Workshop Report

Technical Consultation on Mpox Correlates of Protection with focus on mRNA vaccine candidates

(June 26-27, 2025)

Meeting report prepared by Dr Julia Granerod, PhD (CEPI consultant)

Executive summary

On 26-27 June 2025, the Coalition for Epidemic Preparedness Innovations (CEPI) hosted a technical consultation on mpox correlates of protection (CoP), with a focus on mRNA vaccine candidates. The consultation brought together experts in vaccine development, regulatory science, epidemiology, modeling, clinical trial design, One Health, pre-clinical models, immunological assays, and vaccine policy to review available evidence and identify the evidence gaps most relevant to next-generation mpox vaccine development.

Licensed vaccinia virus (VACV)-based vaccines, including MVA-BN and LC16m8, are available for mpox prevention, which can make placebo-controlled efficacy trials ethically and operationally challenging. At the same time, next-generation subunit and RNA-based mpox vaccines express a limited number of MPXV antigens and therefore cannot necessarily rely on direct immunobridging to whole-virus vaccinia-based vaccines, which encode many viral proteins and elicit broader immune profiles. As a result, identifying immune biomarkers reasonably likely to predict clinical benefit is critical for supporting alternative licensure pathways for novel mpox vaccines.

The consultation focused on the current landscape of mpox vaccine candidates, evidence from natural and vaccine-induced immunity, immunological endpoints, assay development and harmonization, pre-clinical models, passive transfer studies, and regulatory considerations. Across sessions, participants emphasized that no validated human CoP for mpox has yet been established; however, there is a plausible pathway to build an evidence package using animal challenge studies, passive transfer data, human immunogenicity data, assay standardization, and regulatory alignment on relevant endpoints.

Executive summary: key messages by session

Session 1: Introductory session

- In the context of ongoing mpox outbreaks, limited vaccine availability, and unmet product needs, mRNA and other next-generation platforms could contribute to future preparedness if they offer advantages in manufacturing scalability, affordability, thermostability, and durability of protection.
- Preferred product characteristics include durability of at least 12 months, 2-8°C storage where feasible, low cost of goods (<$10 per dose), and manufacturing processes that can support rapid outbreak response and sustainable access in endemic countries.
- Establishing immune biomarkers or CoP is important because conventional phase 3 efficacy trials may be difficult to conduct, and immunobridging to licensed VACV-based vaccines may not be directly transferable to subunit or RNA-based vaccines.
- Early clinical and preclinical data from mRNA and saRNA vaccine candidates support continued development, but additional evidence is needed on durability, clade breadth, assay comparability, and relevance of immune endpoints to protection.

Session 2: Natural versus vaccine-induced immunity

- Natural MPXV infection and VACV- or mpox-vaccine-induced immunity provide complementary information for defining protective immune responses.

- Comparing longitudinal immune responses after natural infection and vaccination can help determine the extent to which vaccine-induced immunity recapitulates immune responses associated with protection after infection.
- Breakthrough infections and reinfections provide important opportunities to identify candidate biomarkers associated with protection by comparing immune responses among individuals with similar exposure risk who do or do not become infected.
- Available evidence suggests that neutralizing antibodies may decline over time, while protection may persist through immune memory and cellular immune responses; therefore, waning antibody levels should not be equated with waning immunity without supporting evidence.

Session 3: Immunological endpoints and assays

- Assays used to support CoP development should be fit for purpose, validated or qualified as appropriate, and capable of generating comparable data across studies, species, and laboratories.
- High sequence conservation across orthopoxviruses results in substantial antibody cross-reactivity, making it difficult to distinguish responses induced by natural MPXV infection, vaccination, or prior orthopoxvirus exposure unless assays are carefully designed and interpreted.
- Neutralizing antibodies remain plausible candidate biomarkers, but protection may also involve binding antibodies, Fc-mediated effector functions, complement deposition, NK-cell activation, phagocytosis, mucosal immunity, B-cell memory, and T-cell responses.
- CEPI-supported assay harmonization activities, including the Centralized Laboratory Network and international antibody standards, are important for improving comparability and future biomarker standardization.

Session 4: Correlates of protection and evidence evaluation

- There is currently no standardized framework for assessing the strength of preclinical evidence supporting pre-clinical models, assays, and candidate biomarkers for vaccine assessment.
- The FEEVA initiative aims to develop a transparent, GRADE-like framework for evaluating evidence generated outside traditional efficacy trials, including pre-clinical models, assays, controlled human infection models, and natural history or real-world evidence.
- Evidence that VACV-binding antibodies may associate with protection against mpox has limited transferability to subunit or mRNA vaccines because these platforms induce different antigen-specific immune responses.

