetamine exposure in pregnant rats (or in the post-littering period) will result in any changes in the early life development of offspring, or in their behavioural responses in later life, at adolescence and adult stages. The results of this study can be translated to human scenarios in order to improve clinical knowledge of the potential effects on exposed children.
Potential benefits likely to derive from this project: Using a range of clinically relevant treatment regimes and scenarios, this project will examine a variety of neurodevelopmental parameters over a range of stages, for example infant, adolescent and adult stages of rat development in order to perform a thorough risk assessment of methamphetamine exposure during pregnancy and lactation, and the effects on subsequent offspring. The effects the exposure has on the development and behaviour of offspring will also be investigated, which will enhance the understanding of the impact of methamphetamine exposure clinically.
Species and approximate number of animals expected to be used: 1500 rats
Expected adverse effects on the animals, the expected level of severity and the fate of the animals: Methamphetamine will be administered orally to groups of pregnant females at different stages of their

pregnancy, with exposure in one group of females extending into the lactation period. This procedure is classed moderate in severity. Adverse effects as a result of this level of methamphetamine exposure are not expected to be seen in the pregnant rats themselves. However secondary adverse effects, which are rare, may include damage to the food pipe of the rat from the instrument used for oral administration. A final group of pregnant rats will also be exposed to an antioxidant in order to investigate reversal of the negative effects of methamphetamine. This is classed as a mild procedure. Adverse effects from exposure to the antioxidant are not expected at the level of drug administered to animals. This is classed as a mild procedure. Upon delivery of litters, pups from each group of methamphetamine exposed pregnant mothers, will undergo behavioural and developmental procedures designed to examine the effect of methamphetamine exposure on methamphetamine exposed pups. Procedures include investigation of righting reflexes, forelimb grips and balance of the pups.

Additional groups of pups will progress to adulthood and will be injected with one of three psychotropic drugs in order to determine how methamphetamine exposed offspring react to psychotropic drugs; the administration of these drugs is classed as mild in severity. Adverse effects from the use of these drugs are not anticipated at the dose levels used. However secondary associated adverse effects, which are rare, may include pain and discomfort due to the physical injection process. Treated animals will then undergo three behavioural test procedures that will assess the responsivity to drugs that will reduce depressive symptoms, anxiety symptoms or that have a stimulant action. These tests all have a mild severity classification. The final procedure is a behavioural test called the forced swim test where animals are placed in a cylinder filled with water and from which they cannot escape. This procedure can cause stress to the animal and so has a severity classification of severe. In order to minimise the distress caused, animals will be dried and kept warm after being in the water; this procedure will be performed by trained and experienced personnel.

A final group of pups will progress to adulthood where the effects of methamphetamine exposure on adult behaviour will be examined. Animals will take part in a series of behavioural procedures such as: ability to locate their home cage; ability to experience anxiety and ability to learn and form memories. Each of these procedures will take place for a short period of time and are not expected to cause distress to the animals. These procedures have a mild severity classification.

At the end of the study, animals will be euthanised humanely.

APPLICATION OF THE 3RS

Replacement - why animals need to be used for this project and why non-animal alternatives could not be used:

As the major objective of these studies is to investigate animal behaviour following exposure to drug substances, therefore use of a non-animal alternative was not possible.

Reduction - how the use of minimum numbers of animals can be assured:

Data from previous studies and literature reviews was used to estimate the minimum number of animals required. Statistical methods were then employed in order provide further information on the minimum number of animals required to display objective results.

Refinement - justification for the choice of species, why the animal model(s) used are the most refined and general measures to be taken to minimise harm to the animals:

The laboratory rat is the most widely used species for the types of experiments covered in this project proposal, and the strain being used is very commonly used worldwide, and suitable for this study.

A number of methods were incorporated into the study design to minimise any unnecessary distress or suffering to the animals. Examples include:

- Use of only trained and experienced personnel throughout the study
- Administration of drugs with minimal adverse effects at site of injection

- Limiting the duration of behavioural procedures to the minimum possible duration
- Drying and keeping animals warm after being placed in water
- Minimising separation times of pups from mother
- A veterinarian will be available for consultation should any animal welfare issues arise