PROTOCOL

Homogenization of formalin-fixed umbilical cord and placenta tissues using the Omni THq homogenizer and 12 mm Omni Tip hybrid probe.

Authors

Gabriella Ryan, M.S.
Amelia Thompson
Chase Forbes
Rodney Nash, Ph.D.

Revvity, Inc.

Summary

Every 25 minutes a Neonatal Abstinence Syndrome (NAS) baby is born (Anbalagan, 2021). NAS is a spectrum of clinical manifestations seen in neonates due to withdrawal from intrauterine drug exposure, commonly associated with maternal opioid use. The incidence of NAS has increased fivefold in the past decade.

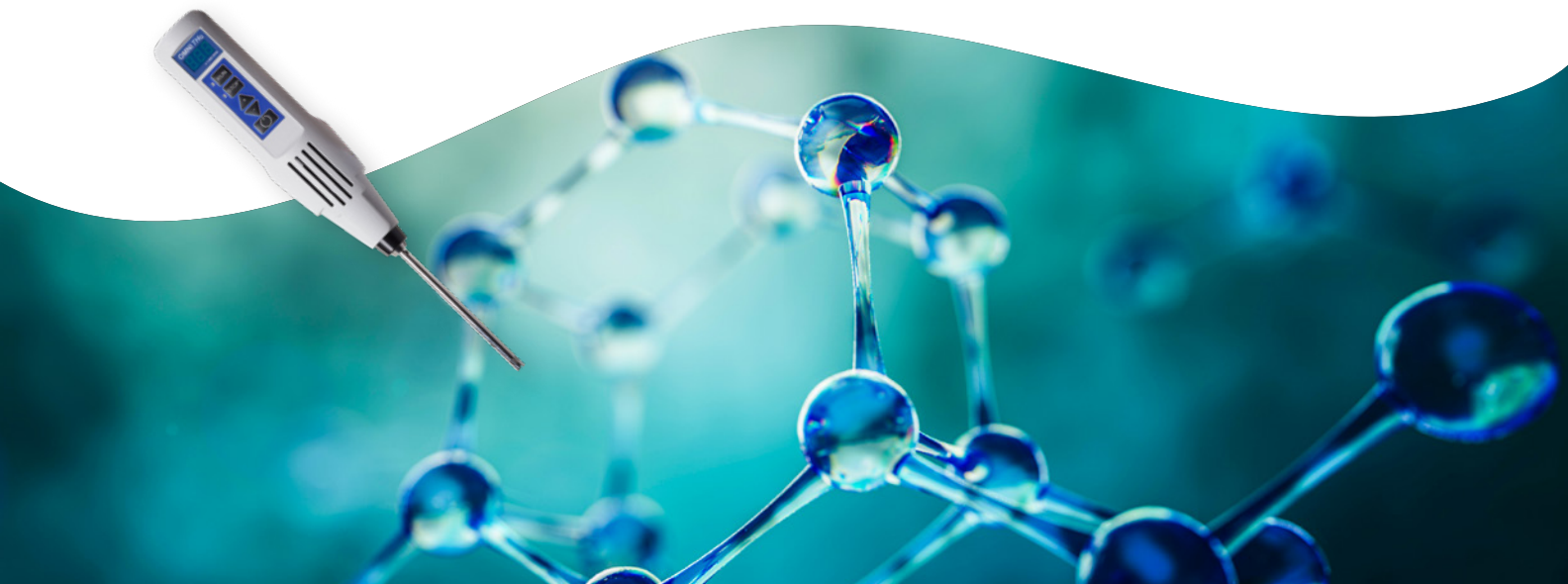
Today's substance abuse landscape is drastically different than it used to be. Testing necessitates the most advanced newborn toxicology options available on the market.

Umbilical cord and placenta tissues are employed as matrices of study in illicit drug and toxicology testing of the newborn. This is mainly due to an improved chain of custody for sample collection, compared to meconium, while still producing quality results when analyzed via LC-MS/MS (Montgomery, 2008). To prepare umbilical cord and placenta tissue for downstream workflows, the samples must be initially homogenized to disrupt the tissue cells and release analytes of interest. Traditional methods for tough sample matrices such as these involve household blenders which can be difficult to clean and may produce inconsistent results.

