mNSET™ Device for Mice 60010 FAQs

FOR TECHNICAL SUPPORT
Please Call +1 (859) 317-9213 or
Email info@paratechs.com

IMPORTANT! Before use with the mNSET Device 60010 please read carefully:

- **mNSET 60010 Instructions**
- **mNSET 60010 Technical Support Letter**
- **mNSET 60010 FAQs**
- **mNSET 60010 Helpful Hints**

mNSET Device for Mice 60010 Technical Support Documents can be found on our web resource page:
https://paratechs.com/pages/mice-nset-60010-resource-page

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

The following Frequently Asked Questions are from our customers, technicians, and scientists which have proved helpful.

1. **Can I reuse the mNSET device to transfer embryos into multiple mice?**
2. **What mouse strain should I use as my embryo recipients?**
3. **What day post coitum (dpc) should the recipient female mice be?**
4. **What developmental stage should my embryos be for transfer?**
5. **Is anesthesia required to perform the mNSET procedure?**
6. **Should I use a lubricant during the mNSET procedure?**
7. **I'm having trouble locating and passing through the cervix. What should I do?**
8. **How many embryos should I transfer into each recipient mouse?**
9. **I performed a non-surgical embryo transfer, but when I removed the mNSET device the catheter was bent. What happened?**
10. **Can I use mice multiple times as embryo recipients?**
11. **Can the mNSET device be used for Artificial Insemination (AI)?**
12. **Does the mNSET device have other applications?**

1. Can I reuse the mNSET device to transfer embryos into multiple mice?
ParaTechs does not recommend using the mNSET device for more than one transfer. Tissue from the mouse reproductive tract tends to clog the catheter. Reuse also renders the catheter pliable and no longer rigid enough to pass the cervix, thus depositing embryos in the vagina. When the device is used multiple times there may be a noticeable drop in the success rate of embryo transfer.

2. What mouse strain should I use as my embryo recipients?
ParaTechs recommends **CD-1 or ICR mice**. For best results, the recipients should weigh ≥ 26 g and be over 60 days old.

3. What day post coitum (dpc) should the recipient female mice be?
We strongly recommend the use of **2.5 dpc pseudopregnant mice** for both training purposes and embryo transfers using the mNSET device. It is possible for the mNSET device to pass through the cervix of a pseudopregnant mouse 1.5 dpc. However, the success rate of embryo transfer at that time has shown to be lower than using the recommended 2.5 dpc pseudopregnant female.

4. What developmental stage should my embryos be for transfer?
Embryos should be in a later developmental stage than the reproductive tract of the pseudopregnant female. For instance, **blastocysts** (e3.5 days after fertilization) should be transferred into a 2.5 dpc pseudopregnant recipient.

ParaTechs Corporation • 1122 Oak Hill Dr • Lexington KY 40505 USA
www.paratechs.com **info@paratechs.com**
Tel. +1-859-317-9213 **Fax. +1-859-317-9452**

5. Is anesthesia required to perform the mNSET procedure?
No. ParaTechs does not recommend the use of anesthesia. A calm conscious animal can be positioned so that the mNSET device catheter can easily pass the cervix. The mouse in the demonstration video on our website is not anesthetized: https://paratechs.com/collections/art-devices/products/nset-device-for-mice#mnset-60010-videos.
Using an unanesthetized mouse also makes the procedure faster and easier while eliminating the risks and stress of anesthesia.[1] Anesthesia may be helpful for training purposes but need not be used under ordinary conditions.

6. Should I use a lubricant during the mNSET procedure?
No. Lubricants can clog the mNSET catheter and prevent the correct placement of embryos in the uterine horn. The specula may be moistened with sterile water or culture media prior to insertion, but even this is unnecessary. If moistening the specula, be sure to shake off any excess moisture before inserting the devices into the vagina.

7. I'm having trouble locating and passing through the cervix. What should I do?
Be sure that your recipient female is 2.5 dpc pseudopregnant. Use gooseneck lighting to inspect and locate the cervical opening before inserting the mNSET catheter, as this will help you position the device correctly. It may be helpful to practice passing through the cervix of 2.5 dpc pseudopregnant females before attempting embryo transfer.

8. How many embryos should I transfer into each recipient mouse?
For most transfers, ParaTechs recommends transferring **12-20 embryos** to each recipient mouse. (Note: optimal number of embryos to transfer will vary depending upon mouse strain and manipulations embryos have received.)

9. I performed a non-surgical embryo transfer, but when I removed the mNSET device, the catheter was bent. What happened?
A bent catheter likely means that the catheter did not enter the cervix or that you applied too much pressure while trying to locate the cervical opening. If the catheter is bent, it is unlikely that the embryos were deposited into the uterine horn of the mouse. **It is important to use gentle pressure when locating the cervical opening.**

10. Can I use mice multiple times as embryo recipients?
Studies by ParaTechs have shown that it is possible to perform multiple mNSET procedures on a female recipient and obtain up to three litters. However, there was a decrease in pregnancy rate and embryo transfer efficiency after the first litter.

11. Can the mNSET device be used for Artificial Insemination (AI)?
Yes. The mNSET device can also be used to deliver sperm to a recipient female mouse to facilitate AI.[2] Please see the article below by Stone et al. (2015). Please email us (info@paratechs.com) if you would like to receive the protocol.

12. Does the mNSET device have other applications?
Yes. The mNSET device can also be used as a novel method for effective transfer of substances for studies of uterine physiology and bacterial infection.[3] Please email us (info@paratechs.com) if you would like to receive more information.

[1]Steele KH, Hester JM, Stone BJ, Carrico KM, Spear BT, Fath-Goodin A. (2013). Non-surgical embryo transfer device (NSET) is less stressful than surgery for embryo transfer in mice. JAALAS. Jan; 52(1): 17-21.
https://aalas.publisher.ingentaconnect.com/content/aalas/jaalas/2013/00000052/00000001/art00004.

[2]Stone BJ, Steele KH, Fath-Goodin A. (2015). A rapid and effective nonsurgical artificial insemination protocol using the NSET™ device for sperm transfer in mice without anesthesia. Transgenic Research Associated with the International Society for Transgenic Technologies (ISTT) 2015 :9887 DOI: 10.1007/s11248-015-9887-3. https://link.springer.com/article/10.1007/s11248-015-9887-3.

[3]Gondek DC, Olive AJ, Stary G, Starnback MN. (2012). CD4+ T cells are necessary and sufficient to confer protection against *Chlamydia trachomatis* infection in the murine upper genital tract. J Immunol. Sep; 189(5): 2441-9. Epub 2012 Aug 1.
https://www.jimmunol.org/content/189/5/2441.

[3]Barrette VF, Adams MA, Croy BA. (2012). Endometrial decidualization does not trigger the blood pressure decline of normal early pregnancy in mice. Biol Reprod. Mar 8; 86(3): 66. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3380067/.

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www.paratechs.com **info@paratechs.com**
Tel. +1-859-317-9213 **Fax. +1-859-317-9452**