SURAT TUGAS
Nomor: 931-R/UNTAR/PENELITIAN/X/2022

Rektor Universitas Tarumanagara, dengan ini menugaskan kepada saudara:

1. **SIUFUI HENDRAWAN, dr., M.Biomed., Dr.**
2. **SUKMAWATI TANSIL TAN, dr., Sp.K.K., Dr.**

Untuk melaksanakan kegiatan penelitian/publikasi ilmiah dengan data sebagai berikut:

Judul	:	Excisional Wound Splinting Model On Rats
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Demikian Surat Tugas ini dibuat, untuk dilaksanakan dengan sebaik-baiknya dan melaporkan hasil penugasan tersebut kepada Rektor Universitas Tarumanagara

10 Oktober 2022
Rektor

Prof. Dr. Ir. AGUSTINUS PURNA IRAWAN

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Nomor dan tanggal permohonan	:	EC00202268824, 26 September 2022
Pencipta		
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Kewarganegaraan	:	Indonesia
Jenis Ciptaan	:	**Buku Panduan/Petunjuk**
Judul Ciptaan	:	**Excisional Wound Splinting Model On Rats**
Tanggal dan tempat diumumkan untuk pertama kali di wilayah Indonesia atau di luar wilayah Indonesia	:	22 September 2022, di Jakarta
Jangka waktu pelindungan	:	Berlaku selama hidup Pencipta dan terus berlangsung selama 70 (tujuh puluh) tahun setelah Pencipta meninggal dunia, terhitung mulai tanggal 1 Januari tahun berikutnya.
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Excisional Wound Splinting Model on Rats	Version: 1
	First draft: THCT
	Approved by:

Materials / Equipment:

1. Biopsy puncher Ø 19 mm
2. Hammer
3. Hair clipper
4. Blade razor
5. Surgical scissors
6. Scalpel
7. Tweezers
8. Sterile marker
9. Ruler

Solutions / Media:

1. Detergent
2. Sterile ddH_2O
3. Sodium hypochlorite (20,000 ppm)
4. 70% Ethanol
5. Povidone-iodine
6. Saline water

Consumables:

1. Nitrile gloves
2. Silicone sheet (0.5 mm thickness)
3. Sterile gauze
4. Sterile biopsy punch (Ø 8.0 mm)
5. Tegaderm™ Transparent Film (3M Health Care)
6. Super glue
7. Nylon suture 3-0
8. Non-woven adhesive tape Hypafix® (Leukoplast)
9. Cohesive elastic bandage

1. Production and Sterilization of Silicone Splint

Method is modified from Wang et al. (2013).[1]

- ☐ A silicone sheet with 0.5 mm thickness is cut into circles using a biopsy puncher (Ø 19 mm) and a hammer.
- ☐ Silicone splints are washed with a detergent and rinsed with water.
- ☐ Splints are immersed in sodium hypochlorite (20,000 ppm) for 30 mins.
  <u>Note:</u> To make 20,000 ppm (2%) of NaClO from 5.25% NaClO stock → 114.29 mL of 5% NaClO is diluted in 185.71 mL of ddH2O to make 300 mL of 2% NaClO.
- ☐ Silicone splints are rinsed with sterile ddH_2O.
- ☐ The splints are incubated in 70% ethanol for at least 30 mins; longer period of incubation could be done until 2 weeks.
- ☐ Prior to use, silicone splints are air-dried on a sterile gauze under biosafety cabinet.
- ☐ The sterile silicone splints could be stored in a sterile bottle for up to 2 weeks. For re-sterilization, repeat the process from sodium hypochlorite incubation (step 3) until finished (step 7).

2. Pre-surgery Preparation

- ☐ Animal model (outbred male Sprague Dawley rat; weighing 180-200 gr) is anesthetized using 10% Ketamine (40-80 mg/kg BW) and 2% Xylazil (5-10 mg/kg BW).
- ☐ The hair removal is performed on the dorsal and side skin of the rat using hair clipper and blade razor.

3. Excisional Wound Splinting Procedure

Method is modified from Wang et al. (2013).[1]

- ☐ The skin surface is disinfected with 70% ethanol and povidone-iodine.
- ☐ Every rat receives two full-thickness excisional wounds (side by side) on the dorsal side. Using a sterile biopsy puncher (Ø 8.0 mm), the wound sites are marked with a biopsy puncher, followed by outlining the mark using a sterile marker (Fig 1a-d).
- ☐ Full thickness skins are excised using a surgical scissor according to the marked area (Fig 1e-f). In this step, pictures are taken using a camera by including a ruler as the scale for initial area measurement (Fig 1f).

<u>Note:</u> After the excision, treatment using matrix implant or other drugs could be done into the wound area prior to the application of Tegaderm™ transparent film.

