Extracta Product Selection Guide

Keywords: Extracta DNA Prep for PCR, Extracta DBS, Extracta Plus DNA, Extracta Plus RNA, nucleic acid extraction, DNA/RNA purification

INTRODUCTION

Nucleic acid isolation is a key step for genetic analysis. There are a wide variety of methods available on the market for DNA and RNA purification. Choosing a specific kit for extraction depends on sample type, input amount and downstream application. Extracta DNA Prep and Extracta DBS kits are suitable for crude extraction, saving time and reagents costs. Extracta Plus DNA and RNA kits are better suited for more sensitive workflows such as next-generation sequencing which require high-purity DNA and RNA as the starting input. Spin-column based nucleic acid extraction, such as the workflows utilized in the Extracta Plus DNA and RNA kits, is one such method for achieving highly-purified nucleic acid. The following guide makes it easy to determine which isolation kit is best suited for downstream needs.

Extracta DNA Prep for PCR – Get DNA Quickly

Crude DNA extraction is a quick and easy way to get DNA for use in PCR and qPCR assays. The Extracta DNA Prep for PCR is a 2-buffer system that allows for rapid extraction of genomic DNA from cells, tissues, tail snips and ear punches. Samples are incubated in Extraction Reagent at 95°C for 10-30 minutes, cooled to room temperature and then samples can either be used directly or Stabilization Buffer can be added for long-term storage. Extracted DNA is then ready for a variety of PCR and qPCR applications such as standard genotyping,[1] species identification[2,3] and CRISPR-modification verification.[4] Since the Extracta DNA Prep is a crude procedure, purified DNA may contain impurities that could potentially inhibit sensitive downstream reactions. Pairing DNA obtained from the Extracta DNA Prep kit with the ToughMix PCR and qPCR technologies allows for quick and easy analysis without inhibition due to impurities. To further speed up experimental workflows, this method is easily adaptable for processing in tubes, plates, or deep-well blocks for automation and liquid-handling workflows. Due to the crude nature of this extraction method, it is not suitable for the isolation of RNA.

Extracta DBS – Get DNA from Blood Spots

Dried blood spots (or DBS) are a quick way to collect blood samples for later analysis and are routinely used for newborn screening[5]. However, extracting suitable DNA from the storage collection cards can be challenging due to the small volumes collected and the presence of PCR inhibitors in whole blood. Extracta DBS has been specifically optimized for extracting DNA from DBS, and has also been used with other bodily fluids such as urine collected on storage cards.[6] This kit includes a single ready-to-use reagent that requires minimal processing and no additional reagents to go from a DBS punch to purified DNA. The one-reagent system is also easily scalable for high-throughput extractions of collected samples. Similar to the Extracta DNA Prep, pairing DNA extracted with the Extracta DBS reagent with ToughMix formulations ensures robust and reproducible PCR and qPCR amplification for maximized assay sensitivity.

An increasing number of newborn screening laboratories are using next generation sequencing for routine screening of target mutations. Extracta DBS has been shown to be suitable for DNA extraction for use in translational sequencing panels for cystic fibrosis.[7]

Extracta Plus DNA – Get Clean DNA

The Extracta Plus DNA kit is the ideal option for isolating genomic DNA for sensitive downstream applications. The use of spin-column technology allows for a quick method of extraction requiring only a microcentrifuge. The technology relies on binding the nucleic acid to a spin column, washing out any impurities such as salt, and then eluting purified, high-quality DNA from the spin column. This technique is recommended for downstream applications such as qPCR or library preparation for NGS technologies. Samples can be processed from a wide variety of sample types such as cell culture, fresh or frozen tissues, blood and bacteria. The extracted genomic DNA is high molecular weight and free of damage and nicks (Figure 1). This high quality genomic DNA is well suited for Illumina NGS library preparation (Figure 2) as well as long-range PCR amplification and long-read sequencing on PacBio® and Oxford Nanopore Technologies sequencers.

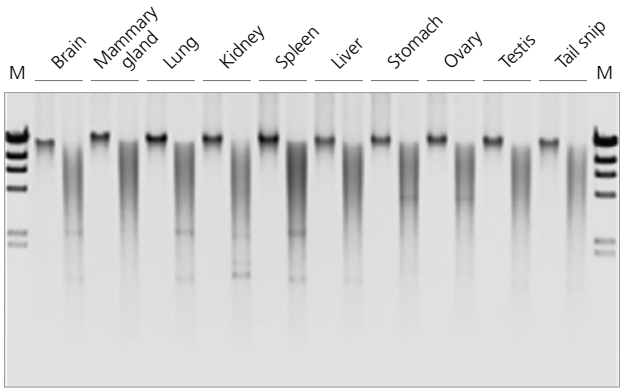

Figure 1 **Extracted gDNA of high molecular weight.** The Extracta Plus DNA Kit was used to extract DNA from a variety of tissues: rat (brain and lung), pig (mammary gland), cow (kidney), mouse (liver, testis and tail snip) and guinea pig (stomach and ovary). Samples were digested with EcoRI and visualized by agarose gel electrophoresis. **M**: Marker, HindIII digest of lambda DNA.

