

# Supplementary Data

## **Global Mpox Lineage Discovery and Rapid Outbreak Tracking with Nanopore Sequencing**

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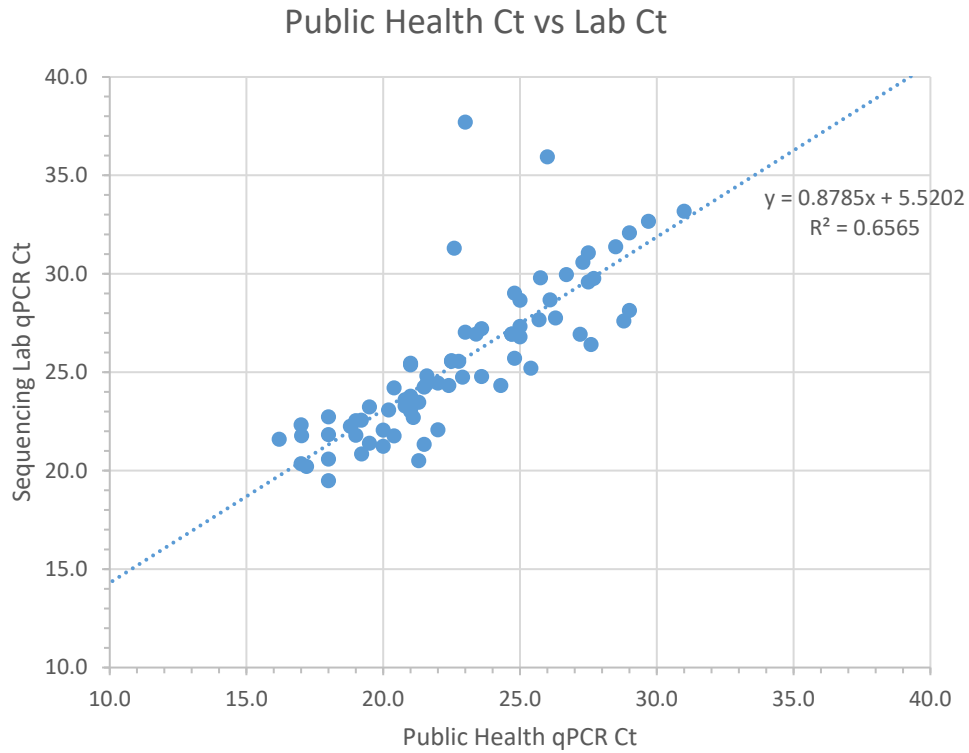
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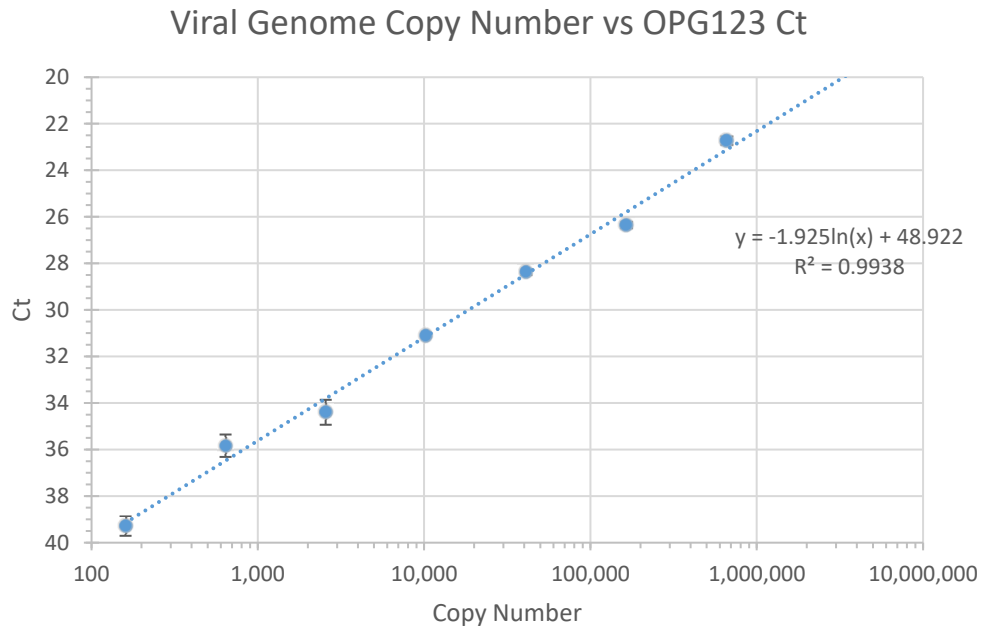
<sup>†</sup> These authors contributed equally.

