## Project

## **Epitope mapping for Diagnostic Testing for MPXV virus**

**Overall criteria**

* **Aas > 9; surface localized, extracellular and antigenic**

**Abstract**

The world has witnessed a resurgence of monkeypox (MPOX) disease driven by mutants of the original West African and Central African clades of monkeypox virus (MPXV:- namely Clade IIb-2022-to-2023; Clade 1b-Sept 2024 todate). Their is paucity of approved point of care, rapid diagnostic tests (POC-RDTs). Here, we map epitopes of two rationally selected diagnostic and possibly therapeutic targets (surface localization; involvement in virus-host attachment and entry): (a) VMP-OPG143 (A17L) and (b) EP-OPG108 (H3L). Six and three best candidates are selected as follows:

1. VMP-OPG143 (myristylayed protein, A17L) (#=6 candidate epitopes)

* 11-**KIETGIADIRD**-21;
* 44-**YMYYYETSPGEI**-55;
* 104-**QNEVTPEYIKDLKYATDYIA**-123;
* 270-**KKDYLLGDSDSVAKCCSKTNTKHCPKIFNNNYKTEHC**-290
* 291-**CKYVGCTINVNSL**-303;
* 110-**EYIKDLKYA**-118

1. EP-OPG108 (heparin binding protein, H3L) (#=3)

* 17-**PPSETFPNVHEHINDQKFDDVKDNEVMQEKRDVVIVNDDPDHY**-59
* 28-**HINDQKFDDVKDNEVMQEKR**-47
* 236-**YVEHDPRLVAEH**-247

**Conclusions**: Across all parameters, the two best identified epitopes for detection of VMP—OPG143 (A17L) and EP-108 (H3L) are respectively as follow:

* VMP-OPG143: 270-**KKDYLLGDSDSVAKCCSKTNTKHCPKIFNNNYKTEHC**-290 (104-**QNEVTPEYIKDLKYATDYIA**-123;)
* EP-OPG108: 236-**YVEHDPRLVAEH**-247 (28-**HINDQKFDDVKDNEVMQEKR**-47)

**Detailed methodology**

1. **Target protein # 1: Monkeypox Virion Membrane Protein (VMP) OPG143 (Gene A17L)**

* Rationale for selection:

1. **Surface localization:** Surface protein found on the viral envelope.
2. **Function**: It plays a crucial role in viral replication by enabling the fusion of the virus with the host cell membrane, and viral egress (Shchelkunov et al., 2002; Gao et al., 2023). VMP is therefore an envelope protein part of the entry-fusion complex(EFC) responsible for the virus membrane fusion with host cell membrane during virus entry. Also plays a role in cell-cell fusion (syncytium formation).
3. **Implications**: Monoclonal antibodies raised against VMP-OPG143 may therefore have both diagnostic and therapeutic utility

