



Helbing/Langlois Laboratory eDNA Technical Bulletin

All eDNA tools are validated through a rigorous multi-step evaluation protocol that includes tests of DNA target specificity and amplification sensitivity¹⁻³.

General eDNA Assay Information

Target Species: Common shrew (*Sorex cinereus*)
Species Code: ma-SOCI

eDNA qPCR Tool: eSOCI1
eDNA qPCR Format: TaqMan

Gene Target: MT-16S
Published in: _____

eDNA Assay Sensitivity Test Summary using gBlocks™ Synthetic DNA

LOD 0.6 95% CI 0.4-0.9 Copies/Rxn LOQ 2.2 95% CI 1.5-3.6 Copies/Rxn LOB 0 hits/8
LOQ_{continuous} 4 Copies/Rxn

Binomial-Poisson model for 8 technical replicates determined using eLowQuant R code⁴.

When the LOQ < LOD, use the LOD for the LOQ.

Enzyme: QIACuity

eDNA Assay Specificity Test Information

Each qPCR reaction in the specificity assay contained 10 picograms of voucher target gDNA (n=25 technical replicates)

| Species | Common Name (<i>Species</i>) | Detection | # Voucher | | Sample Sources/Locations |
|-----------|---|-----------|-----------|--|--------------------------|
| | | | Specimens | | |
| ma-SOCI | Masked shrew (<i>Sorex cinereus</i>) | Yes | 5 | | Quebec |
| ma-SOFU | Smoky shrew (<i>Sorex fumeus</i>) | No | 3 | | Quebec |
| ma-SOHO | Pygmy shrew (<i>Sorex hoyi</i>) | No | 3 | | Quebec |
| ma-BLBR | Northern short-tailed shrew (<i>Blarina brevicauda</i>) | No | 3 | | Quebec |
| ma-SOGA | Gaspé shrew (<i>Sorex gaspensis</i>) | No | 1 | | Quebec |
| ma-HOSA | Human (<i>Homo sapiens</i>) | No | 1 | | Cell line ATCC |
| ma-CALUfa | Dog (<i>Canis lupus familiaris</i>) | No | 1 | | Quebec |
| ma-FECA | Cat (<i>Felis catus</i>) | No | 2 | | Quebec |

References

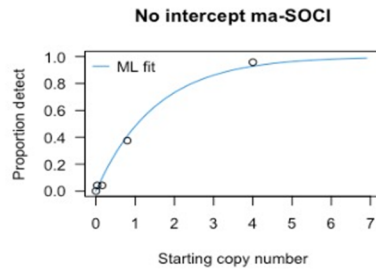
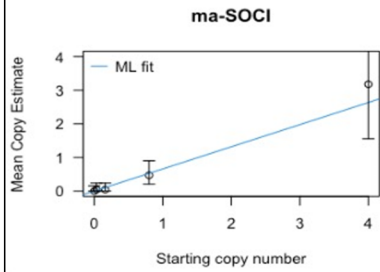
- Hobbs, J, Adams, IT, Round, JM, Goldberg, CS, Allison, MJ, Bergman, LC, Mirabzadeh, A, Allen, H, Helbing, CC (2020) Revising the range of Rocky Mountain tailed frog, *Ascaphus montanus*, in British Columbia, Canada, using environmental DNA methods. Environmental DNA, 2: 350-361. <https://doi.org/10.1002/edn3.82>
- Hobbs, J, Round, JM, Allison, MJ, Helbing, CC (2019) Expansion of the known distribution of the coastal tailed frog, *Ascaphus truei*, in British Columbia, Canada, using robust eDNA detection methods. PLOS ONE 14(3): e0213849. <https://doi.org/10.1371/journal.pone.0213849>
- Langlois, VS, Allison, MJ, Bergman, LC, To, TA, and Helbing, CC (2020) The need for robust qPCR-based eDNA detection assays in environmental monitoring and risk assessments. Environmental DNA, 3: 519-527. doi: 10.1002/edn3.164
- Lesperance, M, Allison, MJ, Bergman, LC, Hocking, MD, and Helbing, CC (2021) A statistical model for calibration and computation of detection and quantification limits for low copy number environmental DNA samples. Environmental DNA, 3: 970-981. doi: 10.1002/edn3.220



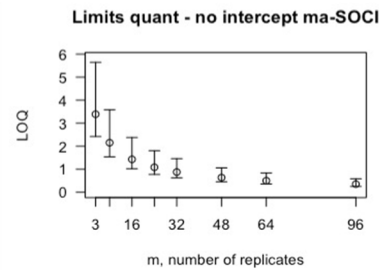
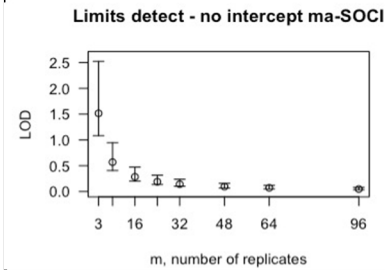
eDNA Assay Sensitivity Test Details using gBlocks™ synthetic DNA

To generate tables for different numbers of replicates, use raw csv data files.

From 8 Technical Replicates

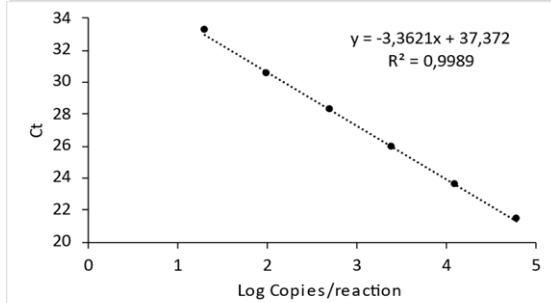


| # Detects | # Copies | SE |
|-----------|----------|-------|
| 0 | 0 | 0 |
| 1 | 0.203 | 0.207 |
| 2 | 0.437 | 0.322 |
| 3 | 0.713 | 0.44 |
| 4 | 1.052 | 0.578 |
| 5 | 1.488 | 0.756 |
| 6 | 2.104 | 1.023 |
| 7 | 3.156 | 1.558 |



Determined using eLowQuant R code⁴.

Applied to reactions with ≥ 95% positive hits



Efficiency 98%

Binomial-Poisson model: No intercept

Determined using eLowQuant R code⁴.

Based on a 2 µL DNA input in a total 15 µL reaction

Field Sample Validation

| Sample Type | Known | | Detected | Location |
|-------------|----------|-----------|----------|---|
| | Presence | # Samples | | |
| Soil | Y | 1 | Y | Parc national de la Jacques-Cartier, Quebec |

Abbreviations

| | | | |
|--------|---|--------|--|
| 95% CI | 95% Confidence interval | LOQ | Limit of quantification |
| eDNA | Environmental DNA | MT-16S | Mitochondrial 16S ribosomal RNA |
| gDNA | Total genomic DNA extracted from voucher specimen | NTC | qPCR no template control |
| LOB | Limit of blank | qPCR | Quantitative real-time polymerase chain reaction |
| LOD | Limit of detection | SE | Standard error |