

Implantation of Radio Frequency Identification Transponders in Neonatal Mice

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Abstract

A previous study in our animal facility revealed that adult mice implanted in the scapular region with a next generation Radio Frequency Identification (RFID) transponder experienced minimal transponder migration, loss, and tissue reactivity. With a 60% reduction in transponder size from the previous generation, our facility explored the feasibility of using micro-transponders for permanent identification of neonatal mice on day 1. A less invasive method of identification than toe clipping, the transponders provide permanent reliable identification, and increase efficiency of long term neonatal to adult dosing. A small pilot study was conducted post mortem on mouse pups to determine if implantation was feasible. Repeated practice on dead animals was performed to improve skin handling technique, rostral vs caudal implantation approach, and technician hand positions. Once a suitable method was developed, micro-transponders were implanted subcutaneously into 1-day-old, 61 BALB/c mice (25 males and 36 females) under cryoanesthesia. An additional 17 male and 14 female mice were used as controls for growth rate comparison. Surgical glue was used in closing the implantation site wound to prevent transponder loss. Mice were monitored daily and weighed weekly for signs of rejection from the mother, transponder loss, or unexpected complications, until weaning. Weekly body weights and daily health monitoring of mice occurred until their scheduled endpoint at either 30 or 90 days post implantation. At the planned endpoint, transponders were collected along with the surrounding skin and subcutaneous tissue for histopathology. Overall, results indicated that there was no maternal rejection or death of any pups post implantation, minimal migration of the transponder, and mice tolerated transponders with minimal tissue reactivity for the duration of the study. Implanted mice grew at the same rate as control animals. The use of these micro-transponders appears to be a well-tolerated method for permanent electronic identification of neonatal mice as early as day 1.

Background

Working with our RFID vendor a new 60% smaller version of the RFID transponder was created. A previous study in our lab showed no adverse findings in adult mice implanted with the smaller transponder.

The next step was to see if implantation was possible in neonatal mouse pups as early as 1 day of age. To refine methods of permanent identification and avoid digit amputation.

Study Objectives

- To determine feasibility of implanting pups with transponders at 1 day of age.
- To evaluate any impact of implantation on maternal behavior toward pups.
- To evaluate cutaneous histopathology for unexpected lesions or inflammatory reaction post implantation.

