Refinements to Intravenous Infusion Systems in Sprague Dawley Rats Aids in the Successful Outcome of a Complex Study Design

T.R. Gleason, A.J. Evans, J.L. Shaffer, M.N. Stamen, J.D. Bultman
Charles River, Ashland Ohio

1 ABSTRACT

The Infusion Department at Charles River Ashland was given a challenge: administer a 15 second intravenous dose to rats every ten minutes for one hour on study days 86 and 149. A surgically implanted catheter was used to allow for intravenous infusion via ambulatory tethered system. Due to the study design, several areas of concern were noted including the extended duration of the study warranting concerns for patency, the condition of the port, and social housing for animal welfare. An exteriorized port using magnetic connections was determined to be the best option with consideration given to animal growth, catheter length and tip style, security of the catheter to the port, while also allowing for social housing. The study initiated with 116 M and 116 F rats (Toxicology-60/sex; Pharmacokinetic-24/sex; Cardiovascular Telemetry-32/sex.) The animals were checked for patency and locked with 20 IU/mL heparinized saline every other week. Two animals were electively euthanized due to a compromised port. Of the surviving animals, 96% of the Toxicology animals remained bi-directionally patent until scheduled main and recovery necropsy on study days 87 and 100 respectively and 94% of the Pharmacokinetic and Cardiovascular remained bi-directionally patent until scheduled necropsy on study day 151. System refinements (catheter length, tip style and security) facilitated the exceptional patency rates, the successful dosing of the study, and allowed for social housing for most of the study.

2 METHODS

The objective of the study was to evaluate the toxicity (Tox) of a known stimulant when administered via IV infusion to Sprague Dawley rats immunized with an IM dose of test article on study days 1, 22, and 64. Cardiovascular (CV) and Pharmacokinetic (PK) data were also collected following stimulant infusion on days 86 and 149.

One hundred thirty male and 129 female rats were implanted at the vendor with polyurethane catheters placed in the femoral vein, advanced to the approximate level of the kidney and exteriorized to an external magnetic port. The rounded tip catheter was bonded to the subcutaneous pin of the port prior to implantation and ports were protected by a magnetically attached aluminum cap. Thirty-four males and 33 females also had telemetry devices implanted for cardiovascular collections.

During acclimation and between infusion dosing, the systems were checked for patency, flushed with saline, and locked with 20 IU/mL heparinized saline every other week. Once connected to the tether via magnetic connection (on the day prior to infusion), animals were maintained on saline at a rate of 0.3 mL/hr. (Figure 1)

On study day 86 (Tox, CV and PK subsets) and study day 149 (CV and PK subsets only), the animals were infused with 1 mL/kg of the stimulant for 15 seconds every 10 minutes for one hour for a total of 7 doses. Due to the unusual dosing regimen, basic (non-programable) infusion pumps were connected to the same electrical circuit and a technician had to complete the circuit at the appropriate times. The CV animals (Figure 2) were dosed in a separate animal room and personnel were not permitted in the room during dosing to ensure integrity of cardiovascular and hemodynamic data. Therefore, the technician controlling the infusion pumps did so from outside of the room.

Animals were socially housed (Figure 3) except during an approximate 2 week surgical recovery period and during the infusion dosing periods. Body weights, food weights and detailed physicals were collected weekly and animals were observed approximately 1-3 hours post dose on dosing days. Clinical pathology, necropsy and histology data were collected for the Tox animals and PK sampling and CV data were collected on days 86 and 149.

