



Nucleic acid extraction and purification technologies



BIOSEARCH TECHNOLOGIES

Extraction and purification solutions from LGC, Biosearch Technologies

Biosearch Technologies is unique in the nucleic acid extraction and purification marketplace, in that we are both a service provider, and a developer of proprietary chemistries and instrumentation. We develop chemistries and instrumentation for customers to use in their own laboratories, and it is these same products which we use in each of our service laboratories. This allows Biosearch Technologies to be highly flexible, and offer individual solutions for each of our customers.

To provide the most efficient and effective extraction or purification method, we offer not only a range of different technologies but also the capability to develop tailor-made kit systems for all applications and downstream requirements.

Our range of technologies allows delivery of optimised extractions and purifications from an unrivalled array of matrices including:

- Plant material (including leaves, seeds, and other plant matrices)
- Livestock samples (including blood, animal tissue, hair follicle, buccal swabs and saliva)
- Human samples (including blood, tissue and forensic samples)
- Plasmid preparations
- Bacteria, prokaryotes and other microorganisms

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DNA extraction chemistries

QuickExtract

For DNA screening and genotyping-type applications, PCR-grade genomic DNA is sufficient, which can be rapidly and efficiently extracted using QuickExtract™ DNA Extraction Solution. In 3-8 minutes, and using only a single-tube protocol, PCR-grade genomic DNA from almost any sample type can be extracted, without the use of centrifuges or spin-columns (Figure 1).



Heat at 65 °C for 6 minutes and 98 °C for 2 minutes



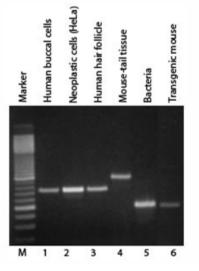
Figure 1: Procedure for obtaining PCR-ready DNA using QuickExtract DNA Extraction Solution.

The range of QuickExtract Extraction Solutions for additional sample types includes:

- QuickExtract DNA Extraction Solution
- QuickExtract FFPE DNA Extraction Solution
- QuickExtract Plant DNA Extraction Solution
- · QuickExtract RNA Extraction Solution

Figure 2: Samples extracted with QuickExtract DNA Extraction Solution. PCR was performed using primers to amplify the regions indicated: Lanes 1-3, human β-globin; lane 4, transgenic mouse GAPDH; lane 5, E. coli 16S ribosomal RNA gene; lane 6, transgenic SV40 T antigen.

QuickExtract Solution has been used to extract DNA from samples such as hair follicles, quill-end cells of feathers, tissue-culture cells, buccal cells, zebrafish organs and scales, and mouse tail snips. The extracted DNA is suitable for PCR analyses (Figure 2), such as genomic, transgenic, or viral DNA screening in animals, or for genetic or environmental research and screening in humans and other organisms.



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DNA purification chemistries

PCR-grade DNA purifications

Kleargene

Kleargene purification chemistry uses spinplates and a glass-fibre support to purify DNA using polar interactions to generate PCR-grade DNA, applicable with high throughput methods. Protocols have been optimised for both plant and animal purifications, and the Kleargene chemistry can generate yields between 100 ng and 10 µg DNA from samples ranging from plant leaf tissue and plant seeds to rodent tails (Figure 4). Kleargene has been highly optimised for automation with both 96-well formats (plant and tissue) and 384-well formats (plant), achieving higher throughput than comparable purification technologies.

The advantages of Kleargene include:

- High throughput DNA purifications (96-well and 384-well formats)
- Semi-automated (via Biosearch Technologies Genespin platform)
- Manual high throughput purifications (via centrifugation)
- Broad range of sample types (plant leaves, plant seeds, animal tissue)
- High yield of DNA (PCR-grade)

Kit	Application areas	Amount of starting material	Expected yield
Kleargene spin 96-well plates	Faster discovery of new traits resulting in faster to market. No laboratory at your	>6 leaf punches	5 µg
Kleargene spin 384-well plates	location required	2-3 leaf punches	1.5 µg