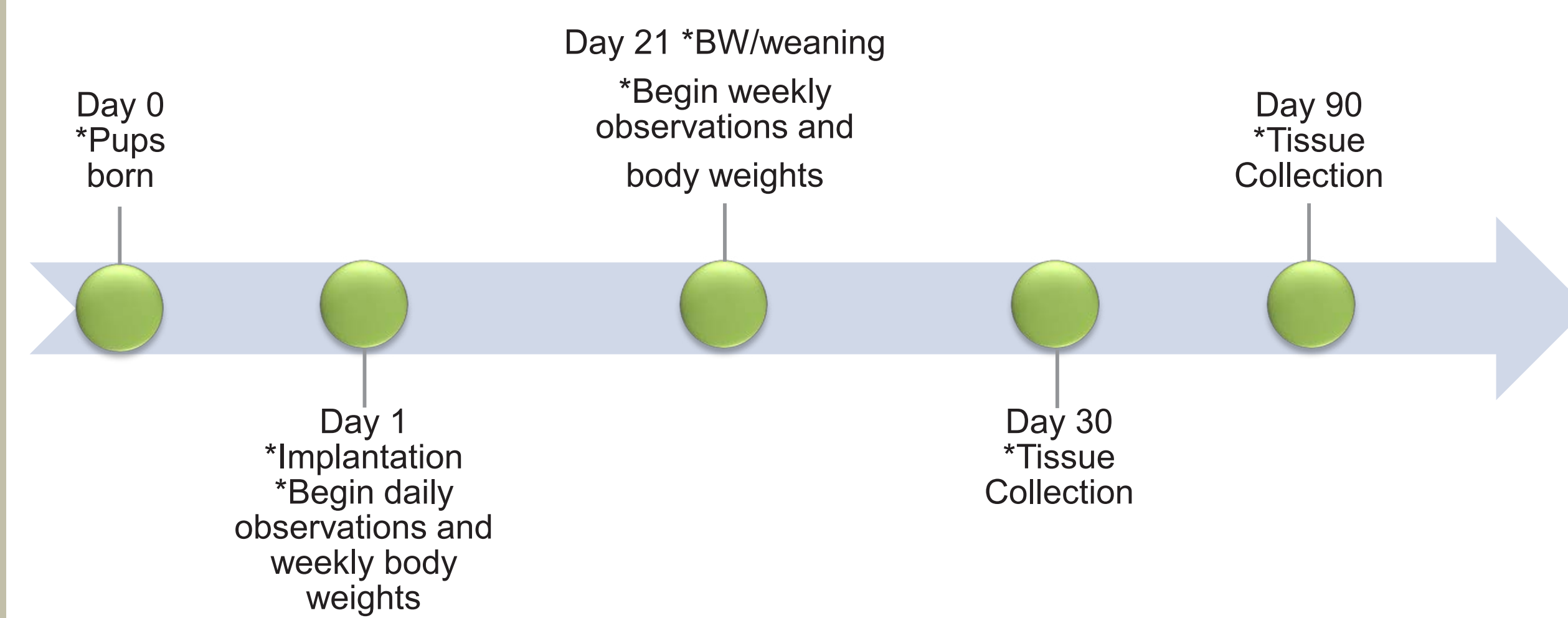
Training

- A small pilot study was conducted on post mortem mouse pups with the smaller transponders to determine if implantation was feasible.
- Repeated practice on dead animals was performed to improve skin handling technique, rostral vs caudal implantation approach, and technician hand positions.
- Once a suitable method was developed live pups were implanted subcutaneously at 1 day of age with the RFID transponder.

Study Design

Group	Animal/Group	Animal Strain	Procedure
1	18 Female/13 Male	BALB/cAnNCrI	Implant RFID on day 1, euthanize day 30
2	17 Female/12 Male	BALB/cAnNCrI	Implant RFID on day 1, euthanize day 90
3	7 Female/11 Male	BALB/cAnNCrI	Control mice - BW for 30 days
4	7 Female/6 Male	BALB/cAnNCrI	Control mice - BW for 90 days

Timeline

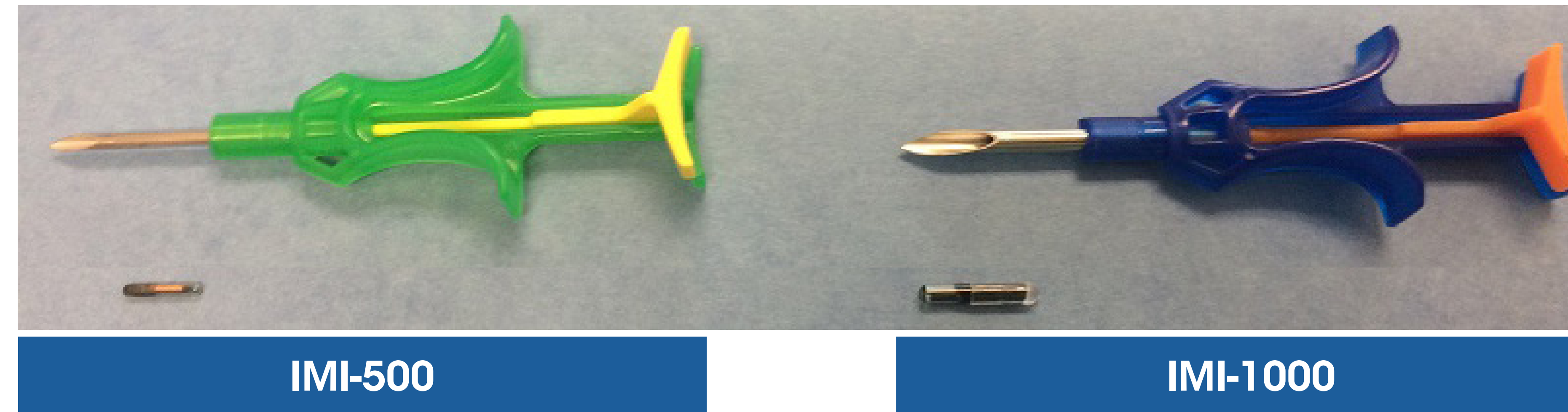


Procedure

- Cryoanesthesia was used to anesthetize pups and minimize procedural discomfort.
- To determine if the pups showed any adverse effects from the implantations the mice were observed daily and weighed weekly until weaning, with observations and body weights then taken weekly until sacrifice.
- The presence of a milk spot was confirmed for all pups 24 hours post implantation.
- At sacrifice mice were humanely euthanized and a 2 X 2cm section of skin was collected with the transponder intact and sent for histopathology review.

