# Rapid and qPCR ready DNA isolation from environmental and human bodily fluid samples using Magnetic bead system

**Luiza Lima[2], Won-Sik Kim[1] and Yousef Haj-Ahmad[1]**

1 Norgen Biotek Corp. 3430 Schmon Parkway, Thorold, Ontario, Canada, 2 Brock University, 500 Glenridge Avenue, St. Catharines, Ontario, Canada

## Abstract

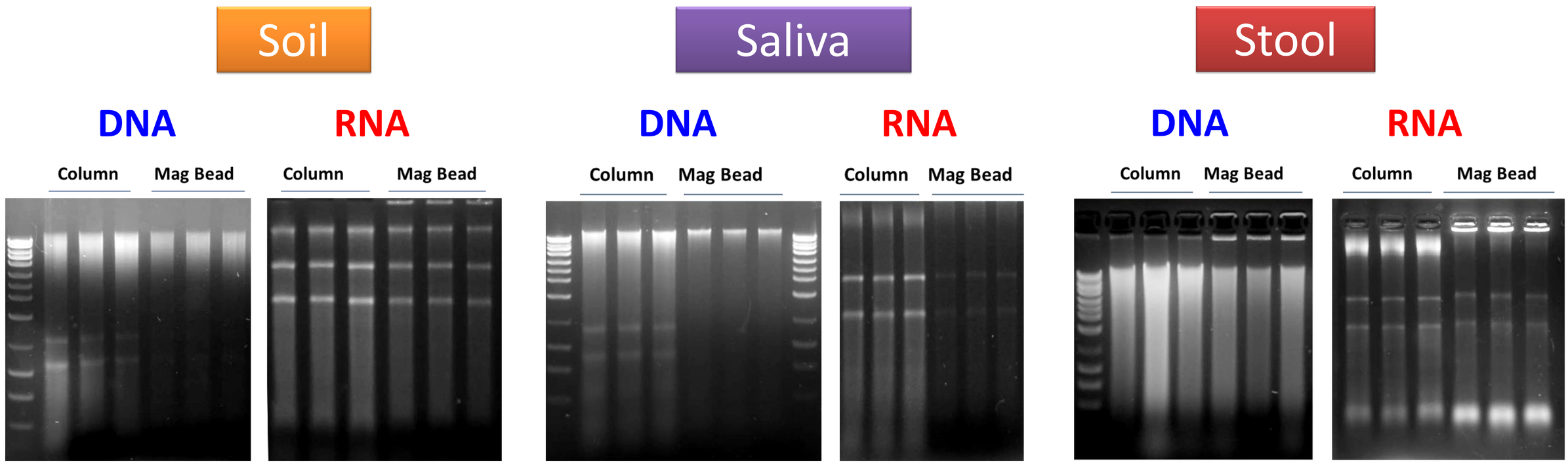
A magnetic bead system for DNA isolation was tested with the aim of producing a general approach for qPCR-ready DNA from environmental and human bodily fluid samples that are challenging to gain high quality of DNA and time consuming. Optimization of lysis, binding and wash conditions were done using several types of tissue samples (Soil, Stool, Saliva and Plasma). In all cases, DNA suitable for PCR was prepared in less than 30 minutes except stool and soil samples that were mechanically homogenized to gain a better DNA yield prior to the magnetic bead binding step. Optimized washing condition removed the PCR inhibitors efficiently and DNA was simply eluted non-EDTA contained elution buffer such as Nuclease-free water. This system is robust and reproducibly yields qPCR-ready DNA. Since there are no centrifugation steps or vacuum steps required whole DNA isolation procedure can be done efficiently in a small space. A robotic manipulation system for DNA bound to magnetic beads is also in progress adapting an automation of DNA extraction system for the types of material tested here.

## Methods & Materials

| Isolation method | Sample | DNA | RNA (spiked with 1.5 ug of E.coli total RNA) |
|---|---|---|---|
| Spin column (Norgen Biotek) | Soil | Soil DNA Isolation kit (Cat. 26500) | Soil total RNA purification (Cat. 27750) |
| | Saliva (preserved)* | Saliva DNA isolation kit (Cat. RU45400 ) | Total RNA purification kit (Cat. 17200) |
| | Stool (preserved)** | Stool DNA isolation kit (Cat. 27600) | Stool total RNA purification (Cat. 49500) |
| Magnetic bead | Same samples applied as above | ➢ 200 mg (soil) 200 µl of preserved stool or saliva sample were mixed with lysis buffer<br>➢ Bead beating for soil and stool samples and collect the clean supernatant only<br>➢ Add Magnetic beads and stand magnetic rack to separate DNA/RNA from impurities<br>➢ Remove the supernatant and wash the magnetic bead twice with washing buffer<br>➢ Dry the magnetic beads at 65˚C and elute with 75-100 µl of elution buffer | |

* Saliva DNA Collection and Preservation Devices (50) (Cat. RU49000)

** Stool Nucleic Acid Collection and Transport Tubes (Cat. 45630, 45660)




## Summary

- This preliminary result showed that Magnetic bead system can be applicable for various challenging samples such as stool, soil and saliva.
- With the simple, convenient and rapid purification procedure, magnetic bead method can be converted into a high throughput system.
- Further optimization is underway to improve over all DNA and RNA yield against the column methods that have shown a consistency, high yield and quality.

NORGEN BIOTEK CORP.

3430 Schmon Parkway, Thorold, ON L2V 4Y6 Canada
Phone: (905) 227-8848 | Toll Free: 1-866-NORGENB (667-4362) | Fax: (905) 227-1061 | email: info@norgenbiotek.com

Brock University