* Uniprot Accession number and fasts format

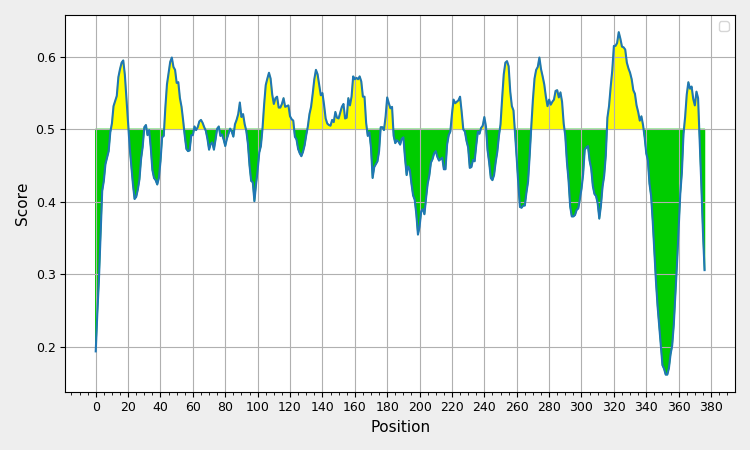
>sp|A0A7H0DNB5|PG143\_MONPV Virion membrane protein OPG143 OS=Monkeypox virus OX=10244 GN=OPG143 PE=3 SV=1 MGAAVTLNRIKIETGIADIRDKYMVLDFNYPEYNRAVRFAEESYMYYYETSPGEIKPKFCLIDGMSIDHCSSFIVPEFAKQYVLIHGEPCSSFKFRPGSLIYYQNEVTPEYIKDLKYATDYIASGQRCHFIKKDYLLGDSDSVAKCCSKTNTKHCPKIFNNNYKTEHCDDFMTGFCRNDPGNPNCLEWLRVKRKPAMSTYSDICSKHMDARYCSEFIRIIRPDYFTFGDTALYVFCNDHKGNRNCWCANYPKSNSGDKYLGPRVCWLHECTDESRDRKWLYYNQDVQRTRCKYVGCTINVNSLALKNSQAELTSNCTRTTSAVGDVHPGEPVVNDKIKLPTWLGASITLVVISVIFYFISIYSRPKIKTNDINVRRR

* Extracellular domain

2-MGAAVTLNRIKIETGIADIRDKYMVLDFNYPEYNRAVRFAEESYMYYYETSPGEIKPKFCLIDGMSIDHCSSFIVPEFAKQYVLIHGEPCSSFKFRPGSLIYYQNEVTPEYIKDLKYATDYIASGQRCHFIKKDYLLGDSDSVAKCCSKTNTKHCPKIFNNNYKTEHCDDFMTGFCRNDPGNPNCLEWLRVKRKPAMSTYSDICSKHMDARYCSEFIRIIRPDYFTFGDTALYVFCNDHKGNRNCWCANYPKSNSGDKYLGPRVCWLHECTDESRDRKWLYYNQDVQRTRCKYVGCTINVNSLALKNSQAELTSNCTRTTSAVGDVHPGEPVVNDKIKLP-341

