

Cell3™ Xtract: Cell-free DNA Extraction Kit

Fast and flexible cell-free DNA extraction kit from plasma and other biological specimens

Highlights

Fast and convenient protocol

Fast and simple protocol enables a 90 minute processing time with 45 minutes hands-on time, to extract cfDNA from a 1-10 ml volume. Requires centrifugation steps only. No specialist equipment such as magnets or vacuum manifolds needed.

Flexible kit

Flexible input volume of 1-10 ml enables increased cfDNA recovery and use of the entire sample volume.

Supports low input DNA and sensitive applications

Cell3™ Xtract technology enables an elution volume of as low as 35 µl, which helps avoid the need for DNA concentration steps and assists with low input or sensitive applications such as quantitative/real-time/digital PCR and Next Generation Sequencing.

Multi-specimen support

Extract cfDNA from plasma, cerebrospinal fluid (CSF), saliva, serum and amniotic fluid.

Accurate quantitation of cfDNA

Accurate quantitation of extracted cfDNA is important for downstream applications, particularly Next Generation Sequencing where input amounts for assays can be critical. Many companies use carrier RNA to improve cfDNA extraction which can lead to false/incorrect cfDNA quantitation. The Cell3™ Xtract kit does not require carrier RNA and therefore enables accurate quantitation of cfDNA.

Introduction

The utility of cell-free DNA (cfDNA) in both translational research and diagnostic settings has increased dramatically since its discovery. Areas of research being undertaken which involve cfDNA are diverse and include prenatal, oncology, transplantation and ageing. CfDNA is typically found in short fragment sizes of ~160 bp and varying but low quantities of 1 to 100 ng/ml in plasma.

It is therefore critical that sufficient cfDNA of good quality can be extracted for downstream applications. To address this need we have developed a simple and flexible cfDNA extraction protocol.

Quick and convenient workflow

Cell3™ Xtract has a very simple and flexible workflow which allows 1-10 ml of biological sample to be processed within 90 minutes with around 45 minutes hands on time. The flexible input volume allows for increased recovery and concentration of cfDNA while avoiding the necessity for multiple 1 ml extractions. No specialist equipment such as magnets or vacuum manifolds are required, just a microcentrifuge, a swinging bucket centrifuge capable of holding 50 ml tubes, and a heat block or water bath (55 °C).

Reduced elution volume

A flexible elution volume of $\geq 35 \mu\text{l}$ avoids the need for DNA concentration steps, which can cause additional sample loss.

Validated kit

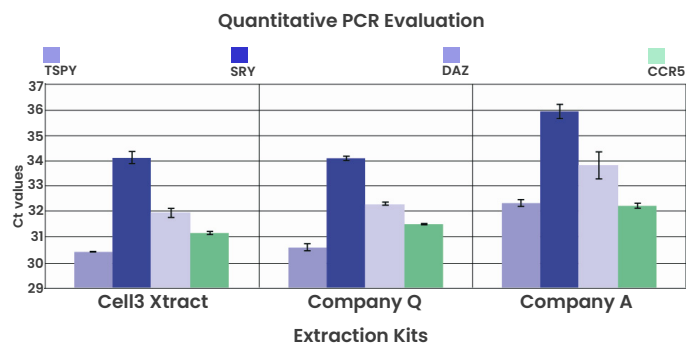
To ensure that our kit performs to our exacting standards, we compared it to manual cfDNA extraction kits from four other companies, including the market leading company (companies Q, A, O, M). These kits covered both spin column and bead based methods. For kit performance evaluation, we used plasma from pregnant women carrying a male fetus.

We extracted cfDNA from 1 ml of plasma following the manufacturer's recommended protocol and ensuring that we used the same patient sample for comparison between the Nonacus Cell3™ Xtract kit and other vendor kits.