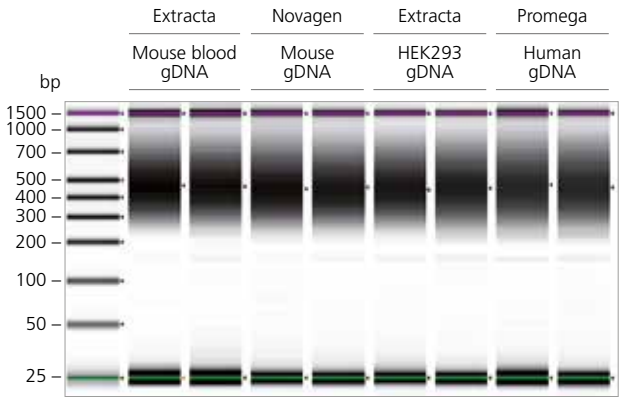

Figure 2 **Reproducible preparation of high quality NGS libraries.** gDNA was extracted from mouse whole blood (100 µl) and HEK293 cells (1 x 10^6 cells) using the Extracta Plus DNA Kit. Illumina NGS libraries were prepared from 100 ng of extracted DNA and an equivalent quantity of commercially available high-quality DNA using the sparQ DNA Frag & Library Prep Kit. TapeStation electrophoresis analysis demonstrates a similar fragmentation pattern and yield for all samples, indicating suitability for NGS library preparation.

Extracta Plus RNA – Get Clean RNA

For RNA extraction, the Extracta Plus RNA is a spin-column based methodology for clean and efficient isolation. Similar to the Extracta Plus DNA kit, samples can be processed from a wide variety of sample types such as cell culture, fresh or frozen tissues, blood and more. This kit features the Extracta Plus DNA Removal column for effective gDNA removal (Figure 3) without the need for a separate DNase I treatment. Column-based DNA removal saves valuable time and ensures that extracted RNA is ready for RT-PCR, RT-qPCR, and RNA-seq with minimal to no DNA contamination.

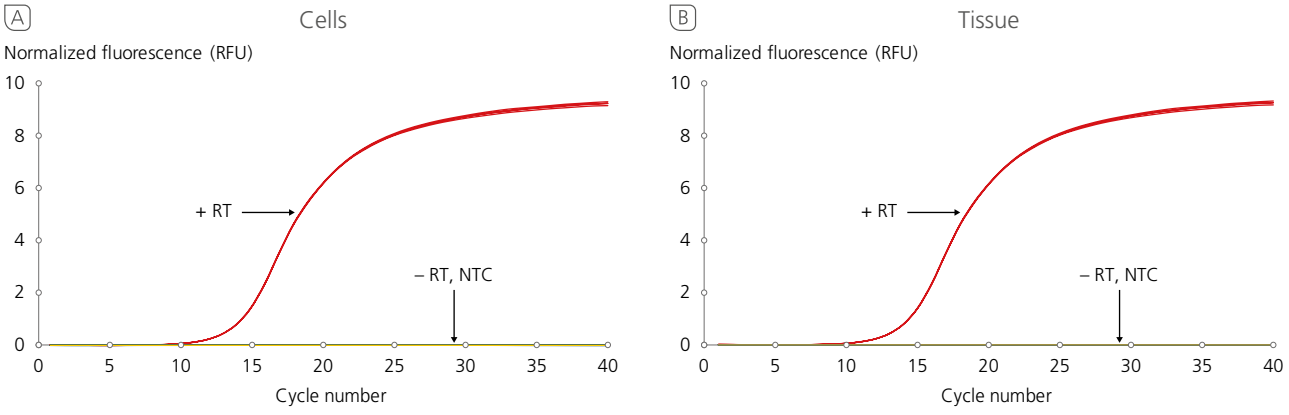

Figure 3 **Effective gDNA removal from cells and tissue.** Total RNA was purified from A 1 x 10^6 HeLa cells or B 10 mg rat kidney tissue using the Extracta Plus RNA Kit. RT-qPCR assays were performed with (+RT) or without (–RT) reverse transcriptase.

Conclusion

Choosing the right kit for nucleic acid extraction can be challenging. Extracta kits from Quantabio deliver nucleic acid ready for a variety of applications. For the fastest extraction for PCR and qPCR studies, the Extracta DNA Prep is an easy-to-use two reagent system. For samples collected on storage cards, the Extracta DBS is the best kit for easy isolation. These extraction systems work best in combination with Quantabio's ToughMix PCR and qPCR mixes to overcome any PCR inhibitors in the crude extraction for maximum assay sensitivity. For all other applications including NGS, the Extracta Plus DNA and Extracta Plus RNA kits provide high-quality DNA and RNA for sensitive downstream applications.

	Extracta DNA Prep for PCR	Extracta DBS	Extracta Plus DNA	Extracta Plus RNA
Sample Type	Cells, tissues, buccal swabs, saliva, tail snips, ear punches, hair	Dried blood spots	Fresh or frozen tissue, cells, blood, bacteria	Fresh or frozen tissue, cells, blood
Analysis	PCR, qPCR	PCR, qPCR, Sanger Sequencing, NGS	PCR, qPCR, Sanger Sequencing, NGS	RT-PCR, RT-qPCR, Sanger Sequencing, NGS
Ideal for	Rapid isolation	Samples spotted on collection cards	Sensitive downstream applications	Sensitive downstream applications

Table 1 **Extracta product selection chart.**

References

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