With the 12 mm Omni Tip™ hybrid probe and Omni THq homogenizer, sample preparation of umbilical cord and placenta is streamlined, enabling efficient homogenization with capacity to translate into an automated sample preparation platform to accommodate high sample-throughput demands.

For research use only. Not for use in diagnostic procedures.

Omni THq digital handheld tissue homogenizer



Materials and methods

Equipment

- Omni THq digital handheld tissue homogenizer (Cat # 12-500)
- 12 mm Omni Tip hybrid probe (stainless steel & plastic) (Cat # 22-012)

Procedure

Sample acquisition

De-identified paraformaldehyde-fixed human umbilical cord and placenta samples were obtained from Emory University Hospital, Tissue Procurement Lab under standard IRB protocols.

Sample preparation

Umbilical cord and placenta samples were sectioned into 0.5, 2.0 and 5.0 g pieces using a scalpel. Tissue sections were weighed out with a tolerance ± 10 mg of desired sample weight. All tissue sections were continuous, meaning that the weighed sample was not further sectioned prior to homogenization. Weighed out samples were divided into separate 50 mL tubes (Cat # 19-6650) along with 10 mL of phosphate buffered saline (PBS), pH 7.2 (Gibco, Cat # 20012027). Samples were processed with the 12 mm Omni Tip hybrid probe (Cat # 22-012) and Omni THq homogenizer (Cat # 12-500) according to sample parameters described in Table 1. To clean and drastically reduce cross-contamination between samples, the 12 mm Omni Tip hybrid probe can be placed in 2-3 consecutive water baths to dilute out left-over homogenate particles that are left on the probe after processing tissue samples.

Homogenate pipette test

After each sample was processed, 1 mL of resulting liquid homogenate was pipetted using a standard 1250 µL filter pipette tip (Gemini Bio, Cat # L1250F) and manual pipette (Mettler Toledo, Cat # PR-1000) to verify homogeneity and validate capability of downstream manual/automated liquid handling.

Table 1. Sample homogenization summary. Parameters for placenta and umbilical cord tissue.

Tissue type	Tissue weight (g)	Speed (rpm)	Time (min)
Placenta, Umbilical Cord	0.5	20,000	1
	2.0	25,000	1
	5.0	30,000	3

Results

The Omni THq homogenizer and 12 mm Omni Tip hybrid probe yielded uniform and complete homogenates when used to process umbilical cord (Figures 1-3) and placenta tissue (Figures 4-6) of varying weights. The resulting homogenate is also suitable for liquid handling using a manual pipette.