☐ The wound areas are covered with Tegaderm™ transparent film (Fig 1g-h).

☐ Super glue is spread through the surrounding edge of the silicone splint and the silicone splints are immediately placed on the Tegaderm™, fixing to the area above the wound (Fig 1i-k).

☐ Using a 3-0 nylon suture, the silicone splints are sutured through the wounded skin; sutures are done on two points of the perimeter (Fig 1l-m).

☐ To secure the silicone splints, a non-woven adhesive tape Hypafix® is applied on top of both silicone splints (Fig 1n).

☐ A cohesive elastic bandage is wrapped enclosing the surgical site around the body. Be mindful not to let the wrapping too tight (Fig 1o-p).

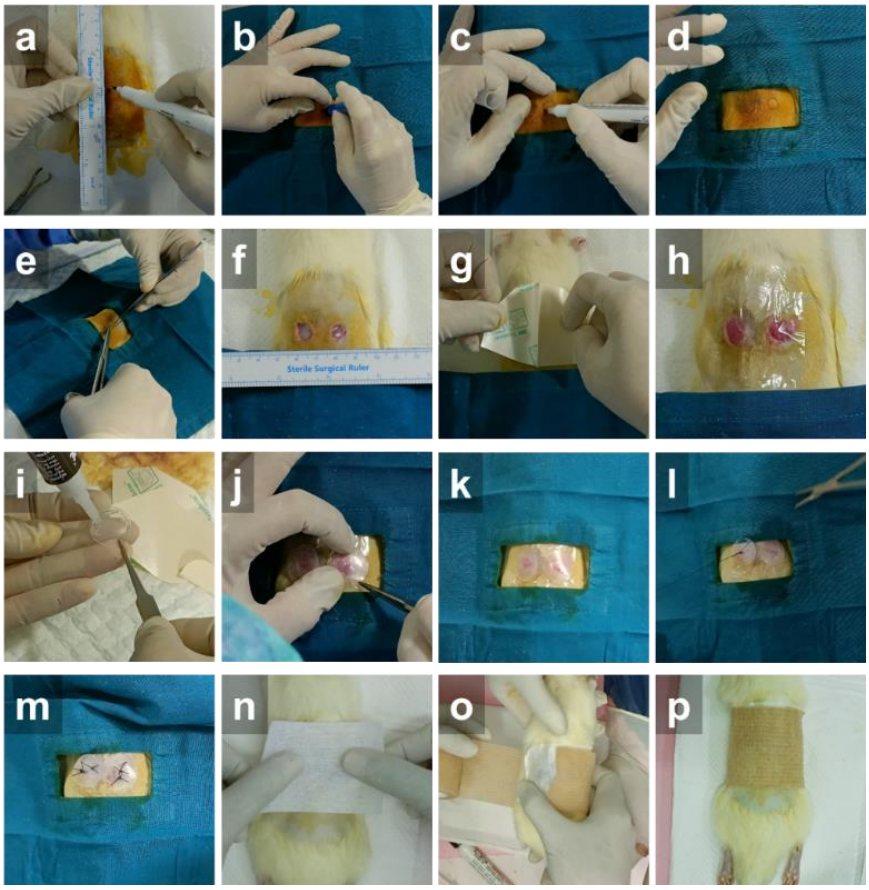

Figure 1 Excisional wound splinting protocol on a rat.

4. Post-surgery Care

☐ After the surgery, rats are placed back into their cage until fully recovered from anesthesia.

☐ Rats are caged individually to prevent wound site interruption by other individuals.

5. Measurement of Wound Area and Wound Closure Percentage

Method is modified from Wang et al. (2013)[1] and Aragón-Sánchez et al. (2017) [2].

☐ After rats are fully euthanized at endpoint, the splints and Tegaderm are removed to measure the final area of the wound.

☐ The wounds are captured using a camera, including a ruler as the scale.

☐ The wound areas are measured using ImageJ software (National Institutes of Health, Rockville, MD; http://imagej.net/ImageJ).

☐ To calculate the wound closure percentage, the following equation is applied:

$$\%\text{Wound closure} = (A_0 - A_t) / A_0 \times 100\%$$

A_0 = initial area of the wound

A_t = final area of the wound

Reference:

1. Wang X, Ge J, Tredget EE, Wu Y. The mouse excisional wound splinting model, including applications for stem cell transplantation. Nat Protoc. 2013;8(2):302–9.
2. Aragón-Sánchez J, Quintana-Marrero Y, Aragón-Hernández C, Hernández-Herero MJ. ImageJ: A Free, Easy, and Reliable Method to Measure Leg Ulcers Using Digital Pictures. Int J Low Extrem Wounds. 2017;16(4):269–73.

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Date	22 September 2022	22 September 2022	