Table 1: Scales of available Kleargene kits

Next-generation sequencing-grade purifications

sbeadex

The requirement to generate high quality and quantity DNA for PCR, sequencing and next-

generation sequencing applications can be met by Biosearch Technologies portfolio of sbeadex® chemistries. All sbeadex kits use surface modified superparamagnetic particles that bind nucleic acids via a novel, two-step binding mechanism (Figure 3). Firstly, the nucleic acids affix to the beads via polar interactions, and secondly, the nucleic acids are bound via an affinity-driven mediator. This second binding step allows the final wash steps to be carried out using water-based wash solutions, eliminating the

Figure 3: Standard work flow for all sbeadex-based purification chemistry



need of a drying stage, and minimising any contaminating ethanol carryover into the final eluate. The switch between binding mechanisms also allows for greater purity of nucleic acids.

The advantages of sbeadex include:

- High-quality DNA: suitable for all genomicapplications (including NGS)
- Compatible with Biosearch Technologies automated Oktopure platform.
- Automation-friendly: suitable for most popular magnetic-based robotic platforms
- Tailor-made lysis conditions according to customer requirements
- · Quicker results: purification-time optimisation
- · Flexible batch sizes and kit volumes
- No organic solvents in final wash buffer: results in higher purity
- No salts in eluates: high OD260/230

sbeadex feature	Specification
Colour	Dark brown
Superparamagnetic bead structure	Proprietary double-coated technology (competitor: single-coated)
Size and shape	Irregular 80% 5-10 μm (diameter); a; <53 μm
Maximum capacity	2 μg DNA/mg sbeadex particle
Binding material and technology	2-step binding mechanism: first silica and second DNA adaptor
Purification format	96
Automation	KingFisher (lab validated); applicable with alternative open liquid handlers
Sample material	Blood; plant; livestock; forensics; plasmids; other human samples
Application	PCR; KASP®; BHQ® Probe chemistry; Sanger; NGS; Microarrays
Storability	>12 months

Table 2: Features and specification of sbeadex beads.

sbeadex chemistry can be used for a variety of species, by modifying the lysis conditions. Although our standard protocols already deliver high quality DNA, sbeadex can be customised and optimised according to customer requirements.

sbeadex plant

Plant samples are often challenging due to secondary metabolites, high levels of sugars and polyphenols, or robust cell walls. sbeadex plant protocols for leaf and/or seed material from a broad range of plant species have been validated and can be automated on all open platforms, including the Biosearch Technologies oKtopure. Validated sbeadex purification protocols have been developed for the following plant species (Table 3):

Plant species with validated sbeadex purification protocols				
Aphid, soybean (Aphis glycines)	Cucumber (Cucumis sativus)	Rice, Asian (Oryza sativa)		
Aubergine (Solanum melongena)	Pepper (Capsicum annuum)	Sunflower (Helianthus annuus)		
Barley (Hordeum vulgare)	Potato (Solanum tuberosum)	Tomato (Solanum lycopersicum)		
Beet, sugar	Rapeseed/Canola/ Oilseed (Brassica	Wheat (Triticale)		

Table 3: Examples of plant species, which have validated purification protocols using sbeadex chemistry.

Kit	Application areas	Amount of starting material	Expected yield
sbeadex mini plant	Plant leaves;	20 – 30 mg	2 μg
sbeadex maxi plant	Plant seeds	80 – 100 mg	10 µg





sbeadex livestock

The majority of NGS-based genomic analysis technologies (including those working with livestock samples) typically require high-quality DNA preparations which are often challenging and labour-intensive to produce. sbeadex livestock is based on one protocol which can be used for all sample types and the oKtopure can be used for all livestock samples (excluding blood).

Sample type	Animal species
Blood* Hair follicle Saliva Semen Tissue (e.g. ear punch) Buccal swab FTA cards	Bovine Chicken Dog Fish Goat Horse Sheep Pig

Table 5: Sample types from different species applicable with the sbeadex livestock kit. *Not compatible with the Oktopure.

sbeadex blood

Blood samples can often vary considerably in quality, and this can lead to wide variations in the consistency and yield of extracted DNA. sbeadex blood provides high yields of high-molecular weight, double-stranded DNA from different blood preparations including those treated with anticoagulants, including:

- EDTA blood
- Heparin blood
- · Citrate blood
- · Buffy coat preparations

Kit	Application areas	Amount of starting material	Expected yield
sbead	Whole blood; buffy coat preparations	100-200 μL	4-12 μg

Table 6: Scales of available sbeadex blood kits.