Session 5: Regulatory pathway

- Regulatory pathways for novel mpox vaccines will depend on the intended indication, the feasibility of efficacy trials, and the strength of evidence linking immune biomarkers to protection.
- A credible evidence package may require animal challenge studies, passive transfer studies, human immunogenicity data, and carefully justified immunobridging approaches.
- Passive transfer studies can provide direct evidence of antibody-mediated protection, but they do not capture contributions from cell-mediated immunity or immune memory.
- Binding antibodies may be acceptable biomarkers if a robust and biologically meaningful relationship with functional measures, such as neutralization, can be demonstrated for the relevant vaccine candidate and assay context.

Session 6: Pre-clinical models

- The ideal mpox pre-clinical model would reproduce relevant human disease manifestations, including lesional disease and severe outcomes, using biologically plausible routes and challenge doses.
- It is important to distinguish between pre-clinical models that best mimic human disease and those best suited to evaluate vaccine efficacy; both considerations matter for translational relevance.
- Overly artificial, high-dose, or non-physiological challenge models may limit the relevance of identified immune correlates for human immunobridging.

Detailed report

The Technical Consultation on Mpox Correlates of Protection aimed to review the pipeline of mpox mRNA and other next-generation vaccine candidates, compare existing evidence and gaps in understanding natural versus vaccine-induced immunity, evaluate immunological endpoints and assay-related advances and challenges, and discuss pre-clinical models and alternative licensure pathways relevant to identifying immune markers associated with protection. The meeting also served as a consultation on supportive data for surrogate endpoints, immunobridging strategies, and regulatory considerations for licensure. Key outcomes include defining the elements needed to build robust data packages for next-generation mpox vaccines and identifying critical gaps, expert recommendations, and next steps for the mpox CoP field.

Reference to final agenda to be inserted as an appendix.

Session 1: Introductory session

Research and development perspective to addressing mpox unmet needs

Mpox continues to circulate globally, with active transmission in several African countries. A major challenge remains the mismatch between vaccine availability and country needs for outbreak control. Low- and middle-income countries face limited access to licensed vaccines beyond donations, suboptimal target product characteristics, constraints in rapidly scaling manufacturing, unclear use cases and demand, and sustainability challenges for access over time.

The current R&D ecosystem is not fully prepared to respond to multiple mpox scenarios, ranging from localized outbreaks to broader prophylactic use in endemic areas. A coordinated research roadmap published in September 2024 identified immediate priorities for mpox research, including the use of pre-clinical models and assays to identify CoP and pathways for licensing next-generation medical countermeasures when licensed products already exist.

New vaccine platforms, including mRNA, saRNA, protein subunit, and VLP-based approaches, may offer advantages if they can improve affordability, speed and scalability of manufacturing, cost of goods, thermostability, and durability of immunity. These attributes are important for outbreak response and for sustainable use in endemic countries.

Definitions and considerations for identifying correlates of risk and protection

It is important to distinguish correlates of risk from correlates of protection. Correlates of risk measure how well immune markers predict a particular mpox-related endpoint within a given cohort or animal challenge model, whereas CoP measure how well immune markers predict, cause, or mediate vaccine efficacy. A correlate of risk may inform progress toward a CoP but is not necessarily sufficient for approval of decisions.

Each candidate CoP is tied to a specific assay and endpoint. Therefore, assay selection and validation are central to evidence generation. Reliable evidence requires assays with high precision, low measurement error, and sufficient dynamic range among vaccinees. Harmonizing assays across studies is key for comparability, and opportunities to establish correlates exist throughout vaccine development, from preclinical studies to post-licensure evaluation.

Mpox situation on the African continent

At the time of this meeting (Epi week 24 of 2025), Mpox cases in Africa were beginning to decline with Sierra Leone accounting for a substantial proportion of confirmed cases. Clades Ib and IIb continued to drive transmission, and most confirmed cases since January 2024 had occurred in Democratic Republic of Congo, Uganda, Burundi, and Sierra Leone. High test positivity suggested ongoing community transmission, and indicators beyond case fatality rate, including health-system burden, remain important for assessing outbreak impact.

Several African countries have received vaccines and some have implemented active vaccination. Priorities include expanding testing and surveillance, increasing vaccination, and ensuring regulatory approval. Dose-sparing

approaches, including one-dose or fractional-dose strategies, may support the current response but must be considered alongside durability, supply constraints, and equity considerations.