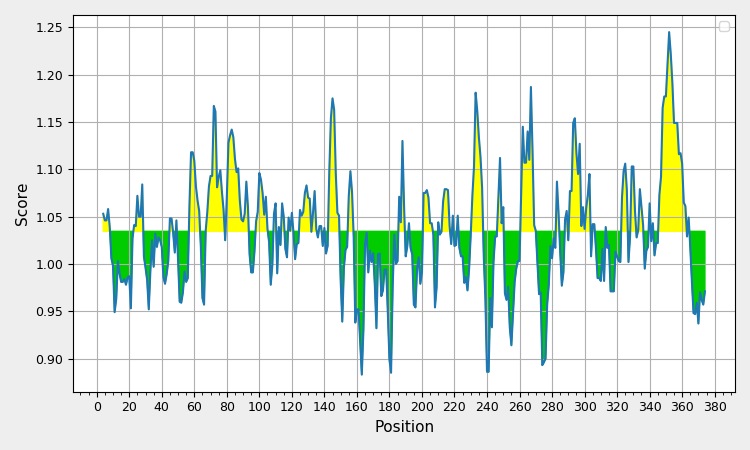
* Epitopes by IEDB-R

1. Bepipred



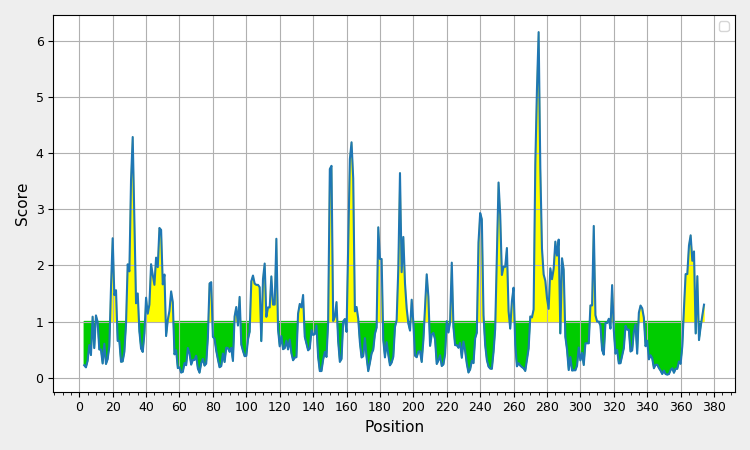
* 11-**KIETGIADIRD**-21
* 44-**YMYYYETSPGEI**-55
* 104-**QNEVTPEYIKDLKYATDYIA**-123
* 132-KKDYLLGDSDSVAKCCSKTNTKHCPKIFNNNYKTEHC-168
* 270-**KKDYLLGDSDSVAKCCSKTNTKHCPKIFNNNYKTEHC**-290
* 317-TRTTSAVGDVHPGEPVVNDKIKL-339
* ~~365-PKIKTNDIN-373 (transmembrane)~~

1. Antigencity



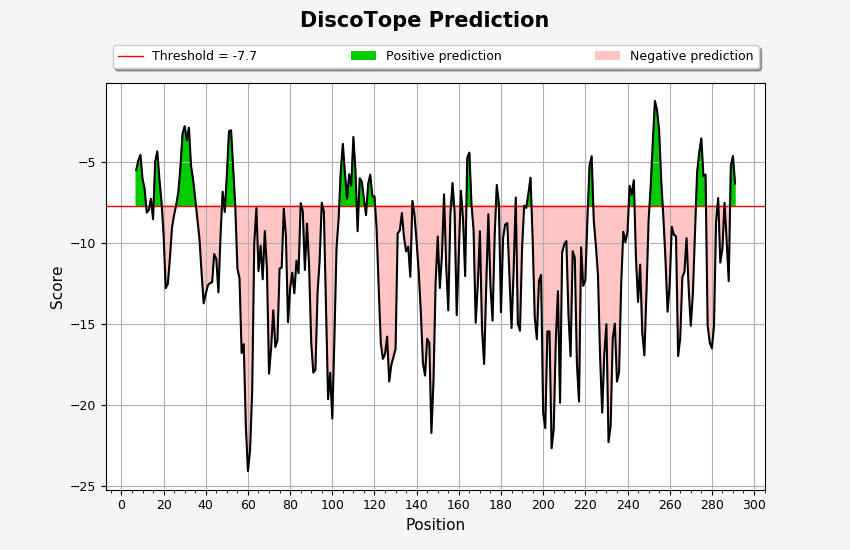
* 67-IDHCSSFIVPEF-78
* 80-KQYVLIHGEPCSSF-93
* 261-GPRVCWLHEC-270
* 291-**CKYVGCTINVNSL**-303
* ~~346-SITLVVISVIFYFISIY-362 (transmembrane)~~

