EchoLUTION Tissue DNA Micro Kit

- for single-step spin column-based purification of genomic DNA
- from 0.1 to 10 mg of fresh, frozen, or stabilized tissue

Genomic research and DNA analyses such as PCR and NGS require high-quality genomic DNA from quick procedures, no matter what the sample amount or source. The EchoLUTION Tissue DNA Micro Kit has been developed to obtain high-purity genomic and mitochondrial DNA from all kinds of tissue and from amounts of starting material as low as 0.1 mg. The purified DNA comprises of longer DNA fragments, free of contaminants and enzyme inhibitors like chaotropic reagents and organic solvents and is highly suitable for all downstream applications.

The EchoLUTION Tissue DNA Micro Kit provides

- **High DNA yield from small input amount** – nearly 100 % recovery, no loss of precious sample
- **Reliable downstream performance** in PCR and NGS – highly pure, long and intact DNA
- **High detection sensitivity** due to greater purity – no carry-over of inhibitors
- **Fast and convenient process** – complete in less than 40 min, no tedious *bind-wash-elute* procedure
- **Sustainability at lower costs** – 70 % less plastic, no hazardous liquids, plastic-free packaging

Faster preparation in half the time & fewer steps

Silica *Bind–Wash–Elute*

1. Prepare buffers.
2. Add lysis reagents to tissue sample. Mix.
3. Incubate.
4. Short spin.
5. Add buffer, vortex.
6. Add ethanol, vortex.
7. Short spin.
8. Transfer sample to column.
9. Centrifuge.
10. Transfer to new tube. Add wash buffer 1.
12. Centrifuge.
12. Transfer to new tube. Add wash buffer 2.
13. Centrifuge.
14. Transfer to new tube.
15. Centrifuge.
16. Transfer to new tube. Add elution buffer, incubate.
17. Centrifuge. DNA is in eluate 1.
18. Transfer to new tube. Add elution buffer, incubate.
19. Centrifuge. DNA is in eluate 2.

20-21. Optional 3rd elution step.

7 minutes hands-on time per sample
8 centrifugation steps
1 hour 56 minutes total time (excl. buffer preparation)

EchoLUTION Tissue Micro

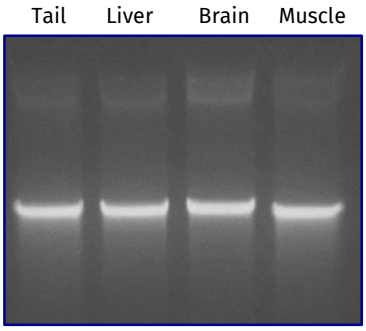
1. Add TurboLyse protease and buffer to tissue sample. Mix.
2. Incubate.
3. Add Clearing Solution.
4. Short spin.
5. Prepare column.
6. Centrifuge.
7. Transfer sample to column.
8. Centrifuge. DNA is in eluate.

3 minutes hands-on time per sample
3 centrifugation steps
37 minutes total time

The innovative EchoLUTION workflow increases the convenience, speed and performance of genomic sample preparation significantly. Genomic DNA from any tissue sample is purified with less hazzle and in a fraction of the time compared to common Silica bind-wash-elute procedures. Yields per mg starting material from EchoLUTION reactions are close to 100% due to highly efficient TurboLyse sample lysis (patent pending) and a subsequent single-step purification process, avoiding harsh adsorption, washing and desorption steps.

Consistently high yields, also from low sample amount

A



Genomic DNA was purified from fresh, frozen or stabilized tissue samples of various origin using either a leading Silica-based kit or the EchoLUTION Tissue DNA Micro Kit. Elution fractions were spectrophotometrically analyzed (Table). **A**, For DNA integrity assessment by agarose gel electrophoresis, samples were normalized to approximately 100 ng per lane. Tissues were 1 fresh, 2 frozen, 3 stabilized with PurifyLater Tissue Stabilizer, 4 with RNAlater.