Landscape of current mpox mRNA vaccines

The mpox vaccine pipeline includes a broad range of technologies, with several candidates in preclinical or exploratory development and a smaller number already registered, stockpiled, or under clinical evaluation. mRNA technology has the capacity to scale rapidly, an important factor for outbreak response. Having more than a limited number of vaccine options in the mpox armamentarium would strengthen preparedness and access.

CEPI's platform readiness assessments evaluate characteristics such as durability, safety, thermostability, cost, and prior knowledge to prioritize rapid-response candidates. The target platform should offer potential for rapid response with durability of at least 12 months, adequate 2-8°C stability where feasible, and low cost of goods. These considerations are directly relevant to future mpox preparedness and response.

Clinical update on orthopox vaccine candidate mRNA-1769

Moderna's vaccine candidate mRNA-1769 includes two mature virion and two enveloped virion antigens with high amino-acid conservation across orthopoxviruses. Interim Phase I/II data in adults indicate acceptable safety and tolerability across evaluated doses, with mostly Grade 1-2 solicited adverse reactions. Immunogenicity data showed increases in mpox- and vaccinia-neutralizing antibodies, strong binding antibody responses to vaccine antigens, and robust CD4 with modest CD8 T-cell responses. Long-term immune-response data remain forthcoming.

Gennova's saRNA platform for mpox

Gennova's self-amplifying RNA mpox vaccine candidate includes viral antigens linked to protection in small animal studies and is currently in preclinical testing. The saRNA platform can support multivalent antigen expression and may offer advantages related to dose-sparing, stability, and suitability for LMIC contexts. No data are currently available in the public domain.

Session 2: Key characteristics of mpox natural immunity versus immunity conferred by vaccination

Observational approaches for generating evidence on CoP

Phase 3 trials remain an important component of vaccine development, but the design and evidentiary role of such studies may differ when traditional pivotal efficacy trials are ethically or operationally difficult. For mpox, the existence of licensed vaccines and the fluctuating nature of outbreaks complicate placebo-controlled efficacy trials and may require alternative evidence packages supported by robust biomarkers and pre-clinical-model data.

Observational studies can help generate evidence on mpox CoP using longitudinal cohorts, field vaccine-effectiveness studies, household transmission studies, and breakthrough infection registries linked to interventional trials. These studies can address infection route, clade-specific differences, immune longevity, immune repertoire, prior smallpox vaccination, and asymptomatic infection. Key study design considerations include collection of baseline and longitudinal biospecimens where feasible, clearly defined clinical endpoints, robust exposure assessment, adequate follow-up duration, and standardized, fit-for-purpose immunological assays.

Breakthrough infections and reinfections are particularly informative because they can allow comparison of immune-marker levels between individuals who become infected and those who remain uninfected despite similar exposure opportunity. Such comparisons may help identify biomarkers associated with protection or reduced disease severity.

Immune CoP against mpox

Subunit and RNA-based mpox vaccines are expected to encode a limited set of MPXV antigens, often representing both mature virion and enveloped virion forms. These virion forms contain distinct, non-overlapping antigens, and immune responses against one form may not fully capture protection against the other. Therefore, vaccine antigen selection and assay strategies should consider both MV- and EV-specific responses.

Because whole-virus VACV-based vaccines encode many viral proteins and elicit broad immune responses, evidence generated with those vaccines may not directly transfer to MPXV-antigen-specific subunit or RNA-based vaccines. Existing evidence on biomarkers associated with protection may therefore have reduced transferability across vaccine platforms, necessitating platform- and antigen-specific evidence to establish relevant CoP.

Long-term follow-up of immunological consequences of infection versus MVA-BN vaccination

Ongoing mpox cohort studies in Belgium and DRC provide important opportunities to compare immune responses after natural infection and vaccination. Longitudinal analyses suggest that people with prior MPXV infection can show strong immunological memory, while responses after MVA-BN vaccination in vaccinia-naïve individuals may be less robust and shorter-lived. Historic smallpox vaccination may also influence current immune responses by boosting pre-existing orthopoxvirus immunity.

Interpretation of these data requires careful attention to prior vaccination, clade, exposure risk, HIV or immunocompromised status, and the specific assays used. Comparing natural and vaccine-induced immunity can help define which immune responses are most closely associated with protection and whether vaccine-induced immunity recapitulates protective immune profiles observed after infection.

Immunology of LC16m8 vaccine

LC16m8 is an attenuated VACV strain approved in Japan for smallpox and mpox. Preclinical and clinical immunogenicity and safety data demonstrate antibody responses in mice, non-human primates (NHPs), and humans, with evidence of responses to VACV proteins and selected MPXV proteins. LC16m8 has demonstrated neutralizing antibody responses across MPXV clades and cross-protection in pre-clinical models. Additional Japanese non-replicating or attenuated vaccine candidates may provide further information on humoral and cellular immunity, but their relevance to CoP development depends on the specific construct, model, endpoint, and assay used.