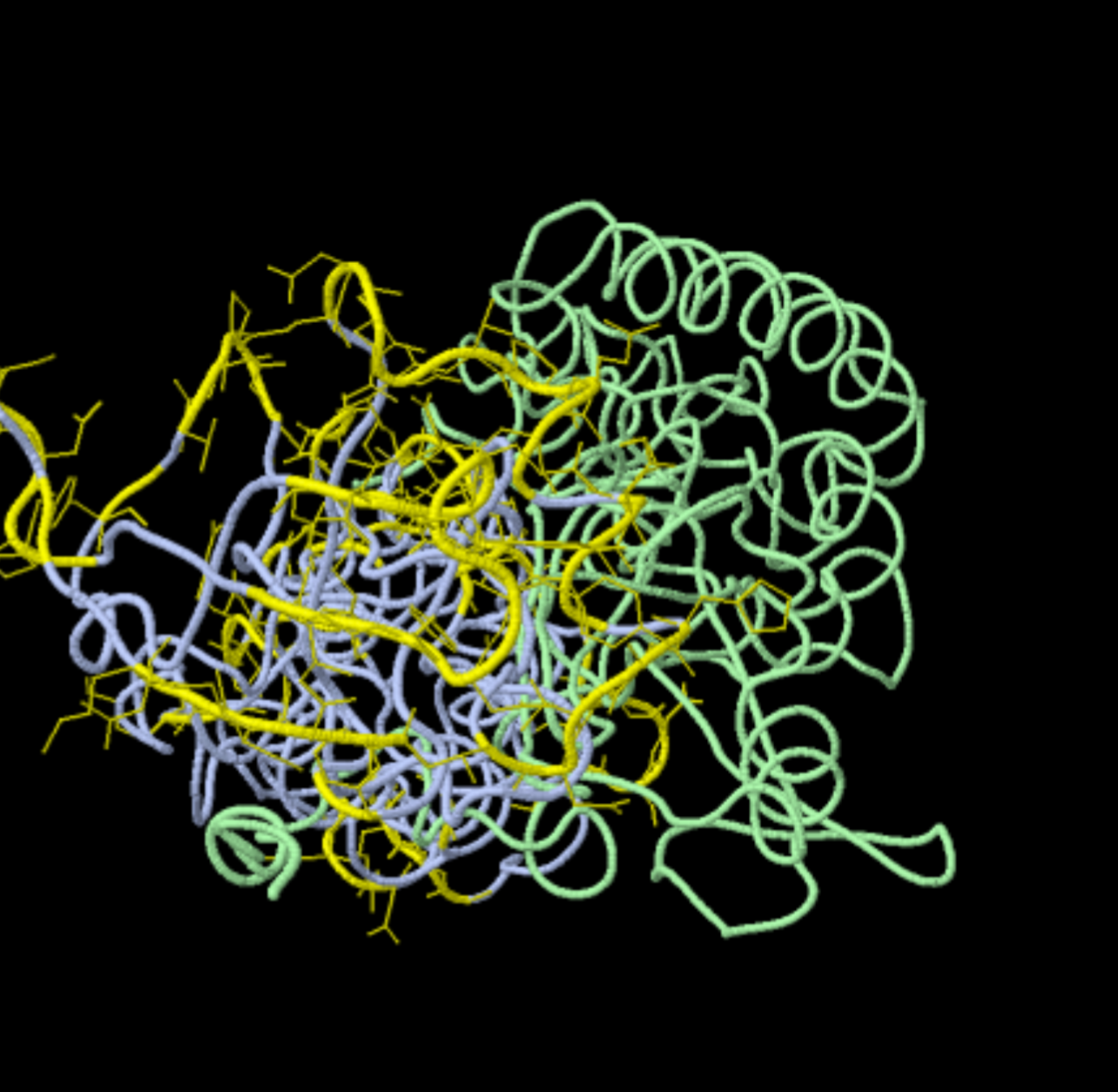
1. Surface accessibility

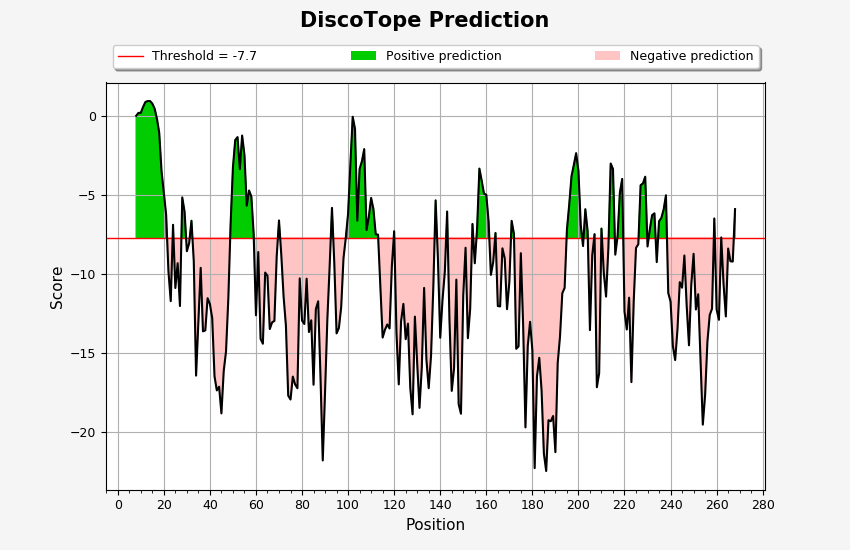


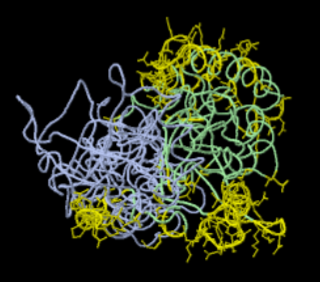
* 40-AEESYMYYYETS-51
* 110-**EYIKDLKYA**-118
* 270-CTDESRDRKWLYYNQDVQ-287

1. Discotope









* Thus, best surface targets between 1-to-20; 25-to-35aa; 50-to-60aa; 100-to120aa; and 200-to-240aa
* Best epitope across all parameters for detection of VMP-OPG143 is: 270-**KKDYLLGDSDSVAKCCSKTNTKHCPKIFNNNYKTEHC**-290

**B. Target protein # 2: Monkeypox virus envelope protein (EP) OPG108 (Gene H3L)**

* Rationale for selection

1. **Surface localization:** Surface, outer envelope protein