Quantitative PCR data revealed that the Cell3™ Xtract kit performed on par with the Company Q kit and outperformed the Company A kit. However, the use of carrier RNA in the Company Q kit resulted in a higher DNA concentration measurement (~50 fold) and the appearance of a high molecular weight peak in the fragment analysis electropherogram.

In the comparison with bead based kits, the Cell3™ Xtract kit performed better than the Company M kit (based on quantitative PCR data and DNA concentration measurements) and on par with the Company O kit.

Comparison to spin column based kits



Fragment size distribution

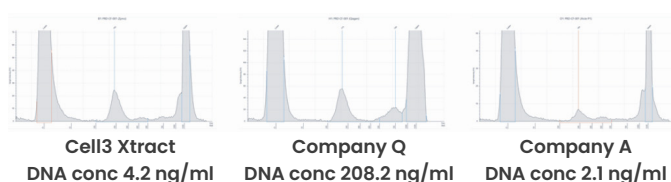
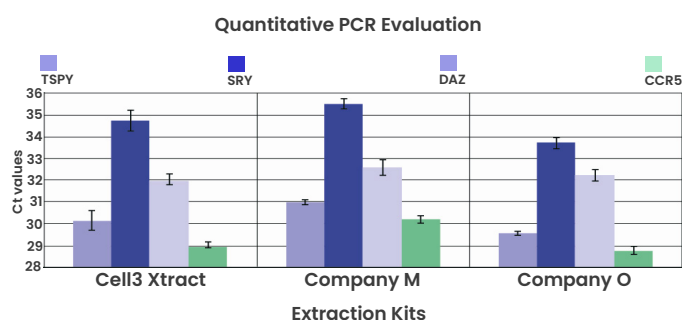


Figure 1. CfDNA was extracted from 1 ml of plasma for each kit: the same sample from a male pregnancy at 14 weeks gestation was used. Quantitative PCR evaluation was conducted to detect and compare the extracted fetal cfDNA portion using the Cell3™ Direct: Fetal Sex Determination kit (Nonacus), which includes chromosome Y specific targets (TSPY, SRY, DAZ) and a control target (CCR5). Fragment size distribution was measured using high sensitivity reagents on the Tapestation 2200 (Agilent). DNA concentration was determined using high sensitivity reagents on the Qubit 3.0 (Invitrogen).

Comparison to bead based kits



Fragment size distribution

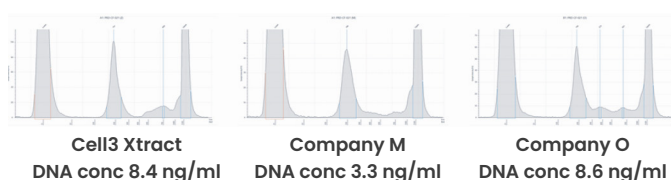


Figure 2. CfDNA was extracted from 1 ml of plasma for each kit: the same sample from a male pregnancy at 10 weeks gestation was used. Quantitative PCR, fragment size analysis and DNA concentration measurements were conducted as explained in Figure 1.

Summary

When comparing its performance to other manual cfDNA extraction kits available on the market, we have found that the Cell3™ Xtract kit delivers:

- a higher recovery of cfDNA (compared to Companies A and M)
 - a cleaner sample by not requiring the use of carrier RNA (compared to Company Q)
 - a faster workflow (compared to companies Q, A, M, O)
 - a more flexible approach to cfDNA extraction (compared to company Q)
 - a simple protocol which does not require the use of specialist equipment (companies Q, M, O)
- Therefore, we have developed a fast, flexible and effective cfDNA extraction kit which allows multiple input volumes, choice of biological input, reduced elution volume and does not require specialist equipment

Learn more

To learn more about the Cell3™ Xtract kit and to download the protocols, application notes, and white papers please visit: www.nonacus.com

Ordering information

Product

Cell3™ Xtract Kit – 16 samples
Cell3™ Xtract Kit – 48 samples

Catalogue No.

PRE_EXT_C3X_16
PRE_EXT_C3X_48

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