# **Rat tickling in gestation females**

## ALICE LOMAX, DAREN LURKINS and HELEN HORNSEY

Labcorp Early Development Laboratories Limited

Correspondence: daren.lurkins@covance.com

#### Introduction

Rat tickling is a positive handling technique promoted by the NC3Rs <u>https://nc3rs.org.uk/3rs-recources/rat-tickling</u>. The website shared links to an online course from Purdue University, developed by Dr. Megan LaFollette. The course, with continuous assessment throughout, once completed successfully, provides certification in rat tickling.

Labcorp successfully trialled and adopted rat tickling in early 2021. Rat tickling was performed in most studies. However it was not recommended to do this to pregnant animals, as advised by the NC3Rs.

From the positive results seen, we then questioned if rat tickling could be used for pregnant females as this had previously been not recommended on the website.

After discussion with our veterinarians, Named Animal Care and Welfare Officer (NACWO) and the study director, agreement was given and we trialled rat tickling on an embryo-foetal study.

The objectives of the embryo-foetal developmental toxicity studies are designed to detect adverse effects on the development of the embryo and foetus consequent to exposure of the dam from implantation to hard palate closure. The pregnant dams are typically dosed on days 6 to15 of gestation.

### **Materials and methods**

In total, 28 female Han<sup>®</sup> Wistar rats were ordered at a minimum age of 11 weeks and 175g. These animals were time-mated, arriving on day 2 *post coitum* (pc).

Animal care and use was performed in conformance with the Guide for the Care and Use of Laboratory Animals in an AAALAC-accredited facility. Technicians started the rat tickling procedure 3 days prior to the commencement of dosing, on day 4 pc.

We followed the guidance from the NC3Rs' website, which at the time was to:

1. Make light quick movements with your fingers on the back of the neck for a couple of seconds (Figure 1).



Figure 1. First stage of rat tickling.

- 2. Pick the rat up for the flip, by placing your index finger in front of the rat's collar bone and your thumb and middle finger under its armpits.
- 3. Then give a flick of the wrist (Figure 2).
- 4. Once the rat is on its back, tickle its belly for approximately 15 seconds and repeat the process for a total of 2 minutes.
- Once oral gavage dosing commenced, rat tickling was performed once a week prior to the weekly detailed examination procedure.



**Figure 2.** The rat has been flipped and ready for the rat tickling procedure.

- Extra care was taken when using the flip method because of the rat's physical condition. Tickling was slightly higher on the belly especially later in pregnancy.
- Tickling procedure ceased on day 20 pc when parturition checks commenced.
- The Magenta<sup>®</sup> Bat4 (Magenta Electronics Ltd) was used to collect the results, demonstrating that gestating females responded positively to rat tickling.
- According to the NC3Rs' website, positive calls occur at 35 to 75kHz, while negative calls occur around 22kHz.
- Behaviour of each animal was also assessed to evaluate if they responded positively.

#### **Results**

Results obtained, clearly displayed that the gestation females enjoyed the interaction.

Most readings were heard at 35 to 50kHz (Figure 3).

No impact was found on the pregnant females at necropsy.

There was no impact on foetus implantation compared to background control data.



Figure 3. Results showing that most had positive reactions.

#### **Discussion**

- The females reacted positively to the rat tickling. Technicians could hear noises of chirping on the Magenta Bat4 detector at 35 to 50kHz. It was fascinating for technicians to hear these positive sounds.
- The dams also responded by licking the handler's fingers and once placed back in the cage, would run back to the handler's hand to be picked up again.
- Some females could also be heard chirping when removed from the cage prior to any procedure being performed, using the Magenta Bat4 detector.
- Technicians commented that the animals in general were very relaxed for the oral gavage procedure.
- Due to the positive findings, we have introduced rat tickling to a range of our reproductive study designs.