When immunity is not absolute: clinical presentations after past infection or vaccination

Breakthrough infections and reinfections can occur after prior infection or vaccination, but observational data indicate that prior immunity can reduce clinical severity. Rather than treating breakthrough infections only as vaccine failures, these events should be considered important opportunities to identify immune biomarkers associated with protection, reduced disease severity, or reduced transmission potential.

Future studies should compare immune responses in breakthrough cases, reinfected individuals, and exposed uninfected controls, while accounting for vaccination status, prior infection, exposure likelihood, and timing of biospecimen collection. Neutralizing antibody levels may decline over time, but durable B-cell memory, T-cell responses, and non-neutralizing antibody functions may contribute to protection. Therefore, data interpretations should distinguish waning antibody levels from waning immunity.

Global equity and booster policy considerations are important but should be addressed separately from the mechanistic question of how incomplete protection can inform CoP discovery. Vaccine strategies should be context-specific and should balance durability, supply, access, safety, and evidence on which immune components are most relevant for protection.

Session 3: Immunological endpoints in the context of mpox CoP

Landscaping of mpox immunological assays

To date, mpox immunological data have mainly evaluated binding antibodies, historically often to whole-cell lysate material, with more recent work considering responses to specific recombinant antigens. Viral neutralization has focused largely on vaccina vaccine-induced responses, with additional studies now considering mpox-specific neutralization and clade-specific readouts. T-cell response data are more limited and have often used IFN-γ ELISpot formats with VACV-infected cell lysates or commercial pan-pox peptide mixtures.

Assays must be selected according to the vaccine antigen, endpoint, and intended evidentiary role. MPXV-based assays preferentially measure MPXV-specific responses, while VACV-based assays emphasize vaccinia-induced immunity. High orthopoxvirus sequence conservation results in substantial antibody cross-reactivity, which can complicate attribution of responses to infection, vaccination, or prior orthopoxvirus exposure. This cross-reactivity can introduce biologically irrelevant signals into assay readouts and weaken evidence supporting a CoP unless carefully addressed.

Ongoing observational and epidemiological studies collecting longitudinal samples from cases, vaccinees, or high-risk contacts may support CoP identification, provided that the assays used are well characterized and sufficiently harmonized. The CEPI Centralized Laboratory Network is developing and validating immunological assays relevant to mpox vaccine development, including orthopoxvirus neutralization assays and T-cell assays, with efforts to transfer assays to partner laboratories, including African laboratories.

CEPI is also supporting a WHO collaborative study to establish international antibody standards for MPXV and VACV. International standards and reference reagents should improve assay calibration, facilitate comparison across laboratories, and support future biomarker standardization, including readouts from neutralization assays.

Mpox assays: advances and challenges

Several immunological assay platforms for mpox are under development or qualification, including MVA-based neutralization assays, live MPXV neutralization assays requiring higher containment, multiplex binding assays against key MPXV antigens, T-cell ELISpot assays using pan-pox peptide mixtures, and non-neutralizing antibody-dependent complement deposition assays. Key challenges include limited availability and volume of suitable clinical samples, narrow analytical range in some neutralization assays, limited data on assay performance characteristics, and the need for appropriate reference materials for multiplexed assays.

Understanding the contribution of MV- versus EV-specific neutralizing antibodies is important because MV and EV have distinct antigenic targets. Neutralizing antibodies against one virion form may not effectively neutralize the other. EV neutralization assays are technically demanding, and both MV and EV readouts may be needed to interpret protective immunity. For binding assays, differences in target antigens, assay format, and readouts make it difficult to compare data across studies without harmonization.

Overview of serological assays for mpox vaccine assessment

Complement-based neutralization assays and PRNT formats may provide informative data, but their use as surrogates of protection requires standardization, validation, and clear evidence linking assay results to protection. Current evidence supports the importance of both functional and binding antibody measures, although neutralizing antibody responses alone may not fully capture protective immunity. Binding antibody responses may be acceptable in regulatory contexts if a robust relationship with functional measures such as neutralization is demonstrated for the relevant vaccine and assay context.

Highly immunogenic antigens such as A35R/A33R and B6R/B5R elicit strong responses and are relevant to candidate vaccine design and biomarker evaluation. However, broad binding cross-reactivity does not necessarily indicate functional neutralization breadth across MPXV clades or orthopoxviruses. Therefore, assay strategies should focus on

clinically meaningful readouts, appropriate antigens, and well-justified testing against relevant MPXV strains or clades.

