



PhoenixDx® Influenza/RSV Multiplex is a multiplex detection system for Influenza A, Influenza B, RSV and a human extraction control (HEC) with the target RNase P for RT-PCR. Viral RNA is converted into cDNA through reverse transcription and unique DNA sequences for each target are subsequently amplified and detected via probe-based qPCR.

Detection and discrimination between the different targets is done in **4 easy steps**:

1) Collect samples: **PhoenixDx® Influenza/RSV Multiplex** works with sputum, nasopharyngeal swabs, nasopharyngeal aspirates, bronchoalveolar lavage samples in Universal Transport Medium (UTM)/ Viral Transport Medium (VTM) and swab samples.

2) Isolate RNA: sample RNA can be isolated manually with a spin column system or magnetic bead system or fully automated with the SphaeraMag® DNA/RNA isolation system, or the Nuclefy spin column series.

3) Perform RT-PCR with PhoenixDx® Influenza/RSV Multiplex: PhoenixDx® Influenza/RSV Multiplex consist of only 3 components for easy sample setup:

COMPONENT	VOLUME
PhoenixDx® RT Enzyme Mix	1 µl
PhoenixDx® 5X MTM Buffer	4 µl
PhoenixDx® Influenza/RSV Assay Mix	1 µl
isolated sample RNA / TPC / NTC	14 µl / 14 µl / 14 µl dH ₂ O

STEP	CYCLES	TEMPERATURE	DURATION
Reverse Transcription	1	50°C	5 minutes
Initial Denaturation	1	95°C	5 minutes
Amplification	45	95°C	5 seconds
		60°C²	30 seconds

HEC Quality & Performance Control

- shows RNA isolation & transcription were successful
- shows if subsequent PCR amplification was successful
- helps to evaluate overall sample RNA quality
- Reduces false-negative results

² Enable Data Collection: **FAM™** for Influenza A
HEX/VIC for Influenza B
ROX for RSV (do not enable ROX normalization)
CY5 for the control (HEC)

4) Analyze your results:

FAM™	HEX/VIC	ROX	Cy5	Result
/	/	/	Ct < 37	The sample does not contain Influenza A, B or RSV RNA . The control was amplified successfully.
/	/	/	/	No amplification in any channel indicates flawed RNA isolation, sample degradation or PCR inhibition. Results cannot be interpreted.
Ct < 37	/	/	Ct < 37	The sample is positive for Influenza A .
/	Ct < 37	/	Ct < 37	The sample is positive for Influenza B .
/	/	Ct < 37	Ct < 37	The sample is positive for RSV .