Figure 1

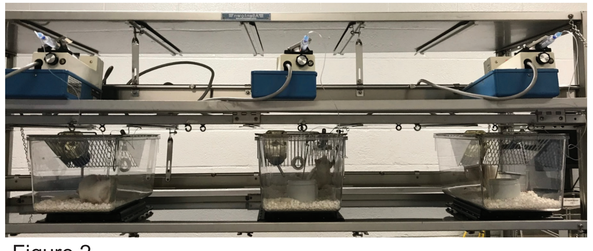

Figure 2

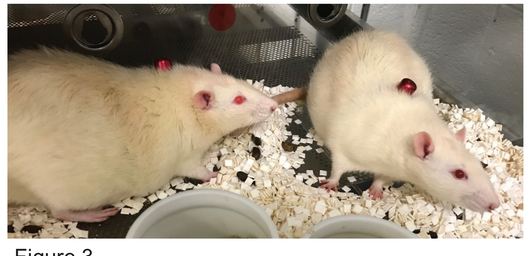

Figure 3

3 RESULTS

Infusion dosing for 15 seconds, once every ten minutes for one hour on study days 86 and 149 was successful. The set-up of the pumps allowed dosing to occur while staff was outside the study room for the Cardiovascular animals. Catheters remained patent for dosing for the entirety of the study. Bi-directional patency rates for the Toxicology animals (Table 1) and Pharmacokinetic and Cardiovascular animals (Table 2) were well above expectations at our facility.

Two animals were electively euthanized (study day 42 and 136) due to ulceration and inflammation of the skin around the exteriorized port. Other early deaths (11 across control and test article groups) were not considered to be test article-related or related to the catheters or ports. There were no test article-related effects on clinical observations. Test article-related, slightly decreased body weight gains were observed for all animals during each week following test article administration. Decreased body weight gains and decreased food consumption were also noted following the stimulant infusions. Differences in hematology, serum chemistry, coagulation and urinalysis were successfully attributable to test-article administration and not hindered by the presence of catheters or ports.

Telemetered cardiovascular data was collected on day of stimulant intravenous dosing. Any significant changes in hemodynamic data were minimal and lacked any clear dose-response relationship. Therefore, these changes were attributed to biological variation. ECG analysis of the cardiovascular waveforms was also conducted. Infrequent changes (*i.e.* premature atrial and ventricular complexes, ventricular tachycardia, AV block) were noted but determined to be a normal variant frequently exhibited in rats.

The pharmacokinetic evaluation of the implanted rats showed minimal variability between sexes and exhibited the anticipated dose response curve of the stimulant interacting with the test-article. Therefore, the implantation of the vascular access button did not result in responses from the animals that would hinder the interpretation of the study design.

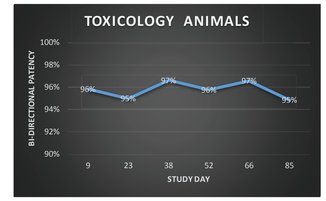

Table 1

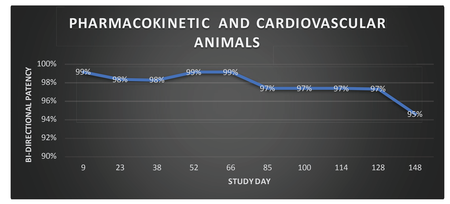

Table 2

4 CONCLUSIONS

- The use of an externalized magnetic port allowed for study required periodic infusions which would have been very difficult using a subcutaneous vascular access port and would not have been possible with the use of an externalized catheter. The metal protective cap allowed social housing during the non-infusion dosing periods.
- Bonding of the catheter to the subcutaneous pin of the port prevented catheter detachment. Use of a rounded tip catheter lessened trauma to the intimal lining of the vena cava. Catheter tip placement at the approximate level of the kidneys allowed for growth. These system refinements contributed to the successful dosing of this long term study.
- Bi-directional patency of the catheters was maintained via every other week flush/lock regimen and adherence to aseptic technique. Also, there were no clinical or gross necropsy findings or changes in white blood cell counts to indicate the presence of infection.
- The simplicity of the non-programable pumps allowed the pumps to be placed on the same electrical circuit and the 15 second IV infusion doses were successfully administered once every 10 minutes on study days 86 and 149. It also allowed for undisturbed collection of cardiovascular data.