2. **Function:**Envelope protein that binds to heparan sulfate on the cell surface and might provide virion attachment to the target cell. Plays role in host virus interaction, virus attachment to host cells and virus entry into host cells.
3. **Implication**: Monoclonal antibodies raised against EP-OPG108 may therefore have both diagnostic and therapeutic utility

* Uniprot Accession number and faster format

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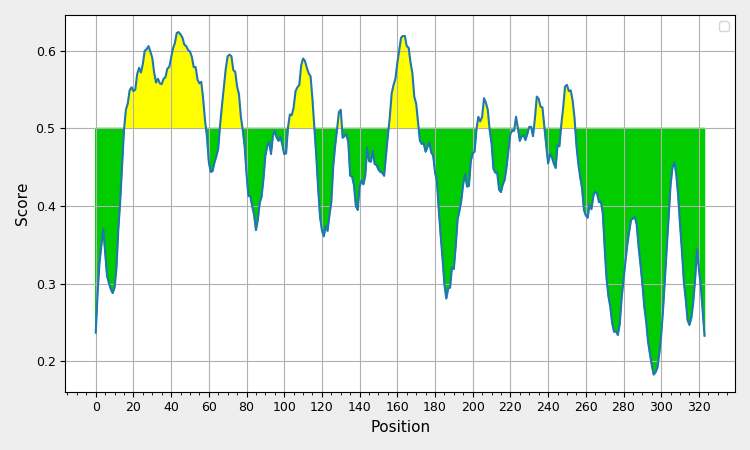
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* Extracellular Domain

1-MAAVKTPVIVVPVIDRPPSETFPNVHEHINDQKFDDVKDNEVMQEKRDVVIVNDDPDHYKDYVFIQWTGGNIRDDDKYTHFFSGFCNTMCTEETKRNIARHLALWDSKFFTELENKNVEYVVIIENDNVIEDITFLRPVLKAIHDKKIDILQMREIITGNKVKTELVIDKDHAIFTYTGGYDVSLSAYIIRVTTALN-284

