Helbing/Langlois Lab
eDNA Inventory

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 sensitivity[1-3].

General eDNA Assay Information

Target Species: Elk (*Alces alces*)
Species Code: ma-ALAL
eDNA qPCR Tool: ma-eALAL4
eDNA qPCR Format: TaqMan
Gene Target: MT-ND1
Published in:

eDNA Assay Sensitivity Test Summary using gBlocks™ Synthetic DNA

LOD 1.4 95% CI 1-2.4 Copies/Rxn LOQ 5.3 95% CI 3.8-9.2 Copies/Rxn LOB 0 hits/8

$LOQ_{continuous}$ 20 Copies/Rxn

Binomial-Poisson model for 8 technical replicates determined using eLowQuant R code[4]. When the LOQ < LOD, use the LOD for the LOQ. Enzyme: Immolase

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 (*Species*)	Detection	# Voucher Specimens	Sample Sources/Locations
ma-ALAL	Elk (*Alces alces*)	Yes	10	MFFP
ma-RATA	Reindeer (*Rangifer tarandus*)	No	10	MFFP
ma-ODVI	White-tailed deer (*Odocoileus virginianus*)	No	10	MFFP
ma-ODHE	Mule deer (*Odocoileus heminonus*)	No	5	MFFP
ma-CALUfa	Canine (*Canis lupus familiaris*)	No	1	INRS
ma-FECA	Cat (*Felis catus*)	No	1	INRS
ma-HOSA	Human (*Homo sapiens*)	No	1	INRS

References

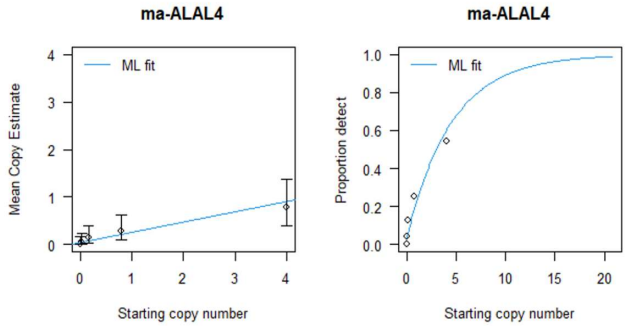
1. Hobbs, J, Adams, IT, Round, JM, Goldberg, CS, Allison, MJ, Bergman, LC, Mirabzadeh, A, Allen, H, Helbing, CC (2020) Revising the range of Rocky Mountain tailed frog, *Ascaphus montanus*, in British Columbia, Canada, using environmental DNA methods. Environmental DNA, 2: 350-361. https://doi.org/10.1002/edn3.82

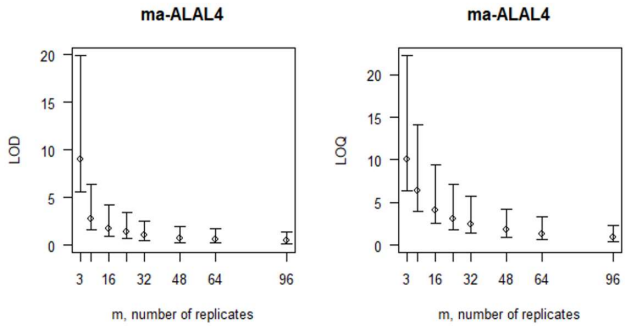
2. Hobbs, J, Round, JM, Allison, MJ, Helbing, CC (2019) Expansion of the known distribution of the coastal tailed frog, *Ascaphus truei*, in British Columbia, Canada, using robust eDNA detection methods. PLOS ONE 14(3): e0213849. https://doi.org/10.1371/journal.pone.0213849

3. 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

4. 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





Binomial-Poisson model: No intercept
Determined using eLowQuant R code[4].
Based on a 2 µL DNA input in a total 15 µL reaction

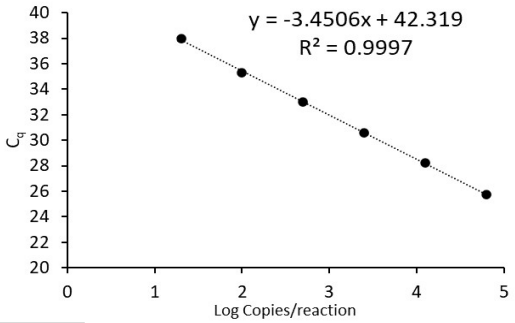
To generate tables for different numbers of replicates, use raw csv data files.

From 8 Technical Replicates

# Detects	# Copies	SE
0	0	0
1	0.502	0.514
2	1.082	0.802
3	1.769	1.098
4	2.608	1.443
5	3.591	1.89
6	5.216	2.56
7	7.816	3.892

Determined using eLowQuant R code[4].

Applied to reactions with ≥ 95% positive hits



Efficiency 99%

Field Sample Validation

Sample Type	Known Presence	# Samples	Detected	Location
Water	Y	2	Y	Refuge Pageau, Amos, Quebec

Abbreviations

95% CI	95% Confidence interval	LOQ	Limit of quantification
eDNA	Environmental DNA	MT-ND1	Mitochondrial NADH dehydrogenase 1
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