DNA/RNA/total nucleic acid purification chemistries

MasterPure

Some applications require flexibility of isolating high-molecular weight genomic DNA, total cellular RNA or total nucleic acid from the same sample. The MasterPure™ Complete DNA and RNA Purification Kit allows for isolation of NGS-grade DNA, RNA or total nucleic acid in 30–60 minutes using a single kit, and is suitable for many different sample types.

Applications for the MasterPure kit include:

- Library preparation for next-generation sequencing of genomic DNA and RNA
- DNA methylation studies (for example, with use with the Illumina® Infinium® HumanMethylation BeadChips)

- Genomic DNA and cDNA cloning
- PCR, qPCR, RT-PCR and qRT-PCR
- Microarray analysis (including CGH and gene expression profiling)

The kit uses a scalable salt-precipitation protocol which eliminates the need for hazardous chemicals (no need for phenol, chloroform or guanidine). Depending on the amount of nucleic acid required, and size of the sample, different MasterPure kits are available, including:

- MasterPure Complete DNA and RNA Purification Kit
- · MasterPure DNA Purification Kit for Blood
- · MasterPure Gram Positive DNA Purification Kit
- MasterPure Yeast RNA Purification Kit

Large volume DNA purifications

Biobanking and other human research markets require large volume purification technologies which provide high quality DNA. Biosearch Technologies

offers two cost-effective, easy and rapid methods for the purification of DNA from large volume samples (Table 9):

Features	Kleargene XL	PLUS XL
Technology	Detergent-driven cell lysis, followed by guanidine-based purification with DNA binding to silica particles. Contaminants are removed using organic solvent-based wash steps, followed by elution.	DNA cell-components are enriched, and residual proteins are digested away. The DNA is subsequently precipitated, washed, dried, and then resuspended.
Automation	Manual purifications only	Manual purifications only
Applications	NGS-grade	NGS-grade
Starting volume	Scalable: Up to 10 mL	Scalable: Up to 10 mL
Sample types	Whole blood; Saliva (including Oragene collection tubes); Tissue;	Whole blood; Saliva (including Oragene collection tubes); Tissue;
Typical DNA yields	10-800 µg	10-800 μg

Table 7: Key features of large volume DNA purification chemistries

Automation

One example for the need of high throughput molecular biology-based technologies is for plant and livestock breeders to accelerate their breeding programmes. Throughput, DNA quality, downstream application requirements, required labour and running costs are the main drivers and deciding factors when selecting high throughput automation platforms. Biosearch Technologies can offer both the oKtopure™ and Genespin™ to meet these requirements, which allows for standardised, (semi) automated, high throughput DNA purifications (Table 8).

Key features	oKtopure	Genespin
Grade of automation	Fully automated	Semi-automated
Throughput	Up to 5,000 samples/day	Up to 20,000 samples/day
Format	96-well	96- and 384-well
Applications	Plant leaf; Plant seed; Livestock (ear punch, tissue)	Plant leaf; Plant seed; Rodent tails
DNA quality	Very high (NGS-grade)	High (PCR-grade)
Downstream applications	All genomics-based applications, including NGS	PCR-based technologies, including KASP, BHQ
Replicator functions	96-well	96- and 384-well
Dimensions (cm)	170 x 68.5 x 65	170 x 68.5 x 65
Laboratory requirements	Air pressure	Centrifuge with plate adaptors
Consumables	Tip wash option	Tip wash option



Table 8: Key features of both oKtopure and Genespin automated platforms, compatible with sbeadex and Kleargene chemistries, respectively.



Purification services

We at Biosearch Technologies have developed purification for both DNA and RNA protocols from a wide range of starting materials and volumes which enable the delivery of services for most applications or custom solutions utilising our unique range of in-house technologies.