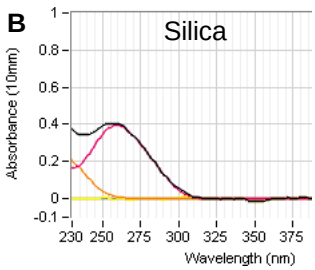
Tissue sample		Yield of genomic DNA		
		Silica µg	EchoLUTION	
			µg	Increase
Human				
Lymphatic tissue, tonsil	10 mg [1]	n/a	9 – 10	n/a
	8.5 mg [4]	n/a	5 – 6	n/a
Mouse				
Tail	10 mg [2]	2 – 4	6 – 15	3-fold
	0.1 mg [2]	n/a	0.1 – 0.2	n/a
Liver	1 mg [2]	n/a	1 – 1.5	n/a
	10 mg [2]	2 – 6	10 – 25	5-fold
Liver	10 mg [3]	8 – 15	20 – 45	3-fold
Muscle	10 mg [2]	1 – 3	3 – 6	3-fold
Brain	1 mg [2]	n/a	1 – 1.4	n/a
	10 mg [2]	1 – 4	7 – 10	3 – 7-fold
Spleen	5 mg [2]	8 – 15	15 – 25	2-fold
Rat				
Tail	10 mg [3]	7	9	1.3-fold
Liver	10 mg [3]	4.5	30.0	6.6-fold
Pig				
Muscle	10 mg [3]	0.8	5.5	7-fold
Liver	10 mg [3]	11	18	1.6-fold

Easy and efficient procedure

The initial lysis step with TurboLyse Protease takes place under physiological conditions where the enzyme activity of the proteases is maximal. All kinds of tissue can be lysed fast and successfully – simple and with the same protocol. The need for mechanical disruption lengthy optimization has been eliminated. The subsequent EchoLUTION single-step spin purification procedure combines an initial filtration, holding back potential cell debris, with an extremely efficient reverse purification. The cleared lysate is directly loaded onto the spin column. After just one single spin, the purified genomic DNA is eluted with high yields (Fig. A and Table) while all impurities and salts are completely removed. The overall procedure is highly suited for particularly precious small samples like biopsies or tissue from small animals and DNA losses are reduced to a minimum. Addition of carrier nucleic acids is not required since matrix interactions have been minimized.

Significantly greater purity

B



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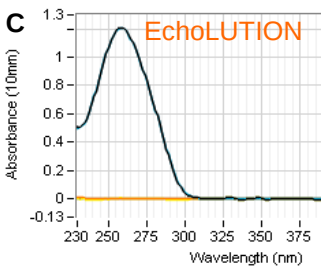


Purity assessment of Silica and EchoLUTION gDNA fractions. Undiluted DNA obtained from mouse tail tissue (**A**) was spectrophotometrically analyzed. Impurities are detected in Silica elution fractions.

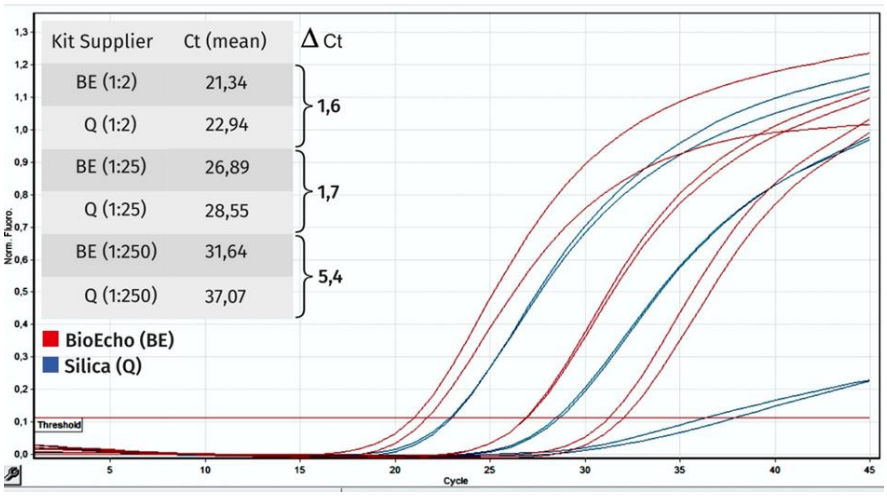
Criterion	Silica	EchoLUTION	Optimum
A_{260nm}/A_{280nm}	1.71	1.84	1.8 – 2.0
A_{260nm}/A_{230nm}	1.09	1.97	2.0 – 2.2
Yield (µg)	2.6	11.0	

The EchoLUTION workflow is based on aqueous solutions only – chaotrophic reagents such as phenol, guanidine-hydrochloride (Gu-HCl), or organic solvents are omitted. Since wash steps are never complete, Silica preps consequently contain trace amounts of these substances as impurities whereas EchoLUTION samples are highly pure. This is confirmed spectrophotometrically (OD ratios, Figure and Table above), where BioEcho eluation fractions typically show values within the optimum range. In addition, the EchoLUTION process removes all interfering substances originating from the sample which results in maximum purity and reliable OD readings.

