Home Office

PPL Standard Condition 18 Notification Form

Please use this form to notify the Animals in Science Regulation Unit (ASRU) of procedural-related adverse effects that have exceeded or are likely to exceed the severity limitations or controls described in the project licence. ASRU will use this information to determine whether any further action is required.

Do not use to report:

- adverse effects that are due to non-procedural issues;
- potential non-compliance (report by other means);
- issues without adverse welfare consequences.

If you are in doubt whether or not you need to fill in this form, please contact your assigned Inspector.

Please clearly title your document: **'ASRU_establishment name_PPL number_PPLh surname_date_sc18'** and send to your Inspector (e.g. using cjsm) with the subject heading 'PPL SC 18 Notification PPL [Number] – **Check Home Office's Updated Bridging Ways of Working for updated advice on email addresses and subject headings. Cjsm no longer necessary.**

Establishment Licence name	University of Cambridge (Mira)
Establishment Licence number	X81BD37B1
Name of PPL holder	Rachel Watson
PPL number	PP6298217
Protocol no	3
Protocol title	Investigation of the pharmacokinetics of test agents (non-instrumented)
Severity classification	Mild
1. Date of incident	04/10/2023

2. Species	Rat

3. Brief details of study and how the limits or constraints have been breached	The aim of this study is to examine the pharmacokinetics of 3 compounds to treat Shwachman-Diamond syndrome. In this study, the compounds (that had previously been administered intraperitoneally (i.p.) in mice at 50 mg/kg) were to be administered by intravenous (i.v.) bolus at a dose of 2mg/kg, in wild-type rats, and blood collected for subsequent pharmacokinetic analysis. On 04/10/2023 the first animal on the study was warmed in a heating cabinet set at 38°C (recorded temp 35°C) for 10 minutes, placed into a restrainer and dosed. The dose was delivered as 2 injections as the animal moved, dislodging the needle, which was then replaced into the vessel to complete the dosing. Upon removal from the restrainer the animal appeared to be unconscious and was resuscitated by the member of staff that dosed the animal. The initial thoughts of the staff were that this may have been related to the restraint. The second animal on study was then warmed and dosed with the second compound with close attention being paid to the restraint. Just before completion of the injection, the tail was felt to go limp and unfortunately upon removal from the restrainer was found to be dead. The mouse was restrained for around 10 seconds. At this point further dosing was halted, and in-person veterinary advice sought while monitoring of the first animal continued. During the monitoring the first animal appeared to be slightly subdued with an unsteady gait although not displaying any signs that would suggest the animal was in pain. The rat was still displaying these clinical signs upon inspection from the NVS at 90 minutes post-dose therefore the decision was taken to cull the animal. A blood sample was taken following culling to examine compound levels, and brain, liver, lung and heart tissue was taken to examine histologically and attempt to

	determine the cause of the issues outlined above. The same tissues were also obtained from the second animal.
4. Have you taken advice from the Named Animal Care and Welfare Officer/Named Veterinary Surgeon/others?	NVS advice was sought in real time and the animal that was resuscitated was observed by the NVS prior to culling.
5. Cause of Problem, if known	Due to the small amount of time that the animals were in the restraining device it is thought that the issues were likely related to toxicity of the substance injected rather than related to the procedure of the iv injection. No macroscopic abnormalities were noted in the brain, heart, liver or lung tissues that were taken from the animals for histological analysis.
6. Action that has been or will be taken *(For example, formulation of drug changed, supportive treatment given, study design modified). If no action needed, indicate why (for example, none required as study has been terminated and will not be repeated)*	Until further investigations have revealed a potential cause of the toxicity seen by the i.v. route in these animals no further i.v. dosing of these compounds will be undertaken. Based on the literature it is unlikely that the toxicity was due to the vehicle (5% DMSO, 40 % Transcutol, in beta cyclodextrin), but we will dose one rat i.v. and monitor closely to rule this out. Further examination of the PK of these compounds will be conducted through the oral route as this will have a slower absorption of the compounds through the GI tract (more similar to how the compound would have been absorbed from the i.p. route previously examined in mice). The first rat to be dosed with each compound will be monitored closely for a minimum of 2 hours and checked regularly throughout the day with a minimum of 24 hours before further dosing.
7. Reported by (name)	Rachel Watson
8. Date	04/10/2023

Inspector's comments and recommendations:

Comments and recommendations	
Inspector name	
Date	