

## INNOVATIVE AUTOMATED NUCLEIC ACID ISOLATION BY THE KEY USE OF MAGNETIC SILICA PARTICLES

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**INTRODUCTION:** Reproducible isolation of highly pure DNA is critical to successful downstream analyses such as routine diagnostic testing, genetic screening, forensics, research and animal testing among others. Reproducibility may be increased by automating time consuming, labor intensive steps. Automation has the additional advantage of allowing for high throughput downstream analyses. We have developed three workstations, GenoM-48, GenoM-96 and GenoM-6, for automated isolation of nucleic acids from a wide range of sample types. The workstations are closed, fully walk-away systems. GenoM-48 isolates from 6-48 samples per run and GenoM-96 from 8-96 samples per run. GenoM-6 isolates from 1-6 samples per run. Sample volumes range from 25-350  $\mu$ l. The workstations use magnetic particle-based technology as the solid phase for capturing and purifying the nucleic acids. This allows for rapid washing procedures and produces highly pure nucleic acids.

**METHODS:** GenoVision has recently developed a second generation of reagents for the nucleic acid (NA) isolation which maintains the high quality of the isolated material but in general increases the total yield compared to our first products. In this poster, we would like to present results of nucleic acid isolations on the basis of this new chemistry. The examples will include NA isolations from a variety of starting materials including fresh and frozen tissue biopsy, blood, cultured cells, and dried blood from filter membrane cards.

All the isolations are based on the use of chaotropic agents (GTC, GuHCl) for lysis and binding, chaotropic agents/alcohols for washing and pure water for elution of the isolated NA.



Fig. 1: GenoM-48 for automated isolation of nucleic acids (6-48 samples per run).

**RESULTS:** Tables 1 and 2 show typical isolated DNA and RNA, respectively, from a variety of different samples, all isolated on the GenoM-48. The elution volumes (in which the NA is isolated) differ typically from 100 to 300  $\mu$ l.

Table 1. Isolated DNA from a variety of samples on the GenoM-48. Comparing old and new kits.

Sample volume/material	Old kit ( $\mu$ g)	New kit ( $\mu$ g)
Whole blood 100 $\mu$ l	1.1-3.1	1.7-3.0
Whole blood 200 $\mu$ l	2.4-5.2	3.5-6.0
Whole blood 350 $\mu$ l	-	5.0-10
50 $\mu$ l buffy/1x10 <sup>8</sup> cells		6-10
75 $\mu$ l buffy/1x10 <sup>8</sup> cells		8-12
100 $\mu$ l buffy/1x10 <sup>8</sup> cells	4.4-6.3	10-15
2x10 <sup>6</sup> HL60 cells	-	6-9
Porcine muscle/60 mg	-	8-9
Rat liver/20 mg	-	6-10

Table 2. Isolated total RNA from cultured cells on the GenoM-48. Comparing old and new kits. Typical values.

Cell samples	Old kit	New kit
2x10 <sup>6</sup> Jurkat	6-8 $\mu$ g	8-12 $\mu$ g
2x10 <sup>6</sup> Bjab	7-9 $\mu$ g	10-12 $\mu$ g
2x10 <sup>6</sup> HL60	13-15 $\mu$ g	22-28 $\mu$ g
4x10 <sup>6</sup> HL60	-	45-50 $\mu$ g

**DISCUSSION & CONCLUSIONS:** Although the old NA isolation kit gives excellent quality of isolated DNA (ratio 280/260 = 1.8) and RNA (ratio 280/260 ca 1.9), the yield for some applications are in the lower acceptable range.

With the new chemistry, we have improved the yields of isolated NA's without compromising the quality (DNA: ratio 280/260 ca 1.87 and RNA: 280/260 ca 2.0). In addition, the second generation kits allow for larger input volumes of samples, and the isolated yield of NA in general shows an improved linearity with respect to sample input.

**REFERENCES:** For more information on GenoVision and its products (handboks etc), please consult [www.genovision.com](http://www.genovision.com).