Materials

- 92 BALB/cAnNCrI mouse pups (50 Females and 42 Males)- Bred in-house
- RFID transponders (IMI-500 BMDS)- Size: 8mm X 1.4mm, 28.6mg
- RFID scanner (BMDS)
- Wet Ice (crushed)
- Petri-dish or weigh boat
- Surgical glue - Vetbond (3M)
- Forceps
- Scissors
- Tissue cassettes/ paper towels
- CO₂
- Scale

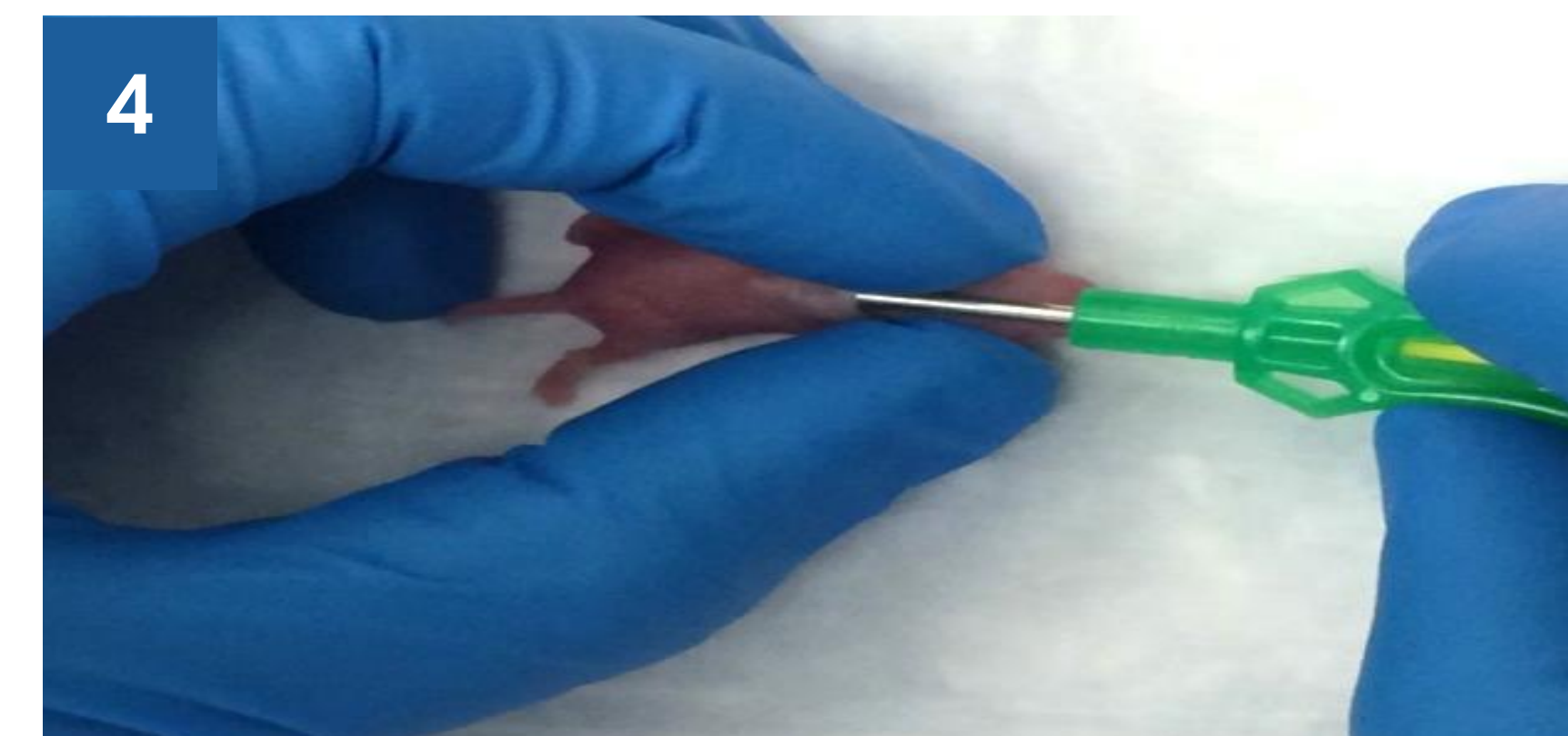
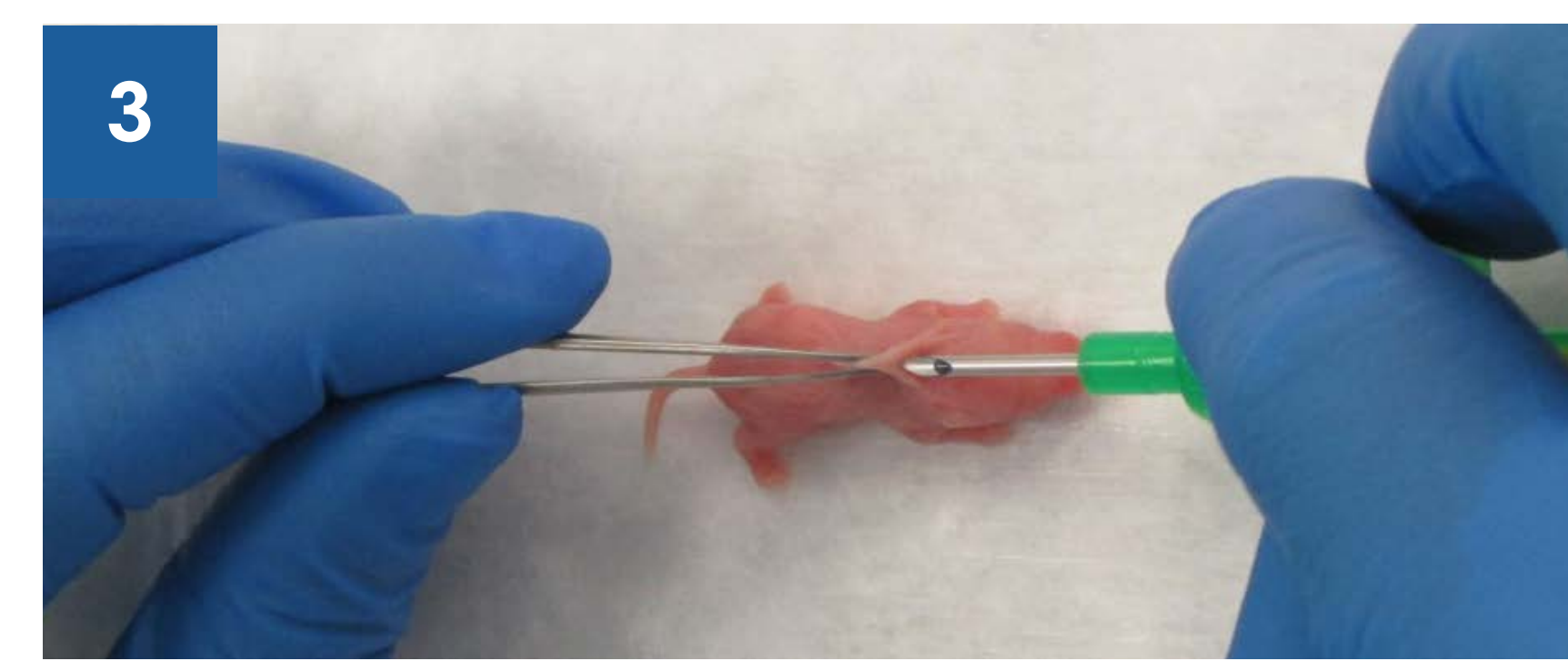
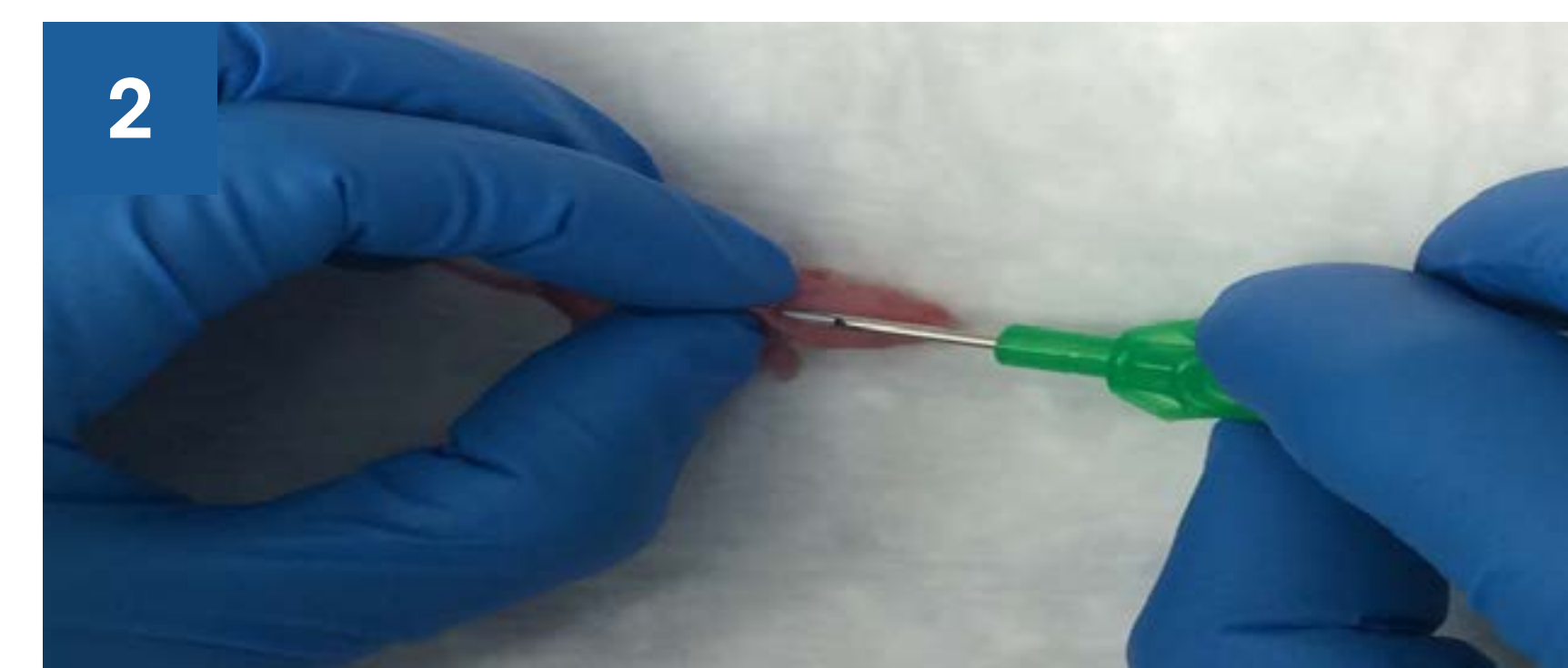