Systems-level analysis of vaccine-induced preclinical humoral CoP

Vaccine effectiveness estimates for licensed vaccines vary across studies and populations, and the clinical endpoint of interest should be clearly specified, such as infection, symptomatic disease, severe disease, lesion burden, or transmission-related outcomes. Neutralizing antibodies remain important candidate biomarkers, but they may wane rapidly, whereas memory B-cell responses, T-cell responses, and non-neutralizing antibody functions may contribute to durable protection.

Emerging data suggest that mucosal viral control may involve neutralizing antibodies together with Fc-mediated mechanisms, including NK-cell activation, complement deposition, and phagocytosis. Protection against lesions and transmission-related pathology may involve immune mechanisms beyond neutralization alone. These findings support a broader, multi-parameter view of protective immunity while recognizing that regulators generally require a clearly justified and validated primary immunogenicity readout.

Assay selection and sample testing should be aligned with the intended mpox indication. For mpox-specific indications, MPXV-relevant neutralization and binding assays are likely to be more informative than VACV-only readouts. Broader orthopoxvirus claims would require wider cross-virus testing. Inclusion of licensed vaccines such as MVA-BN or ACAM2000 may help contextualize immune responses but should not be used to assume direct immunobridging across substantially different vaccine platforms.

Session 4: Correlates of protection

Assessing the evidence for vaccine effectiveness

In an ideal product approval pathway, a pivotal randomized controlled trial demonstrating clinical effectiveness supports regulatory approval, followed by additional evidence from clinical studies, real-world data, systematic reviews, and meta-analyses. For mpox and other outbreak-prone diseases, traditional efficacy trials may be difficult to conduct, creating a need to assess evidence generated through pre-clinical models, assays, controlled human infection models, natural history studies, and real-world evidence.

There is no widely accepted framework for assessing or comparing the adequacy of data from pre-clinical models and immune biomarkers for vaccine assessment. The FEEVA project was launched to develop a transparent, GRADE-like framework for evaluating evidence generated outside standard efficacy trials across domains including pre-clinical models, assays, controlled human infection models, and natural history or real-world evidence. Such a framework could help structure evidence discussions, guide study design, reduce low-value studies, and support alignment among developers, regulators, funders, and public-health stakeholders.

Immunobridging as a basis for licensure: lessons from Ebola vaccine development

A nontraditional licensure pathway using immunobridging was applied for Janssen's Ebola vaccine, where immune responses associated with protection in NHPs were used to infer likely clinical benefit in humans. This approach linked vaccine-induced immune markers in animals to survival and compared those responses with human immunogenicity data. Both animal and human samples were tested in the same assay to enable bridging. This example demonstrates that animal-to-human translational approaches can support regulatory decision-making when efficacy trials are not feasible, although they require strong assay performance, robust pre-clinical-model data, and early regulatory engagement.

For mpox, lessons from Ebola are relevant but not directly transferable. The protective immune mechanisms, available pre-clinical models, vaccine platforms, and assays differ. Therefore, any mpox immunobridging approach should be

justified based on the specific vaccine antigen composition, immune endpoint, pre-clinical model, and intended indication.

CEPI's CoP program and how correlates may support CEPI's mission

CEPI's mission to enable vaccines to be ready for initial authorization and large-scale manufacturing within 100 days of recognizing a pandemic pathogen depends in part on identifying reliable immunological markers and CoP for priority pathogens. CoP and biomarkers can accelerate vaccine development, support immunobridging, optimize dosing and regimens, de-risk late-phase trials, guide rational vaccine design, and inform policy decisions. Challenges include inconsistent data collection and analysis, limited harmonization, fragmented stakeholder coordination, and unclear communication of evidence requirements.

CEPI's CoP strategy and playbook aim to consolidate evidence mapping, systematic reviews, biomarker suitability assessment, gap analysis, regulatory insights, and vaccine-development strategies. For mpox, these activities should align with ongoing assay harmonization, pre-clinical-model evaluation, and evidence-generation plans for next-generation vaccine candidates.

Evidence and limitations of CoP against mpox

A systematic review of vaccine-effectiveness evidence related to mpox indicates that VACV-binding antibodies may be associated with protection against mpox, but current evidence is limited and requires further validation. Even if confirmed, a vaccinia-binding antibody biomarker would have limited utility for subunit or RNA-based mpox vaccines because these platforms express a limited number of MPXV antigens and induce different immune-response profiles.