DNA purification services

We have developed highly-optimised DNA purification service protocols, starting from a wide variety of sample types, including:

- Plant (including roots, leaves and seeds)
- Blood (human; animal, FTA cards)
- Tissue (hair, saliva, liver, skin)
- Tissue culture cell preparations
- Plasmid preparations from bacterial growth cultures

Feature	Customer benefit
Flexible high-throughput laboratories utilising our oKtopure and Genespin systems.	Applicable for any project size and sample type.
High quality nucleic acid with optimal yield from column-based methods and superior purity of nucleic acid using magnetic bead-based methods.	Efficient purification of high quality DNA with the highest quantity of DNA possible.
16+ years of experience in DNA extraction and genotyping.	Trustworthy, robust, reproducible and reliable data.
Standard and rapid sample turnaround time options.	Turnaround times ranging from 2-8 weeks depending on the project needs
Convenient services like sample management, sample storage, quantification and normalisation, dispensing, and more.	Additional customisable services to suit your needs.

Table 9: Key feature of Biosearch Technologies purification services.

RNA purification services

Our RNA service laboratories in Germany can purify high quality RNA for specialised downstream processing applications. Typical sample types include:

- Blood and tissue
- RNAlater® preserved tissue
- Plant (including leaves and seeds)
- Samples collected in PAXgene® blood tubes

Overview

	QuickExtract	Kleargene	sbeadex	MasterPure
Key features	3-8 minute quick and easy extraction technology for PCR- grade DNA	Highest throughput purification by 384 spin plate format	Magnetic beads with flexible protocols and no chaotrophic salts/alcohol in final wash buffer	DNA and RNA purification from same workflow
Mechanism of action	Quick and easy extraction and direct usage	Glass fibre solid support inserted into a microtiter plate	Surface-modified superparamagnetic particlessuperparamagnetic particles	Salt-precipitation based chemistry
Products	QuickExtract DNA Extraction Solution QuickExtract FFPE DNA Extraction Solution QuickExtract Plant DNA Extraction Solution QuickExtract RNA Extraction Solution	Kleargene plant spin Kleargene tissue spin	sbeadex plant sbeadex livestock sbeadex blood sbeadex tissue sbeadex forensic sbeadex plasmid	MasterPure Complete DNA and RNA Purification Kit MasterPure DNA Purification Kit for Blood MasterPure Gram Positive DNA Purification Kit MasterPure Yeast RNA Purification Kit
Automation potential	Semi-automated (heating required)	Semi-automated (centrifugation required)	Fully automated (excluding lysis)	Semi-automated (centrifugation required)
Corresponding Biosearch Technologies instrument	Liquid handler	Genespin	oKtopure	Liquid handler
Scalability	96-well format	96- or 384-well format	Customised kits adapted in 96-well format	96-well format
Options for customisation	Adaption volume	Adaption of buffer composition	Adaption to any sample material (lysis)	Adaption volume

Table 10: Overview of extraction and purification chemistries from Biosearch Technologies.

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Order information

DNA Extraction- QuickExtract			
Part number	Volume	Description	
QE09050	50 mL	QuickExtract DNA Extraction solution	
QEP70750	50 mL	QuickExtract Plant DNA Extraction solution	
QER090150	50 mL	QuickExtract RNA Extraction solution	
QEF81050	50 mL	QuickExtract FFPE DNA Extraction solution	

Table 11: Order information for QuickExtract products.

High Volume Preparations - Kleargene and PLUS XL			
Part number	Preparations	Description	
NAP40801	10	PLUS XL (up to 10 mL)	
NAP40810	50	PLUS XL (up to 10 mL)	
KBS-1012-005	8	Kleargene blood (up to 10 mL)	
KBS-1012-006	32	Kleargene blood (up to 10 mL)	
KBS-1012-007	160	Kleargene blood (up to 10 mL)	

Table 12: Order information for Kleargene products for high volume blood samples.