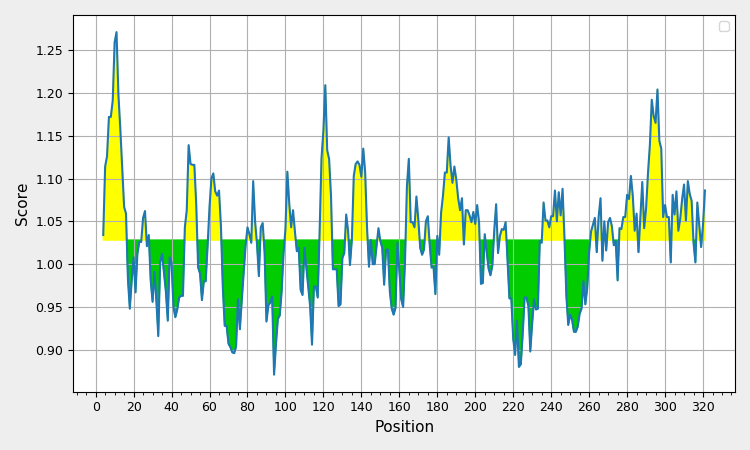
* Epitopes

1. Bepipred



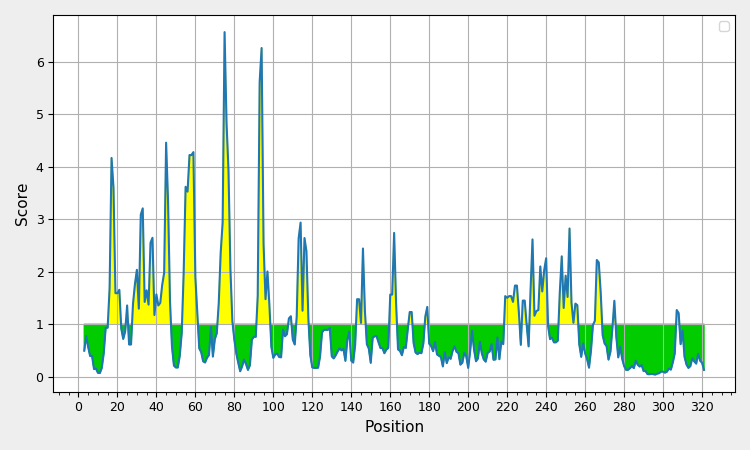
* 17-**PPSETFPNVHEHINDQKFDDVKDNEVMQEKRDVVIVNDDPDHY**-59
* 67-WTGGNIRDDDKY-78
* 103-ALWDSKFFTELENKN-117
* 157-ITGNKVKTELVIDKDH-172

1. Antigenicity



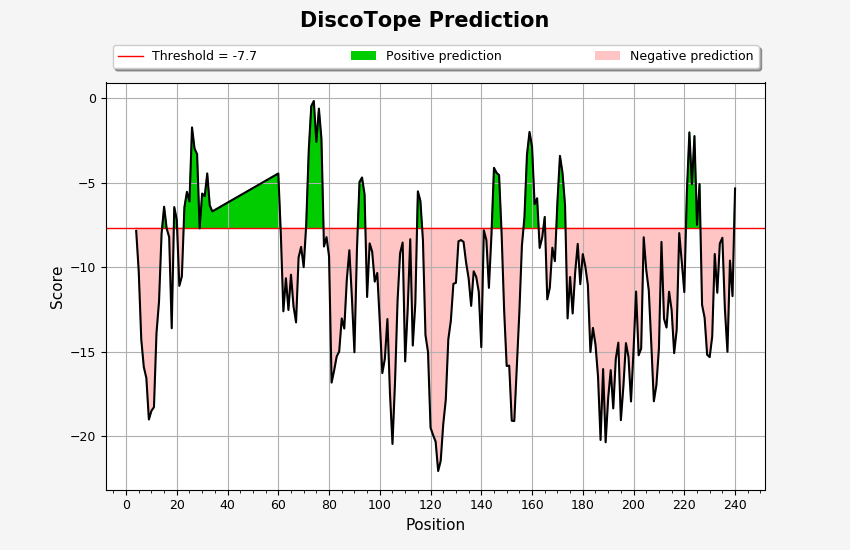
* 4-VKTPVIVVPVIDR-16
* 135-FLRPVLKAI-143
* 182-DVSLSAYIIRVT-193
* 236-**YVEHDPRLVAEH**-247
* 276-TTPLISFFGL-285
* ~~287-DINVIGLIVILFIMFM-302 (transmembrane)~~
* ~~304-IFNVKSKLLWF-314 (transmembrane)~~