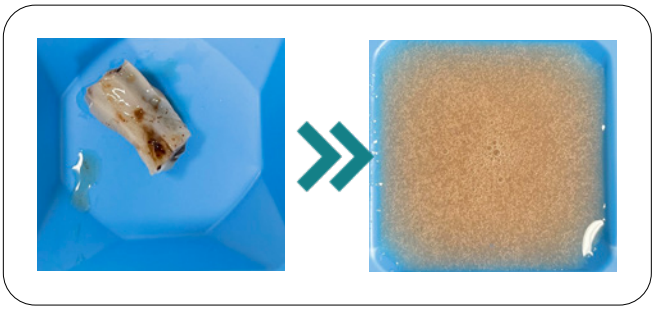


Figure 1: Pre- and post-homogenization photos of 0.5 g umbilical cord tissue

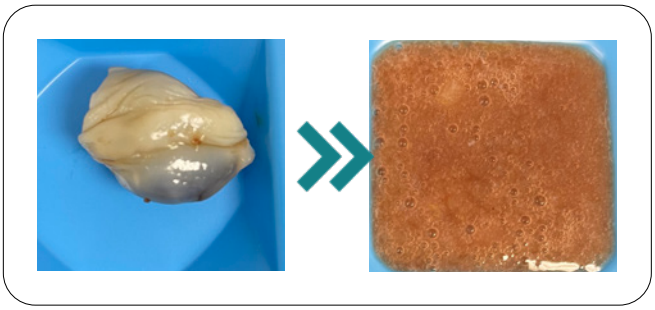


Figure 2: Pre- and post-homogenization photos of 2.0 g umbilical cord tissue

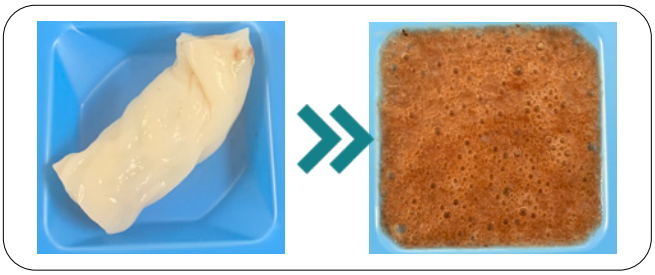


Figure 3: Pre- and post-homogenization photos of 5.0 g umbilical cord tissue

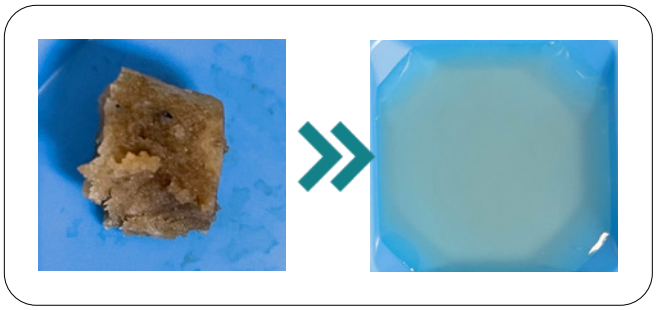


Figure 4: Pre- and post-homogenization photos of 0.5 g placenta tissue

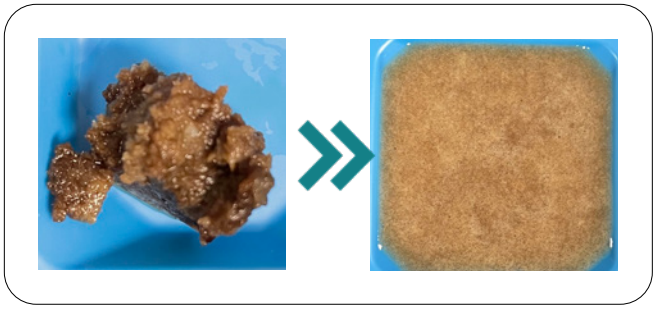


Figure 5: Pre- and post-homogenization photos of 2.0 g placenta tissue

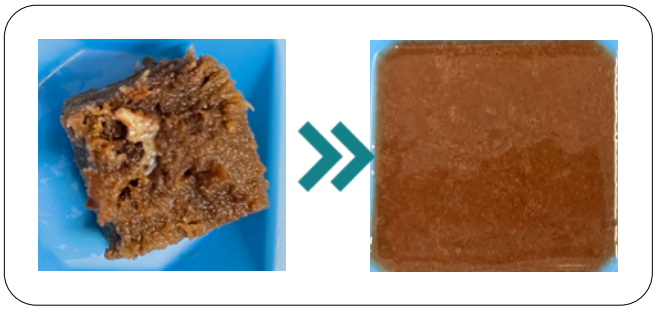


Figure 6: Pre- and post-homogenization photos of 5.0 g placenta tissue

Conclusions

The 12 mm Omni Tip hybrid probe packs a punch for sample preparation of the toughest tissue matrices, like umbilical cord and placenta. Whether processing a few samples or a few hundred samples a day, the hybrid probe fits into either workflow with the Omni THq homogenizer or Omni Prep 96 and LH 96 automated platforms, respectively. The resulting homogenate is uniform and compatible with manual pipetting techniques, indicating proof-of-concept for automated liquid handling workflows and downstream analysis methods.

References

1. Anbalagan, S., & Mendez, M. D. (2021). Neonatal Abstinence Syndrome. In StatPearls. StatPearls Publishing.

2. Montgomery, D. P., Plate, C. A., Jones, M., Jones, J., Rios, R., Lambert, D. K., Schumtz, N., Wiedmeier, S. E., Burnett, J., Ail, S., Brandel, D., Maichuck, G., Durham, C. A., Henry, E., & Christensen, R. D. (2008). Using umbilical cord tissue to detect fetal exposure to illicit drugs: a multicentered study in Utah and New Jersey. Journal of perinatology : official journal of the California Perinatal Association, 28(11), 750–753. https://doi.org/10.1038/jp.2008.97

Revvity, Inc.
940 Winter Street
Waltham, MA 02451 USA
www.revvity.com