**IMPORTANT Read All:**

- **mNSET 60010 Technical Support Letter**
- **mNSET 60010 Instructions**
- **mNSET 60010 Helpful Hints**
- **mNSET 60010 FAQs (Frequently Asked Questions)**

## mNSET (Non-Surgical Embryo & Sperm Transfer) Device for Mice 60010 Technical Support Letter

Before beginning any trials with the mNSET Device, please carefully read this Technical Support Letter, the mNSET Instructions, mNSET FAQs, and mNSET Helpful Hints (PDFs). These important support documents have proven useful for success of the mNSET device and technique. **It is of utmost importance to read these documents thoroughly.** Please contact us directly to answer any questions/comments you may have. We are always willing to help. These items, as well as mNSET poster PDFs and mNSET publications can be found on our website's mNSET Resource page, https://paratechs.com/pages/mice-nset-60010-resource-page.

### Getting started with the mNSET Device for Mice 60010:

The following information is very helpful when getting started with the mNSET device. This information is specifically important for learning the correct stage of estrous the recipient mouse should be in for traversing the mNSET tip through the cervical opening (regardless of what is transferred inside the mNSET tip into the recipient; embryos, sperm, material, or pathogens). We strongly encourage you to practice the technique several times with one of the devices without valuable embryos (or other substances) until you are successful traversing the cervix. ***Please see the red starred section on page 2 for further details.

### About the mNSET Device for Mice 60010:

This single-use plastic veterinary device is intended for research purposes only and not intended for therapeutic or diagnostic use. Each mNSET device tip is packaged with 2 specula (with same specified length, but different diameters). All 3 are sealed in a pouch and there are 10 pouches per box. mNSET 60010 is Ethylene Oxide (EtO) Sterilization processed. No further sterilization is necessary.

### mNSET 60010 Demonstration Videos

The mNSET technique as a procedure takes approximately 2 minutes. Please view the two mNSET Demonstration videos on ParaTechs' mNSET webpage, https://paratechs.com/products/nset-device-for-mice#mnset-60010-videos. The quick procedure video only covers the mouse procedure and demonstrates how easily mouse uterine embryo transfer can be accomplished using the mNSET device for mice. The full procedure video details the entire setup and preparation through the actual mouse procedure.

### Visualizing the Mouse's Cervix:

The image to the right is the cervical opening of a CD1 mouse (our recommended strain) as seen with good lighting through the wider of the two specula (as described above) that come with each device. This image is only intended as a guide to help the end-user know how to visually locate the opening before gently aiming the mNSET tip. Of course, each female mouse is a bit different, and the cervix may not always appear "centered" as seen here. Also, your hand and the P2 (recommended) pipette will block your view of the cervix during the procedure.

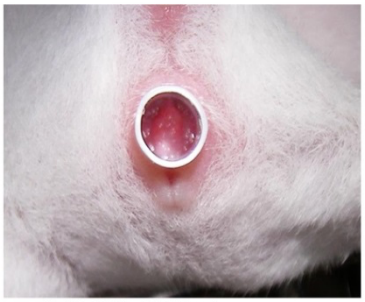

Photo by: Dr. Marcelo F. G. Nogueira
Department of Biological Sciences
FCL, Sao Paulo State University (UNESP) Campus Assis

### mNSET 60010 Recipient Mice

We recommend using CD1 female recipients starting at 8 weeks old. The litter size and pregnancy rate are improved at that age. Late-stage morulae/blastocysts are important to use since the device delivers the embryos to the uterine horn and not the oviduct. **A key factor for success is to make sure the recipient mouse is in the right stage of estrous in order for the mNSET tip to easily pass through the dilated cervix to a uterine horn. She needs to be 2.5dpc (days post coitum) pseudo-pregnant after mating with a vasectomized male, the copulatory plug has been visualized and has fallen out naturally. Do not force the plug out.**



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[mNSET (Non-Surgical Embryo and Sperm Transfer) Device for Mice 60010 Technical Support Letter continued]

***It is important to point out "No. 2" of the "mNSET 60010 Helpful Hints". We suggest the initial practice of finding and traversing the mNSET tip thru the cervical opening without valuable embryos, unless you have spare embryos for practicing. The thought behind this is that while a new end-user is getting used to the feel of the technique, you could potentially lose some embryos in the vaginal opening if applying too much pressure against the vaginal wall while "finding" the cervix. Due to the extreme flexibility of the Teflon® mNSET tip, embryos can get knocked out of the tip when too much pressure is applied and the tip bends too far. Once the mNSET device technique is practiced, the researcher/technician is better poised for successful embryo transfer.***

Gentle, calm, repeated attempts to locate the cervix is of most importance. If the tip simply won't pass through after several tries, place her back in the cage and begin with another 2.5dpc pseudo-pregnant female. As you are most likely aware, if the end-user exerts nervous energy it will transfer to the mice and could negatively affect their calm behavior and thus the trial(s) and results.

When depressing the pipette plunger to the first stop to expel the embryos, we suggest counting to 3 and then slowly removing the device and then the speculum from the mouse. However, do not release the plunger until after the mNSET tip has been completely removed from the mouse. If released before removal, the suction could pull your embryos back into the device tip.

ParaTechs does not recommend using the mNSET device for more than one transfer. Repeated use will clog the mNSET tip/catheter with cervical tissue from the mouse reproductive tract. Reuse also renders the catheter pliable and no longer rigid enough to pass the cervix, thus potentially depositing embryos in the vagina without the end-users knowledge of this occurring. When the device is used multiple times there may be a noticeable drop in success rate.

During LabRoots/VetBio Institute's 2014 LAS BioConference Live, Dr. Stone gave an "Industry Track Presentation" on the NSET device and procedure, "The Future of Mouse Embryo Transfer: Achieving the 3Rs with the NSET Device". That PDF can be found on our mNSET Resource Page, https://paratechs.com/products/nset-device-for-mice - mnset-60010-webinar. Watch the webinar, https://www.labroots.com/webinar/the-future-of-mouse-embryo-transfer-achieving-the-3rs-with-the-nset-device.

**(AI) Artificial Insemination with the mNSET Device 60010**

Dr. Barbara Stone, a Senior Research Scientist and the Director of NSET Technology, has published a technical report in Transgenic Research, https://link.springer.com/article/10.1007/s11248-015-9887-3, "A rapid and effective nonsurgical artificial insemination protocol using the NSET device for sperm transfer in mice without anesthesia", the article is Open Access. If you are interested, and at your request, we can send you the actual protocol Dr. Stone has developed.

**Material or Pathogen Transfer with the mNSET Device 60010**

Innovative researchers have developed a protocol for the novel use of the mNSET device for material or pathogen transfer to the uterine horn of female mice. If you are interested in this protocol, please email your request to info@paratechs.com.

We want your trials to be successful. As stated earlier, please don't hesitate to contact us for any technical mNSET 60010 support and we certainly look forward to hearing from you. Thank you again for your interest in the Non-Surgical Embryo Transfer Device for Mice 60010.

Revised: April 2021

**The mNSET™ Device 60010 is manufactured in the USA by an FDA Registered Medical Device Manufacturer and ISO 13485:2003 registered company and is EtO (Ethylene Oxide) sterilization processed. Patent Information: Non-Surgical Embryo Transfer Method and Apparatus, United States Patent 9,615,903.**



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