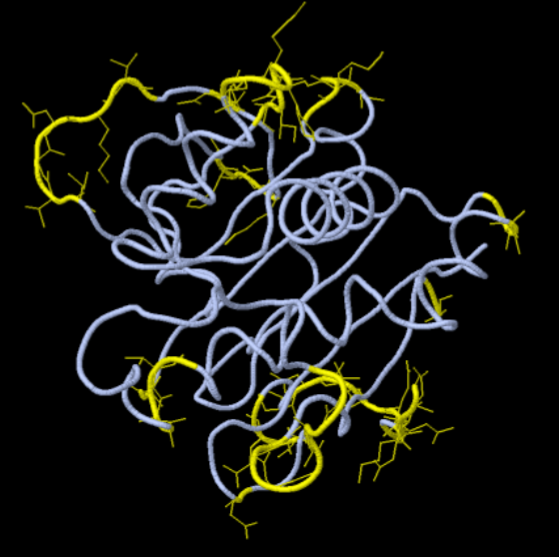
1. Surface accessibility

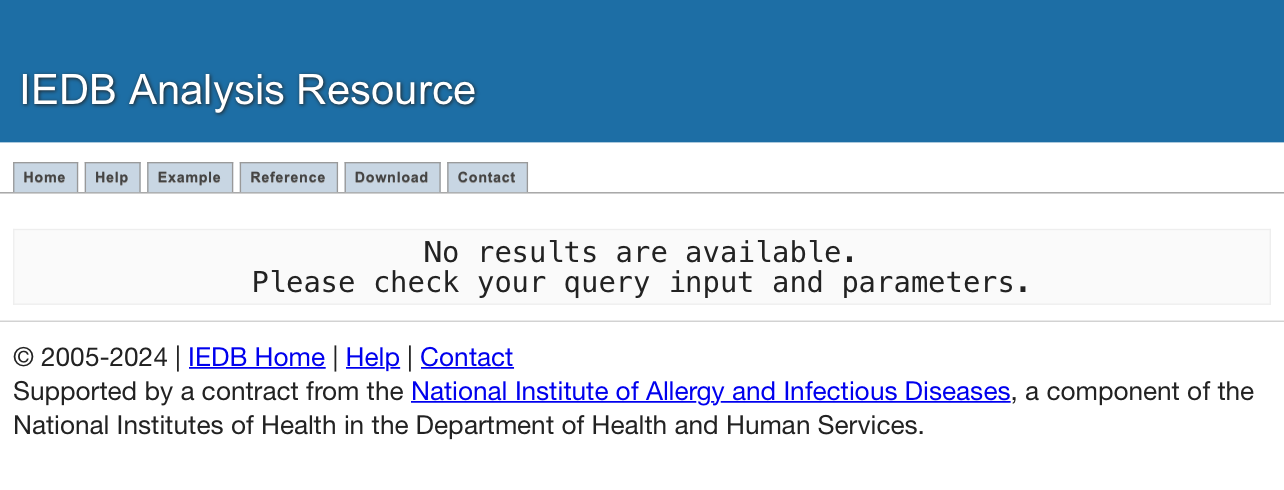


* 28-**HINDQKFDDVKDNEVMQEKR**-47
* 232-NSAKYVEHD-240
* 247-HRFETMKPNF-256

1. Discotope







* Thus best surface antigens located between 20-to-60; 70-to-75;145-to-150; 155-to-165; and 220-to-225
* The best epitope across all parameters for EP-OPG108 is 236-**YVEHDPRLVAEH**-247

**Next Steps**

* Produce purified peptide epitopes to test (ELISA) across the ability to capture host specific Abs (IgM, IgG) in convalescent plasma or serum
* Produce monoclonal antibodies (mAbs) to test (ELISA) for ability to capture recombinant MPXV target proteins or their native analogies on MPXV/ virion like particles (VLPs)