Methods

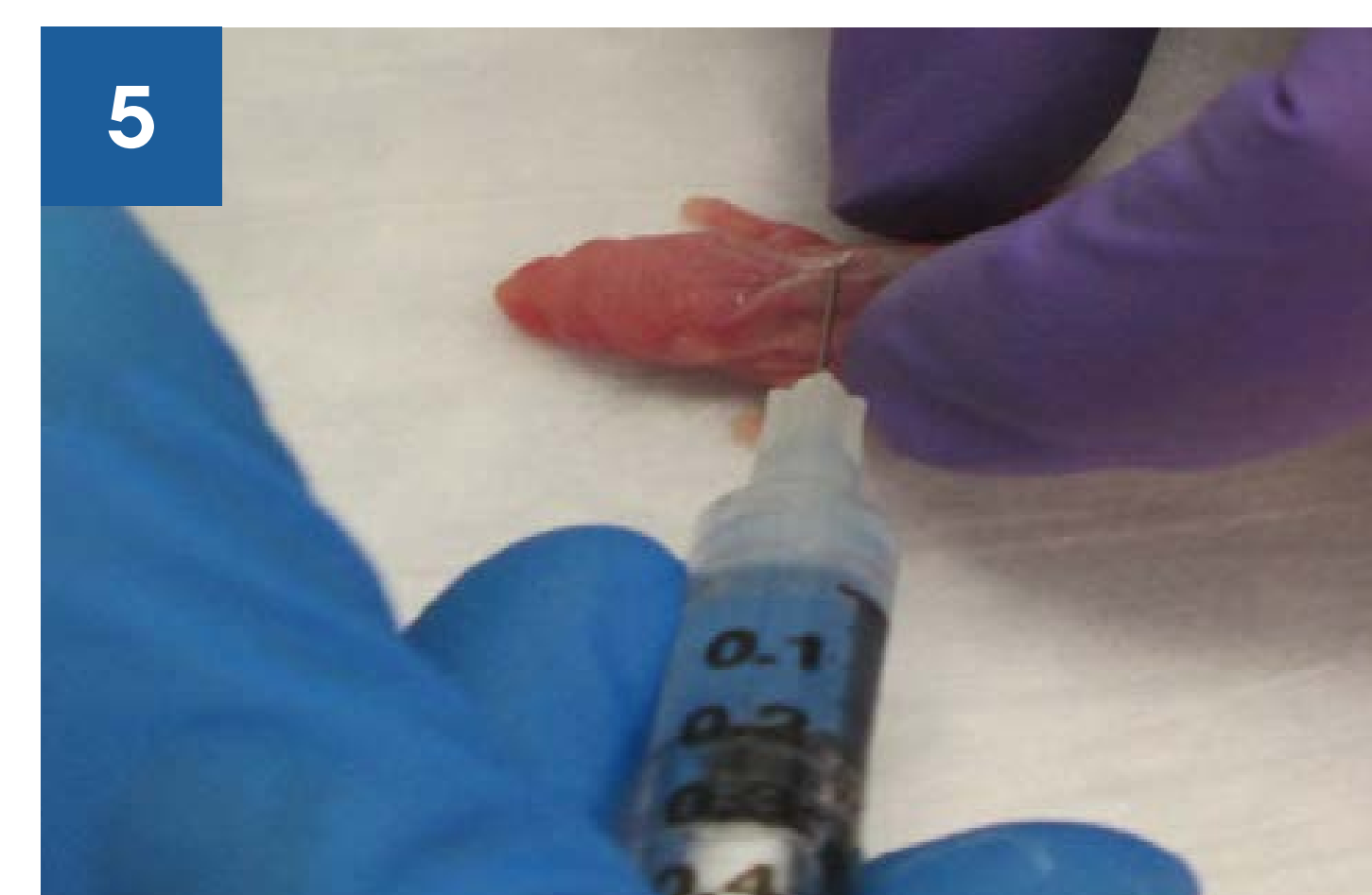
1 day old pups were anesthetized using cryoanesthesia.



The interscapular skin was tented using either fingers (Fig. 2) or forceps (Fig. 3) and the transponder was inserted into the subcutaneous space (Fig. 4) directed caudally.



A small drop of surgical glue was used on the skin at the implant site to prevent transponder loss through the insertion defect.



Following implantation pups were returned to the mother and covered with nesting material. Mice were monitored post procedure until fully recovered and throughout the day for signs of maternal rejection or other complications. The presence of a milk spot was verified for all pups 24 hours post implantation.



Mice were observed daily (Fig. 7, day 10) and weighed weekly until weaning on day 21.



Post weaning, mice were observed and weighed weekly until sacrifice. Fig. 8 (30 days) Fig. 9 (90 days).



Mice were humanely euthanized using CO₂ on day 30 or 90.