For subunit and RNA-based vaccine candidates, MPXV-specific functional measures, such as neutralization against relevant MPXV strains or clades, may be more informative. However, neutralizing antibody responses alone may not fully explain protection, and the role of binding antibodies, Fc-mediated functions, mucosal immunity, B-cell memory, and cellular immunity should be considered as supportive evidence. The totality of evidence will be important for regulatory confidence.

Session 5: Regulatory pathway

How can CoP help the regulatory process for licensure of new mpox mRNA vaccines?

There are significant challenges with conducting traditional efficacy trials for mpox, including ethical and operational constraints created by the availability of licensed vaccines and the unpredictable nature of outbreaks. Regulatory pathways for new mpox vaccines will depend on the intended indication, the feasibility of efficacy studies, the relevance of pre-clinical models, the strength of immune-marker evidence, and the availability of standardized assays.

A credible evidence package for next-generation mpox vaccines may require a combination of animal challenge studies, passive transfer studies, human immunogenicity data, and carefully justified immunobridging approaches. NHP experiments may be important to generate curves relating protection to neutralizing antibody or other immune-marker levels, ideally identifying thresholds or ranges associated with protection. Additional data across MPXV clades may add value, although the need for cross-clade challenge studies should be justified by the intended indication and available immunogenicity data.

Passive transfer studies can provide direct evidence of antibody-mediated protection and help identify functional antibody levels associated with protection. However, passive transfer studies do not capture contributions from B-cell memory, T-cell immunity, or anamnestic responses and therefore provide only a partial view of vaccine-induced protection. They are most informative when interpreted alongside active vaccination challenge studies and human immunogenicity data.

Clinical immunobridging to MVA-BN is complex because MVA-BN is a whole-virus vaccinia-based vaccine that expresses many antigens, whereas mRNA or subunit vaccines typically encode a limited number of MPXV antigens.

Clinical immunobridging alone is unlikely to be sufficient unless supported by fit-for-purpose pre-clinical-model data and a scientifically justified immune endpoint. Neutralizing antibodies remain plausible biomarkers associated with protection, but they should be interpreted within a broader immunological framework. Binding antibodies may be acceptable biomarkers if they show a strong and consistent relationship with relevant functional measures.

Cell-mediated immunity is unlikely to be the pivotal component of an initial licensure package unless strongly justified, but it should be included as supportive evidence because it may contribute to disease control, memory responses, and durability. Regulatory engagement should focus on intended indication, clinical endpoint, pre-clinical model relevance, assay selection, biomarker thresholds, and the totality of benefit-risk evidence.

Session 6: Pre-clinical models of mpox virus infection and disease

Pre-clinical models in mpox CoP research

In traditional licensure pathways, pre-clinical models are used primarily in preclinical studies to assess safety, immunogenicity, and occasionally efficacy before progression to human studies. Nontraditional pathways may require well-characterized pre-clinical models that demonstrate vaccine efficacy and provide immune endpoints translatable to humans. Pre-clinical models can support fundamental CoP work, including assessment of antibody-mediated protection through passive transfer and evaluation of cellular responses.

The ideal mpox pre-clinical model would mimic both lesional disease and severe outcomes observed in humans using biologically plausible challenge doses and routes. It is important to distinguish between a model that mirrors human disease and a model that tests vaccine potential for human use; both are relevant but serve different purposes. Mild disease models may not reveal vaccine effects, while overly artificial, high-dose, or non-physiological challenge models may reduce translational relevance for human immunobridging.

Natural transmission routes, including cutaneous or sexually associated mucosal exposure, should be considered when feasible to ensure that identified immune correlates are relevant to human protection. Pre-clinical models can also inform clade virulence, route-dependent disease, rapidity of protection, and differential contributions of MV- and EV-specific responses. However, limitations include evolving clades, variable disease severity across species, ethical considerations, high containment requirements, NHP availability, cost, and logistical constraints.

Pre-clinical-model strategies should also consider the 3Rs—replacement, reduction, and refinement of animal use. Alternative systems may be useful for toxicity, safety, or mechanistic studies but are less likely to replace animal challenge models for evaluating protective vaccine effects unless validated against historically accepted models.

Passive transfer studies

Passive transfer studies are particularly useful when functional antibodies appear to be a major driver of protection. They can test whether defined quantities or qualities of antibodies from vaccinated humans or animals protect against challenge and can support identification of antibody levels reasonably likely to predict clinical benefit. Nonetheless, passive transfer does not capture cell-mediated immunity (CMI), immune memory, or vaccine-induced anamnestic responses, and therefore should be integrated with other evidence sources.

Care is needed when comparing antibodies elicited by different vaccines or platforms, particularly when comparing VACV-specific and MPXV-specific responses. Equal neutralizing titers may not necessarily imply equal protective capacity if antibody specificity, virion-form coverage, Fc functions, or assay context differ. Combining MV and EV neutralization data, and potentially linking functional readouts to simpler binding assays, may be informative but requires empirical support.