Reliable downstream performance

DNA from tissue samples prepared with the novel EchoLUTION Tissue DNA Micro Kit is highly pure, long and intact, due to the gentle lysis and purification process. As a consequence, it is perfectly suited for downstream applications such as quantitative PCR or NGS. The purified genomic DNA does not contain any traces of chaotropic salts, organic solvents, heavy metal ions or other inhibitory impurities. A superior performance is seen in downstream applications such as real-time PCR or PCR, even with undiluted samples with the DNA solution making up to 45% of the total PCR reaction volume. This advantage can make a huge difference, e. g., for small tissue samples with corresponding low quantity of purified DNA or when highest sensitivity is required as in certain diagnostics applications.

D



Genomic DNA from 5 mg of mouse tail tissue was purified using the EchoLUTION Tissue DNA Micro Kit (red) and a Silica kit (blue). Aliquots of the obtained elution fractions were applied to qPCR detection of a mouse amplicon.

Threshold Cycle (Ct) definition:
ΔCt = 1: 2-fold recovery/sensitivity
ΔCt = 2: 4-fold recovery/sensitivity

Genomic DNA from the EchoLUTION workflow is more sensitively detected than when prepared using a Silica kit. The difference is the more pronounced the more gDNA template is applied to the amplification reaction (low dilution). The improved sensitivity stems from the higher purity and the absence of inhibitory process reagents (Gu-HCl, organic solvents) in the BioEcho procedure.

Sustainable genomic research – 70% less plastic waste

Plastic waste produced from a Silica-based DNA kit and from an EchoLUTION kit (250 reactions each; including kit components and required consumables that are not part of the kits). Bags contained in BioEcho kits are cellophane-based. In numbers:

Waste	Silica	EchoLUTION
Hazardous liquid	80 ml	0
Plastic	540 g	170 g

Ordering information

EchoLUTION Tissue DNA Micro Kit	Reactions	Product No.
For single-step purification of genomic DNA from 0.1 to 10 mg of human or animal tissue, yielding up to 40 µg highly pure DNA suitable for all molecular biology applications	10	010-002-010
	50	010-002-050
	250	010-002-250

BioEcho's growing portfolio of nucleic acid extraction kits and accessory reagents further comprises the product listet below. For availability of test kits for additional applications, please refer to www.bioecho.de.

EchoLUTION nucleic acid extraction kits	Reactions	Product No.
EchoLUTION Blood DNA Kit For single-step purification of genomic DNA from up to 60 µl liquid blood (human or animal) or dried blood (FTA cards) yielding up tp 2 µg of highly pure DNA suitable for all molecular biology applications	10	010-001-010
	50	010-001-050
	250	010-001-250
EchoLUTION CellCulture DNA Kit For purification of genomic DNA from up to $2 \cdot 10^6$ cultured cells (cell lines and primary cells), yielding up to 50 µg of highly pure DNA suitable for all molecular biology applications	10	010-006-010
	50	010-006-050
	250	010-006-250
EchoCLEAN DNA & RNA CleanUp & ReDUCE DNA or RNA concentration		
EchoCLEAN DNA CleanUp Kit (for desalting & removal of DNA/Primer < 50 bp)	50	020-002-030-050
	250	020-002-030-250
EchoCLEAN Organic Solvent DNA CleanUp Kit (for single-step CleanUp of DNA from organic solvents in just 3 minutes)	50	020-002-040-050
	250	020-002-040-250
EchoCLEAN RNA CleanUp Kit (for single-step CleanUp of RNA from organic solvents salts and chaotrophs in just 3 minutes)	50	020-002-050-050
	250	020-002-050-250
ReDUCE DNA Concentrator (for quick 20-fold volume reduction)	10	040-011-010
	50	040-011-050
ReDUCE RNA Concentrator (for quick 20-fold volume reduction)	10	040-012-010
	50	040-0112-050
Buffers and Reagents	**Volume**	
EchoSAFE FFPE Deparaffinization Solution Inert non-volatile solution enables rapid dissolution of paraffin. Allows working outside of the hood. With drop dispenser for fast and convenient dispensing.	5 ml	030-001-005
	10 ml	030-001-010
	100 ml	030-001-100
PurifyLater Tissue Stabilizer Stabilizes tissue samples for convenient storage and protection against breakdown by DNases and RNases.	100 ml	030-002-100
	500 ml	030-002-500
Lab Tools	**Pieces**	
CeraTool Ceramic Blade Scalpels (various blade shapes)	1	050-002-00x
BioEcho Cap Puncher	1	050-001-001

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