



**510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY AND INSTRUMENT**

I Background Information:

A 510(k) Number

K221658

B Applicant

Centers for Disease Control and Prevention

C Proprietary and Established Names

Non-variola Orthopoxvirus Real-time PCR Primer and Probe Set

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
PBK	Class II	21 CFR 866.3315 - Nucleic Acid Based Reagents For Detection Of Non-Variola Orthopoxviruses	MI - Microbiology

II Submission/Device Overview:

A Purpose for Submission:

Modification of device

B Measurand:

Non-variola *Orthopoxvirus* DNA target sequence

C Type of Test:

In vitro molecular diagnostic test

III Intended Use/Indications for Use:

A Intended Use(s):

See Indications for Use below.

B Indication(s) for Use:

The Non-variola Orthopoxvirus Real-time PCR Primer and Probe Set is intended for the in vitro qualitative presumptive detection of non-variola Orthopoxvirus DNA extracted from human pustular or vesicular rash specimens and viral cell culture lysates submitted to a Laboratory Response Network (LRN) reference laboratory. The assay detects non-variola Orthopoxvirus DNA, including Vaccinia, Cowpox, Monkeypox and Ectromelia viruses at varying concentrations. This assay does not differentiate Vaccinia virus or Monkeypox virus from other Orthopoxviruses detected by this assay and does not detect Variola virus. Refer to the CDC algorithm, Acute, Generalized Vesicular or Pustular Rash Illness Testing Protocol in the United States for recommended testing and evaluation algorithms for patients presenting with acute, generalized pustular or vesicular rash illness.

Results of this assay are for the presumptive identification of non-variola Orthopoxvirus DNA. These results must be used in conjunction with other diagnostic assays and clinical observations to diagnose Orthopoxvirus infection. The assay should only be used to test specimens with low/moderate risk of smallpox. If a high risk of smallpox exists, viral culture should not be attempted. Negative results obtained with this device do not preclude Variola virus infection and should not be used as the sole basis for treatment or other patient management decisions.

Use is limited to Laboratory Response Network (LRN) designated laboratories.

C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

Distribution of device is limited to designated laboratories in the Laboratory Response Network.

D Special Instrument Requirements:

Real-Time PCR Instrumentation and Software

IV Device/System Characteristics:

A Device Description:

The Non-variola *Orthopoxvirus* Real-time PCR Primer and Probe Set uses a fluorogenic probe, consisting of an oligonucleotide with a reporter dye (FAM) attached to the 5' end and a quencher dye (BHQ1) attached at or near the 3' end. The probe anneals to a specific target sequence located between the forward and reverse primers. During the extension phase of the PCR cycle, the 5' nuclease activity of the *Taq* polymerase degrades the probe causing the reporter dye to separate from the quencher dye, thereby generating a fluorescent signal. With each cycle, additional reporter dye molecules are cleaved from their respective probes and the fluorescence intensity is monitored during the PCR in real-time. The *Taq* polymerase used in this assay is inactive at room temperature and is activated by incubation at 95°C, thus minimizing the production of nonspecific amplification products.

Each extracted DNA sample is tested using the Non-variola *Orthopoxvirus* Real-time PCR Primer and Probe set along with an internal control primer and probe set(s) to demonstrate adequate DNA extraction, proper function of common reagents and equipment, and the absence of inhibitory substances.

B Instrument Description Information:

Testing with the Non-variola *Orthopoxvirus* Real-time PCR Primer and Probe set is conducted using real-time PCR instrumentation and software.

V Substantial Equivalence Information:

A Predicate Device Name(s):

Non-variola Orthopoxvirus Real-time PCR Primer and Probe Set

B Predicate 510(k) Number(s):

K181205

C Comparison with Predicate(s):

Device & Predicate Device(s):	<u>K221658</u>	<u>K181205</u>
Device Trade Name	Non-variola <i>Orthopoxvirus</i> Real-time PCR Primer and Probe Set	Same
General Device Characteristic Similarities		
Intended Use/Indications For Use	The Non-variola <i>Orthopoxvirus</i> Real-time PCR Primer and Probe Set is intended for the <i>in vitro</i> qualitative presumptive detection of non-variola <i>Orthopoxvirus</i> DNA extracted from human pustular or vesicular rash specimens and viral cell culture lysates submitted to a Laboratory Response Network (LRN) reference laboratory. The assay detects non-variola <i>Orthopoxvirus</i> DNA, including <i>Vaccinia</i> , <i>Cowpox</i> , <i>Monkeypox</i> and <i>Ectromelia</i> viruses at varying concentrations. This assay does not differentiate <i>Vaccinia virus</i> or <i>Monkeypox</i>	Same

	<p><i>virus</i> from other <i>Orthopoxviruses</i> detected by this assay and does not detect <i>Variola virus</i>. Refer to the CDC algorithm, <u>Acute, Generalized Vesicular or Pustular Rash Illness Testing Protocol</u> in the United States for recommended testing and evaluation algorithms for patients presenting with acute, generalized pustular or vesicular rash illness.</p> <p>Results of this assay are for the presumptive identification of non-variola <i>Orthopoxvirus</i> DNA. These results must be used in conjunction with other diagnostic assays and clinical observations to diagnose <i>Orthopoxvirus</i> infection. The assay should only be used to test specimens with low/moderate risk of smallpox. If a high risk of smallpox exists, viral culture should not be attempted. Negative results obtained with this device do not preclude <i>Variola virus</i> infection and should not be used as the sole basis for treatment or other patient management decisions.</p> <div style="border: 1px solid black; padding: 5px; width: fit-content;"> <p>Use of this assay is limited to Laboratory Response Network (LRN) designated laboratories.</p> </div>	
Principle of Operation	Nucleic acid amplification and fluorescent probe detection	Same
Sample Types	<ul style="list-style-type: none"> • Vesicle fluid, skin, crust, “roof” • Dry or wet swab of lesion (dry swab is 	Same

	preferred) <ul style="list-style-type: none"> • Touch prep (slide) of lesion • Fresh biopsy of pustule or vesicle (no formalin) • Viral cell culture lysates 	
Instrumentation and Software	Real-time PCR instrumentation and software	Same

VI Standards/Guidance Documents Referenced:

N/A

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

N/A

2. Analytical Specificity/Interference:

Inquiries regarding performance characteristics for the Non-variola *Orthopoxvirus* Real-time PCR Primer and Probe Set should be directed to the Centers for Disease Control and Prevention.

3. Detection Limit:

The limit of detection for the Non-variola *Orthopoxvirus* Real-time PCR Primer and Probe Set was determined through an analytical sensitivity study.

4. Assay Cut-Off:

N/A

B Comparison Studies:

1. Method Comparison with Predicate Device:

N/A

2. Matrix Comparison:

N/A

C Clinical Studies:

Inquiries regarding performance characteristics for the Non-variola *Orthopoxvirus* Real-time PCR Primer and Probe Set should be directed to the Centers for Disease Control and Prevention.

D Clinical Cut-Off:

N/A

E Expected Values/Reference Range:

N/A

VIII Proposed Labeling:

The labeling supports the finding of substantial equivalence for this device.

IX Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.