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## RT-PCR-Ready Viral RNA from Nasopharyngeal Samples

### OVERVIEW

Major disease outbreaks are associated with the circulation of influenza virus Types A and B in human populations. Infection with Influenza B virus is usually milder than infection with Influenza A virus, but both agents are significant causes of upper respiratory illness. Isolation of viral RNA is a key step for much influenza research. The Akonni Biosystems TruTip Extraction Kits offer a competitive solution for fast and efficient isolation of Influenza A and B viral RNA from challenging clinical sample types to deliver inhibitor-free, RT-PCR-ready RNA in just minutes.

- Clear your backlog in TruTip Speed
- Remove inhibitors and contaminants with TruTip Purity
- Eliminate spin column and centrifugation steps
- Achieve yields equal to industry leaders
- Highly adaptable to fit your throughput needs

### AUTOMATED SOLUTIONS

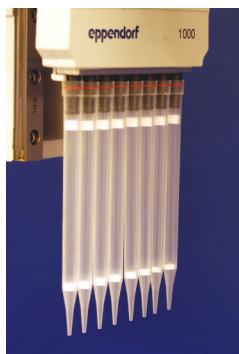
Akonni TruTips are available for Eppendorf's epMotion automated pipetting system. Automating the process allows for higher sample throughput and eliminates user error. Twenty-four samples can be processed on a single plate in 40 minutes, or less than 2 minutes per sample. The program includes set-up time for aliquoting all reagents, which allows the user to process samples easily with the press of a button.

### TruTip RNA EXTRACTION RESULTS

Extensive testing on the extraction of influenza A (FluA) and influenza B (FluB) from both nasopharyngeal aspirate (NPA) samples and nasopharyngeal swab (NPS) samples in viral transport media (VTM) has demonstrated detection at clinically relevant concentrations with high reproducibility.

#### Reproducibility Study

Column 1		Column 4		Column 8	
Sample	Ct	Sample	Ct	Sample	Ct
EP1	32.73	EP9	32.84	EP17	32.93
EP2	33.25	EP10	33.05	EP18	33.24
EP3	32.71	EP11	32.84	EP19	32.80
EP4	32.80	EP12	32.92	EP20	32.75
EP5	32.81	EP13	32.98	EP21	32.70
EP6	32.53	EP14	32.91	EP22	33.06
EP7	33.50	EP15	33.02	EP23	32.84
EP8	32.53	EP16	32.81	EP24	33.20
Avg. Ct		Std. Dev.			
32.85		0.35			



#### Eppendorf epMotion® 5070



Extracted RNA was detected by real-time PCR on the Roche LightCycler 480, using the Invitrogen SuperScript One-Step RT-PCR kit.

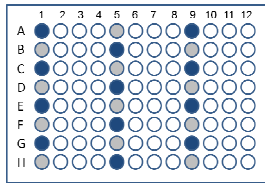
Input FluA Virus Conc.	Avg. Ct. (n=15)	Std. Dev.
10 <sup>6</sup> gc mL <sup>-1</sup>	33.74	0.50
10 <sup>5</sup> gc mL <sup>-1</sup>	36.97	0.57
10 <sup>4</sup> gc mL <sup>-1</sup>	37.29	0.54

**Table 1.** Dilution series of FluA spiked into 5 different Flu-Negative NPA samples (n=3 ea.). FluB was also reproducibly detected (100%) at 10<sup>5</sup> genome copies (gc) mL<sup>-1</sup>, with 10<sup>4</sup> gc mL<sup>-1</sup> approaching the detection limit of the real time RT-PCR assay (data not shown). Extractions performed on the epMotion 5070 with 1000 µL tips.

(Left) Twenty-four replicate extractions of 100 µL FluA at 10<sup>6</sup> gc mL<sup>-1</sup> in Flu-negative NPA were processed on a single plate. Results demonstrated high precision of this extraction method using the automated system.

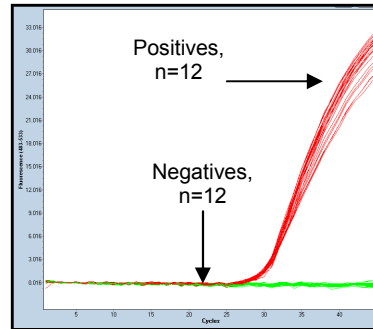
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## Cross-contamination Study



● Positive Influenza  
○ Negative Control

Avg. Ct (n=12)	Std. Dev.
30.16	0.14



## Akonni TruTip Extraction Kit



(Above) Flu-negative NPA spiked with or without  $10^6$  gc  $\text{mL}^{-1}$  FluA in an alternating pattern and processed on a single plate according to the plate map. Results demonstrated no cross-contamination.

## ORDERING INFORMATION

**TruTip Influenza RNA Kit:** To extract and purify influenza RNA from nasopharyngeal swab and aspirate samples.

## AUTOMATED SOLUTIONS

**300-11120 EPM TruTip Influenza Kit (EPM Tips, 1000  $\mu\text{L}$ , g6 SPT)** For use with the *epMotion* Automated Pipetting System. Includes TruTip SPT tips and buffers for g6 extractions.

To order call +1 301.698.0101

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For Research Use Only. Not for use in diagnostic procedures. No claim or representation is intended for the diagnosis, prevention, or treatment of a disease.

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TTP-DS-010 1005.5