Mpox multiprotein virus-like nanoparticle vaccine

VLP-based vaccine approaches using MPXV proteins such as M1, A35, and B6 have demonstrated immunogenicity and protection in pre-clinical models. Combination MV and EV VLPs have shown protection in mice against lethal

orthopoxvirus challenges and can induce neutralizing antibody responses. Passive transfer data may help clarify the contribution of antibody-mediated protection. Future work should assess durability of antibody responses, the role of scaffold-specific immune responses, and comparability with other platforms such as mRNA vaccines.

mRNA-1769 preclinical development

mRNA-1769 preclinical studies demonstrated broad neutralization and protection across diverse orthopoxvirus models, including dose-ranging studies in NHPs and challenge studies in mice and rabbits. Passive transfer of immune sera provides additional evidence for the protective role of antibodies, while durability of protection remains an important gap that regulators may raise. Long-term protection may depend on both antibody levels and memory responses, and booster studies could help define how rapidly protective humoral responses are re-established.

BNT166 preclinical development overview

BNT166a is an investigational quadrivalent mRNA orthopoxvirus vaccine encoding MPXV EV antigens A35 and B6 and MV antigens M1 and H3. Preclinical data indicate robust antibody and T-cell responses and protection in pre-clinical models against VACV and MPXV clades. Clinical evaluation is ongoing in vaccinia-naïve and vaccinia-experienced participants. Future studies should continue to clarify the relevance of infection route, challenge model, antigen specificity, and assay readout for CoP development.

Key Conclusions and Next Steps

- No validated human CoP currently exists for mpox. A structured evidence-generation strategy is needed to identify immune biomarkers reasonably likely to predict clinical benefit.
- Neutralizing antibodies remain leading candidate biomarkers, but they are unlikely to fully explain protection. Binding antibodies, Fc-mediated functions, mucosal immunity, B-cell memory, and T-cell responses should be considered as supportive or complementary evidence.
- Assay selection is central to CoP development. Assays should be fit for purpose, validated or qualified as appropriate, and harmonized across laboratories through reference reagents, international standards, monitoring programs, and shared protocols.
- Evidence from licensed VACV-based vaccines is informative but not directly transferable to MPXV-antigen-specific subunit or RNA-based vaccines. Platform-specific and antigen-specific evidence will be needed.
- Longitudinal studies of naturally infected individuals, vaccinees, breakthrough cases, reinfections, and exposed uninfected controls are essential for identifying biomarkers associated with protection, reduced severity, or durability.
- Well-characterized pre-clinical models and passive transfer studies will likely be central components of alternative licensure pathways, but pre-clinical-model relevance, challenge route, clinical endpoint, and assay readout must be carefully justified.
- Regulatory alignment should focus on intended indication, endpoints, acceptable evidence packages, biomarker thresholds or ranges, assay validation expectations, and how totality of evidence will be assessed.

Supplemental Appendix S-1. Mpox CoP Workshop Agenda

"Technical Consultation on Mpox Correlates of Protection (CoP): with Focus on mRNA Vaccine Candidates," 26-27 June 2025. Coordinated by the Coalition for Epidemic Preparedness Innovations (CEPI).

Meeting Objectives for the technical consultation:

- Review the landscape and pipeline of current mpox mRNA vaccine candidates.
- Review the key characteristics of mpox natural immunity vs immunity conferred by smallpox and/or mpox vaccination.
- Review the immunological endpoints in the context of mpox CoP including assay related advances and challenges.
- Immune correlates from pre-clinical models: Explore surrogate outcomes and lethal challenge studies in pre-clinical models for identification of immune markers.
- Regulatory pathway to licensure: Focus on surrogate endpoints, immunobridging strategy and guidance from regulators relevant to licensure of mpox mRNA vaccines.