Precipitation- MasterPure Complete DNA and RNA Purification Kit		
Part number	Description	
MC85200	100 RNA or 200 DNA purifications	
MC 89010	5 RNA or 10 DNA purifications	

Table 13: Order information for MasterPure products.

Spin plates – Kleargene			
Part number	Preparations	Description	
KBS-1012-201	1 x 96	Kleargene plant	
KBS-1012-202	4 x 96	Kleargene plant	
KBS-1012-210	16 x 96	Kleargene plant	
KBS-1012-211	64 x 96	Kleargene plant	
KBS-1012-204	1 x 384	Kleargene plant	
KBS-1012-205	4 x 384	Kleargene plant	
KBS-1012-212	16 x 384	Kleargene plant	
KBS-1012-213	80 x 384	Kleargene plant	
KBS-1012-400	1 x 96	Kleargene tissue	
KBS-1012-401	4 x 96	Kleargene tissue	
KBS-1012-442	16 x 96	Kleargene tissue	
KBS-1012-443	64 x 96	Kleargene tissue	

Table 14: Order information for Kleargene spin plates products.

Superparamagnetic beads – sbeadex				
Part number	Preparations	Description		
NAP41301	96	sbeadex plasmid		
NAP41310	960	sbeadex plasmid		
NAP41405	96	sbeadex tissue		
NAP41450	960	sbeadex tissue		
NAP41501	96	sbeadex forensic		
NAP41510	960	sbeadex forensic		
NAP41601	96	sbeadex mini plant		
NAP41610	960	sbeadex mini plant		
NAP41602	96	sbeadex maxi plant		
NAP41620	960	sbeadex maxi plant		
NAP44701	96	sbeadex livestock trial		
NAP44702	960	sbeadex livestock		
NAP41640	960	sbeadex maxi plant (oKtopure)		
NAP41607	960	sbeadex mini plant (oKtopure)		
NAP41606	96	sbeadex mini plant trial		

Table 15: Order information for sbeadex products

Genespin	
Part number	Description
KBS-0010-001	96 Head Genespin high-throughput Kleargene prep robot
KBS-0010-002	384 Head Genespin high-throughput Kleargene prep robot
KBS-0900-023	On-site fully inclusive service contract

Table 16: Order information for Genespin products.

oKtopure		
Part number	Description	
KBS-0009-001	oKtopure high-throughput DNA extraction robot	
KBS-0009-002	oKtowash™, concentrated wash buffer (500 mL)	
KBS-0009-003	oKtopure offline tip wash option	
KBS-0009-004	oKtopure mix plates (Thermo 1.2 mL deep well plate)	
KBS-0009-005	Wash buffer bulk reservoirs (pack of 4)	
KBS-0009-999	Extended on-site fully inclusive service contract	
KBS-0010-003	oKtopure tips	
KBS-6212-027	Magnetic plate	

Table 17: Order information for oKtopure products.



Products and services

Products

DNA extraction products (sbeadex®, mag[™] and Kleargene™ kits)

oKtopure[™] and Genespin DNA extraction systems

BHQ[®] Probes (Dual-labeled, BHQplus[®], BHQnova[™])

Enzymes and PCR reagents (KlearKall™, KlearTaq™, KlearTaq™ HiFi)

Whole Genome Amplification (WGA) kits

IntelliQube® qPCR and end-point PCR system

SNPline[™] high-throughput SNP genotyping system

Nexar® ultra-high-throughput SNP genotyping system

DNA fragment library prep kits (NxSeq® AmpFREE and UltraLow Library Kits)

NGS enzymes (NxGen® T4 DNA Ligase, T4 PNK, T4 **DNA Polymerase, Taq98 DNA Polymerase)**

Services

DNA and RNA extraction services

Sample management and project support services

KASP® SNP and InDel genotyping

All-inclusive genotyping services

Sanger sequencing

SSR to KASP SNP marker conversion service

Next generation sequencing services

Genotyping by Sequencing (GBS) services

Whole Genome Amplification (WGA)

SeqSNP targeted GBS service

For research use only. Not for use in diagnostic procedures

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GENOMIC ANALYSIS BY LGC