DISCOVERY OF ABERRANT GENOMIC REARRANGEMENT IN AN MPOX VIRUS GENOME OBSERVED IN A JAPANESE PATIENT

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BACKGROUND

- MPXV has a 197-kb linear dsDNA genome, which consists of a core, left and right variable regions including ITR.
- The ITRs are identical but in opposite direction.
- World-wide outbreak of ***MPXV clade IIb*** was declared by WHO in 2022-2023.
- Over 250 cases have been reported in Japan since July 2022 till present. ***No clade I*** is found so far.
- We experienced ***3 Mpox cases*** at CUH, and performed the genomic analysis for tracking viral dissemination and evolution.

METHODS

- **Genomic analysis:** WGS by tiling PCR and digital PCR based-copy number quantification
- **Viral properties:** Virus isolation, microscopic observation, and growth kinetic analysis

We have discovered a ***large-scale genomic rearrangement*** in one of the MPXV strains with holding its ***replicability and pathogenicity.***

This might be one of the ***key mechanisms*** for drastic ***viral evolution*** in MPXV.

RESULTS

1. Cases

- The patients were all MSM and PLWH. Good virological control of HIV had been achieved.
- They had history of sexual intercourse with plural men several days before onset.
- They all showed ***typical symptoms of Mpox*** (Fever and pustules, etc.).

2. Genomic Analysis

- WGS were successfully attained. All 3 MPXV belonged ***to clade IIb, lineage C.1 (Fig. 1A)***.

Mpox-CH2

- A nonsense mutation (OPG195:Q88*) was detected at the 3´-variable region.

Mpox-CH3

- 5.5 kb-long ***large deletion*** at 5´-variable region was detected (**Fig. 2A**).
- The copy numbers of 160–191 kb region were increased 1.6–2.3 fold (**Fig. 2B**).
- 30.5 kb-long 3´-genomic portion was ***invertedly inserted*** in the deleted region (**Fig. 2C**).
- ***The ITR was expanded from 6.5 kb to 37 kb by the above mentioned large-scale genomic rearrangement (Fig. 1A).***

3. Viral properties

- Viral isolation was succussed in all the 3 patients' specimens.
- Mpox-CH3 alone exhibited a ***slight fusogenicity*** at 3-4 d.p.i. (the other 2 strains did not show such a property.) ***(Fig. 1B)***
- All the 3 MPXV strains ***propagated in a similar manner*** in VERO cells and reached a peak at 5–6 d.p.i. ***(Fig. 1C)***

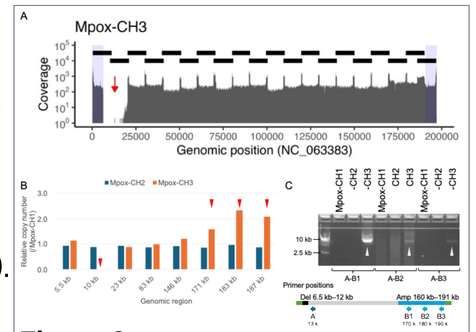


Figure 2.
A) Mapping result of WGS.
B) Relative copy number of each genomic region.
C) Unidirectional PCR.

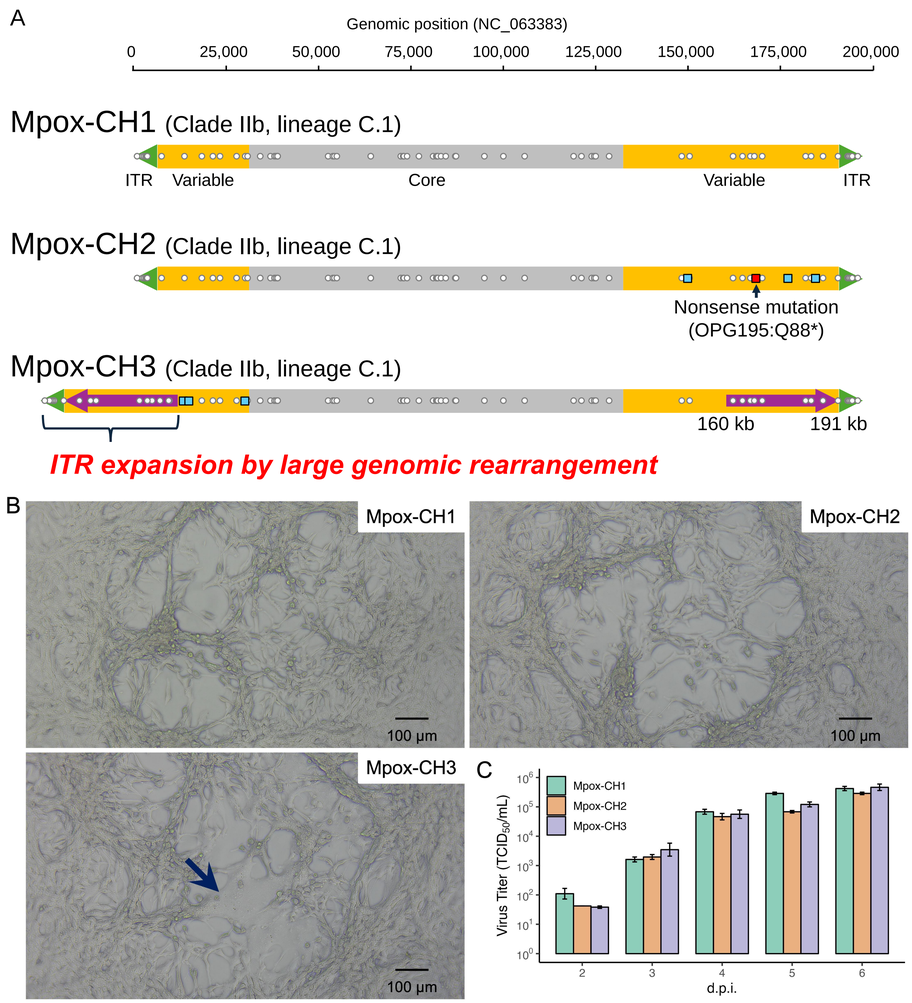


Figure 1. The genomic structures **(A)**, CPEs in VERO cells **(B)**, and viral growth kinetics **(C)** of 3 MPXV. (A) Open circles and blue squares indicate common and individual mutations, respectively. (B) The allow indicates a fusogenic part.

CONCLUSIONS

- All clinical features of the patients with Mpox were ***typical***, and ***no noteworthy differences*** among them had been observed.
- ***Large genomic rearrangement*** was identified in one of three strains, which maintained its robust ***replicability*** in VERO cells showing ***slight fusogenic*** CPEs.
- The future genomic analyses must be conducted carefully.