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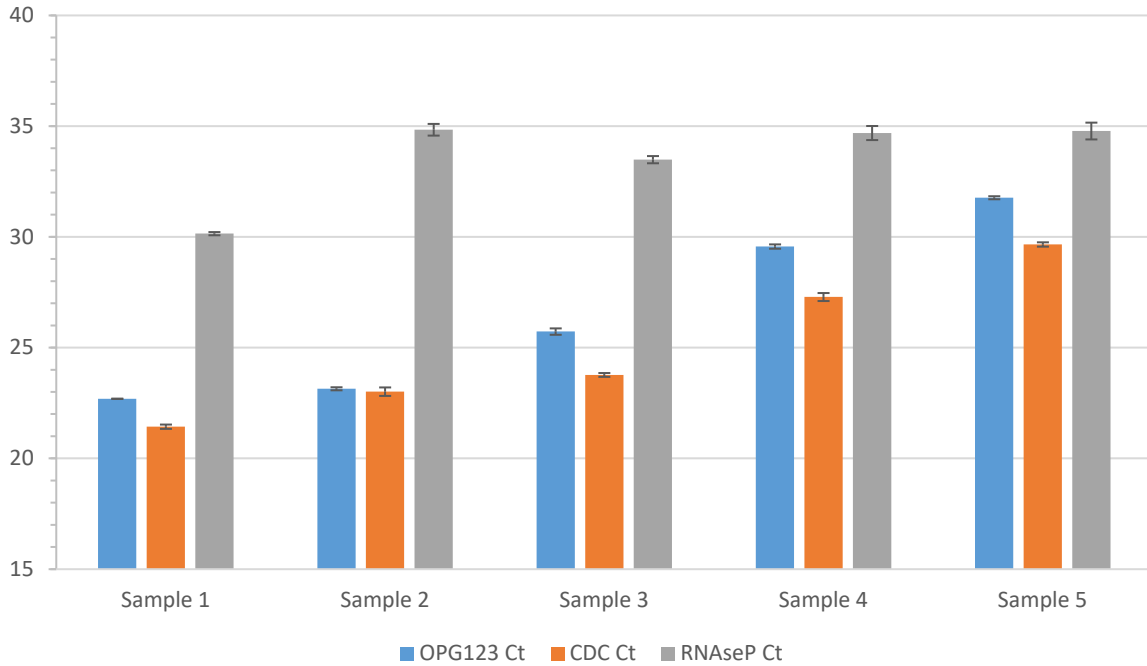
## Supplementary Figures



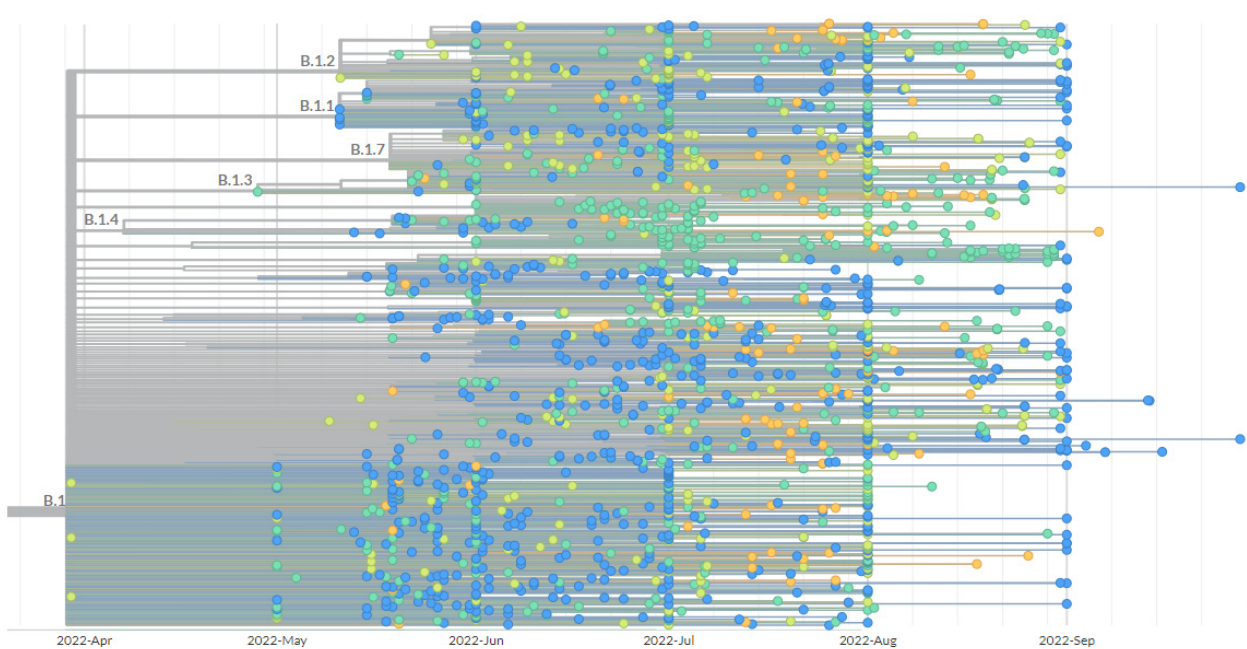
**Supplementary Figure S1:** Comparison of qPCR Ct values from Illinois Department of Public Health and those generated using OPG123 qPCR probes.