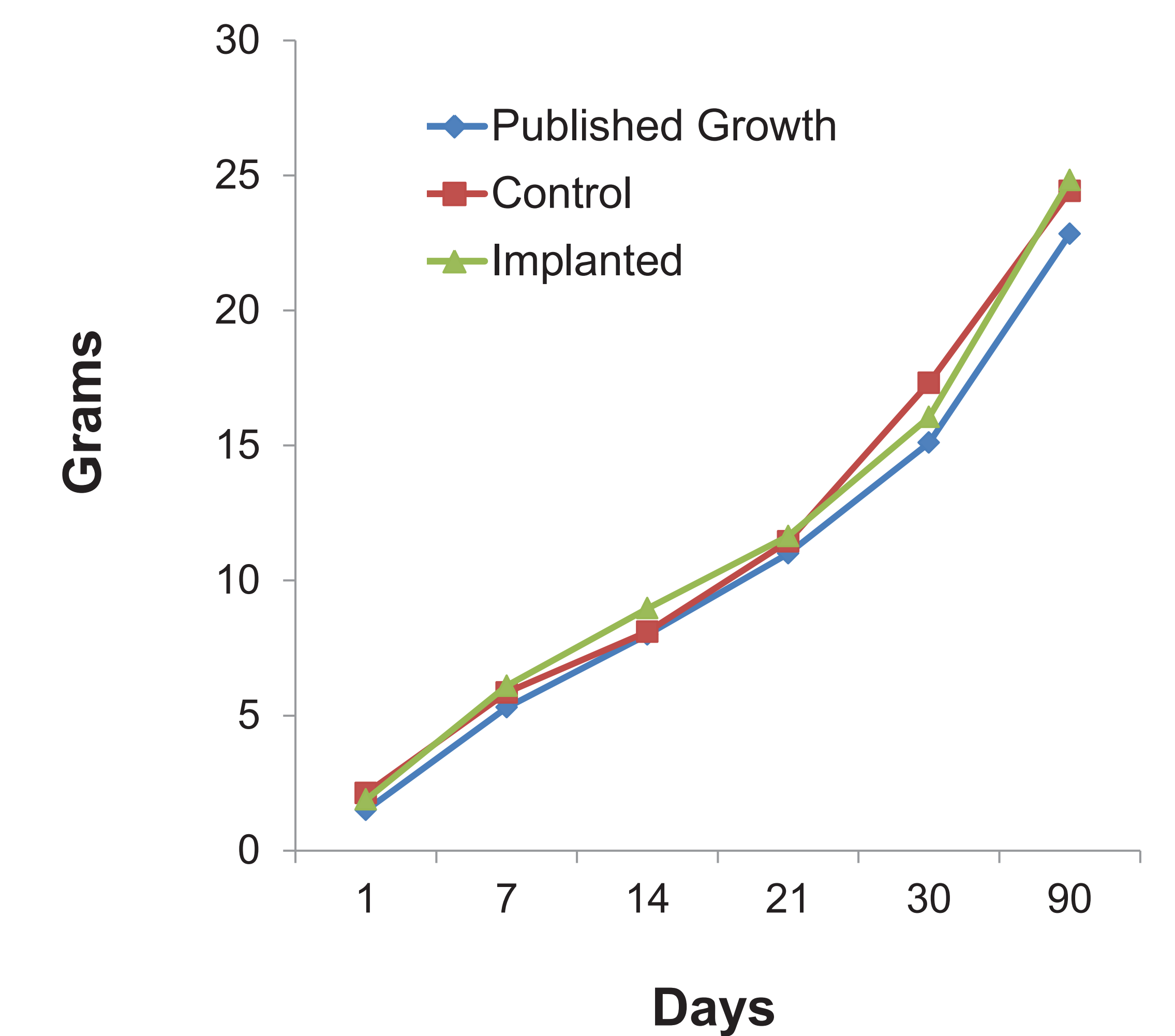


A 2 x 2cm section of skin and subcutaneous tissue was collected 10% NBF with the transponder intact and sent for histopathology review following euthanasia.



Results

- 16% of implanted transponders were not present 24 hours post implantation.
- 100% of transponders present 24 hours post implantation were present at time of euthanasia.
- No pups were rejected by the mother post implantation.
- Histopathology results were unremarkable.
- At 30 and 90 days, rare inflammatory cells were present within the dermis and subcutaneous tissue along with regional thin bands of fibrous connective tissue.
- Three mice at 30 days and a mouse at 90 days had small foci of dermal/subcutaneous tissue mineralization likely associated with the local trauma.
- There was no statistical difference in bodyweights between implanted pups, controls, and published growth data (Chart 1).



Observations

- No statistical differences were seen in the weights of the implanted mice, control mice, and published growth weights, concluding that there was no adverse effects of RFID implantation on health/weight gain of the pups.
- Migration of the transponder was not observed.
- Pathology results indicated mice tolerated transponders with minimal tissue reactivity for the duration of the study. Findings associated with local trauma and inflammation were attributed to trocar use.
- Chip loss occurred within the first 24 hours post implantation. No transponders were lost when mice on a subsequent study were implanted.
- Study technicians report more efficient identification with transponder use as compared to tattoo, toe clip or skin markers.

Conclusion

Implantation of an RFID transponder in neonatal mouse pups as early as day 1 is a reliable and efficient method for long term identification.

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