Acronyms:

Africa CDC -- Africa Centres for Disease Control & Prevention

CEPI – Coalition for Epidemic Preparedness & Response

ITG – Instituut voor Tropische Geneeskunde [Institute of Tropical Medicine—Antwerp]

NIAID – US National Institute of Allergy & Infectious Diseases

PATH – formerly Program for Appropriate Technology in Health

UKHSA – UK Health Security Agency

USAMRIID – US Army Medical Research Institute of Infectious Disease

WHO – World Health Organization

Day	Sessions/Topics	Speakers/Moderator
Day 1: **June 26**	**Session 1: Introductory Session:** CEPI's efforts on mpox, introduction for CoP, consultation objectives, landscape of mpox mRNA vaccines and mRNA vaccine candidates R&D perspective to addressing mpox unmet needs Objectives of Mpox CoP Technical Consultation CoP concept and relevance for mpox	Moderator: Nina Wressnigg (CEPI) Kristine Rose (CEPI) Haritha Adhikarla (CEPI)

	Overview of the epidemiological situation of mpox in Africa Vaccine Platform Readiness Dashboard Landscape of current mpox mRNA vaccines Clinical Update on Orthopox Vaccine Candidate mRNA-1769 Gennova's saRNA Platform for mpox Roundtable: Key summary points Discussion/questions.	David Benkeser (CEPI) Mercy Kyeng (Africa CDC) Cathy Hoath (CEPI) Philipp Mann (CEPI) Hiwot Hiruy (Moderna) Sanjay Singh (Gennova) Moderator: Nina Wressnigg (CEPI)
Day 1: **June 26**	**Session 2**: Key characteristics of mpox natural immunity vs immunity conferred by (smallpox and/or mpox) vaccination Observational approaches for generating evidence on CoP Immune correlates of protection against mpox Long-term follow up of immunological consequences of MPXV infection vs MVA-BN vaccination Immunology of LC16m8 and new mpox vaccine candidate (Japan) When Immunity Isn't Absolute: Mpox Clinical Presentations After Past Infection or Vaccination Roundtable: Key summary points Discussion/questions	Moderator: Rosamund Lewis (WHO) Daniel Rhee (CEPI) Dan Barouch (Harvard Medical School) Christophe Van Dijck (ITG) Ken Ishii (University of Tokyo) Aniruddha Hazra (University of Chicago) Moderator Rosamund Lewis (WHO)
Day 1: **June 26**	**Session 3:** Immunological endpoints in the context of mpox CoP	Moderator: Bassam Hallis (UKHSA)

	a) Landscaping of Mpox Assays/Challenges	
	Overview of CEPI Centralized Lab Network mpox efforts	Gathoni Kamuyu (CEPI)
	b) Mpox Assays: Advances and Challenges	
	Mpox assays: Advances and Challenges	Sue Charlton (UKHSA)
	Overview of serological assays for mpox vaccine assessment	Alessandro Manenti (VisMederi)
	A systems level analysis of vaccine induced preclinical humoral correlates of protection against mpox	Galit Alter (Ragon Institute)
	Roundtable: Key summary points Discussion/questions	Moderator: Bassam Hallis (UKHSA)
	Wrap up of Day 1 Sessions	Haritha Adhikarla
Day 2: **June 27**	**Session 4: Correlates of protection**	Moderator: Kristine Rose (CEPI)
	A Framework for Evidence Evaluation in Vaccine Assessment (FEEVA)	Miles Davenport (Kirby Institute)
	Immunobridging Strategy: Lessons learnt from Ebola vaccine	Jenny Hendriks (CEPI)
	CEPI's CoP Program	Christine Dahlke (CEPI) and
	Stepwise development plan for mpox playbook to ensure alignment with use cases	Rama Raghunandan, Cynthia Lee (PATH)
	Evidence and limitations of a correlate of protection against mpox.	David Khoury (Kirby Institute)
	Roundtable: Key summary points Discussion/questions	Moderator: Peter Dull (Gates Foundation)

Day 2: **June 27**	**Session 5: Regulatory Pathways** (closed session, excluding developers) a) How can CoP help the regulatory process for license of new mpox mRNA vaccines? b) Regulatory pathway related guidance	 Debra Yeskey (CEPI) Regulatory agency representatives
Day 2: **June 27**	**Session 6: Pre-clinical models of mpox virus infection and disease** Explore surrogate outcomes, lethal challenge studies in pre-clinical models for identification of immune markers Pre-clinical models in mpox CoP research Mpox multiprotein virus-like nanoparticle vaccine induces neutralizing and protective antibodies in pre-clinical models. mRNA-1769 broadly protects against diverse *Orthopoxviruses* in preclinical models BNT166 mpox vaccine pre-clinical development overview Roundtable: Key summary points discussion/questions	Moderator: Jay Hooper (USAMRIID) Victoria Graham (CEPI) Bernard Moss (NIAID) Alec Freyn (Moderna) Adam Zuiani and Leela Davies (BioNTech) Moderator: Jay Hooper (USAMRIID)
Day 2: **June 27**	**Roundtable:** Key summary points for regulatory pathway (open for all developers) Closing remarks Acknowledgements	Moderator: Adam Hacker (CEPI) Jakob Cramer (CEPI) Haritha Adhikarla (CEPI)