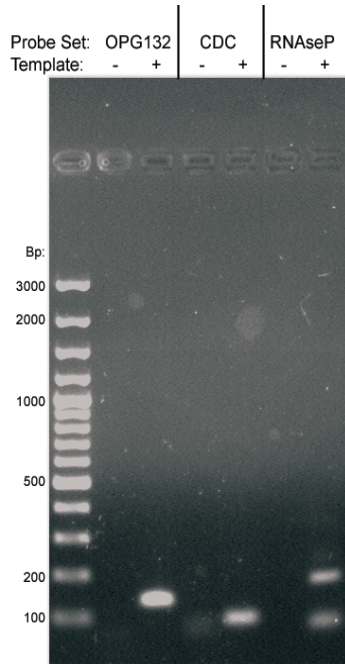
**Supplementary Figure S2:** Comparison of number of copies of a DNA gBlock and the qPCR Ct values generated from them using the OPG123 qPCR probe set.



**Supplementary Figure S3:** Ct values for five different mpox samples, using three different qPCR probes. Each sample/probe combination was replicated three times.

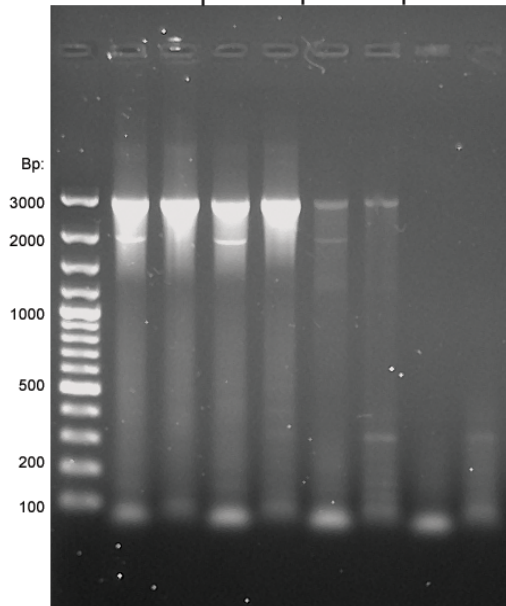


**Supplementary Figure S4:** Phylogenetic tree showing Clade IIb, Lineage B.1 mpox sequences. Color of each sequence indicates status of indicated deletion starting at base 179077. Teal: no deletion. Lime-green: Deletion to base 179094 (17 bases removed). Blue: Deletion to base 179148 (71 bases removed). Orange: Deletion to base 179175 (98 bases removed). Each lineage follows no clear pattern in deletions, indicating this could be an artifact of sequencing method, rather than actual deletion.



**Supplementary Figure S5:** 1% agarose gel showing the positive and negative control results for three qPCR probe sets. Ladder size markers are shown on the left.

Template:	IDPH-22		IDPH-42		IDPH-52		No template	
Pool:	1	2	1	2	1	2	1	2
Coverage:	99.74%		99.42%		99.95%		-	



**Supplementary Figure S6:** 1% agarose gel showing three randomly-selected multiplex PCR samples, along with a negative control (no template). Final sequence coverage of each sample is also shown. Ladder size markers are shown on the left.

