Role of Cryo-Electron Microscopy of Arena Virus

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Growth Project (Livestock Component) World Bank Assisted, Government of Sindh, Hyderabad-Sindh, Dr. Benjamin W. Neuman, School of Biological Sciences, University of Reading, UK. ⁽³⁾ M.Ali Bhatti, Assistant Professor IBA-Sulkkur ⁽⁴⁾ Dr. Gul Hassan Sethar-Consultant PhysicianAl- Amiri Hospital Kuwait city, Kuwait ⁽⁵⁾ Dr. Nargis Khan- Senior Registrar Medicine Department Dow University of Health Sciences, Karachi

Abstract: The current research Role of Cryo-Electron Microscopy of arena virus It was revealed that The natural hosts of Junin virus are rodents, particularly Mus musculus, Calomys spp and Akodon azarae (Vesper Mouse). Direct rodent to human transmission only transpires when contact is made with excrement of an infected rodent. Hazardous group is 4. This commonly occurs via ingestion of contaminated food or water, inhalation of particles within urine or via direct contact of broken skin with rodent excrement. As described in material method about CTFit an image generated after Contrast transfer function is given in fig 9 showing clear difference of image before and after CTFit.

Key words: Role, Cryo-Electron, Microscopy. Arena virus

Introduction:

(1)

Low-dose Cryo-EM was performed at 120kV as described previously (26). The electron dose was minimized to reduce specimen damage. Digital Cryo-EM images were recorded by two methods. SARS-CoV film negatives were digitized using a Zeiss SCAI microdensitometer at a final resolution of 1.84 Å/pixel. All other images were recorded directly via a CCD image sensor at a resolution of 2.26 Å/pixel using Leginon (35). The signal strength and signal-to-noise ratio differed subtly depending on recording methodology. Image contrast was inverted so that protein density appeared white. The density histogram for each image was normalized to a common median grey value prior to analysis. How electron beam hits the sample is shown in fig 4.

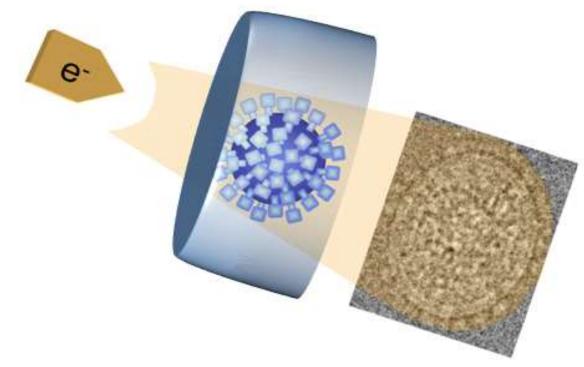


Figure 4.Cryo-EM: Cryo-Electron microscopy is a form of electron microscopy where the sample is studies at cryogenic temperatures. The Cryo-EM imaged sample is safe from any damage caused by radiation. It also allows the samples to be imaged without stained or fixed. In this way samples is just in a native environment. **Arenaviruses used in this study:**

Lymphocytic Choriomeningitis Virus (LCMV).

Lymphocytic Choriomeningitis is a rodent borne viral infectious disease caused by Lymphocytic Choriomeningitis Virus (LCMV). LCMV also causes aseptic meningitis, encephalitis or Meningoencephalitis.

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Its name was coined by Charles Armstrong in 1934. LCMV provides one of the most widely used model systems to study viral persistence and pathogenesis (24, 27). LCMV is found worldwide, probably because of its association with the common house mouse Mus musculus (30).

LCMV is an Old World arenavirus (see fig 5). Its hosts are Mus musculus, Mus domesticus and it is classified as a hazardous group 3 agent in the UK. LCMV causes aseptic meningitis, encephalitis or meningoencephalitis disease.

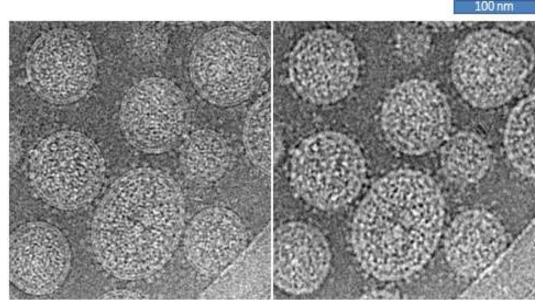


Figure 5. Cryo-EM images of elliptical and round LCMV particles.

Pichinde virus (PICV):

Pichinde virus (PICV) belongs to the New World arenaviruses (8) of clade A (see fig 6). Its natural host is Oryzomys albegularis (Tomes's Rice Rat) and it was discovered in Columbia in 1967 (36). It is a hazardous group 2. It does not cause any disease in humans but pathogenic in guinea pigs and hamsters.

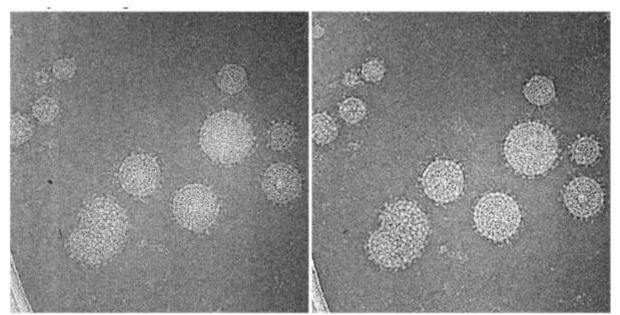


Figure 6. Pichinde Virus Cryo-EM images show the natural variation in size and shape.



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Tacaribe virus (TCRV) is the prototype of the taxonomic group of Tacaribe virus complex together with other species of New World arenaviruses (see Fig 7). It was first found in the blood of *Artibeus* bat species in 1956 in the Gran Tacaribe Cave on the north coast of Trinidad. This virus does not cause any significant disease and not a human as dangerous human pathogen as Junin virus and other new world arenavirus. To date only a single case of febrile disease caused by Tacaribe virus known. The Tacaribe virus currently found in fruit bat populations on West Indies and Jamaica. So far, two subtypes have been isolated, the Tacaribe virus p2b2 (V5 and V7) and TRVLII573.

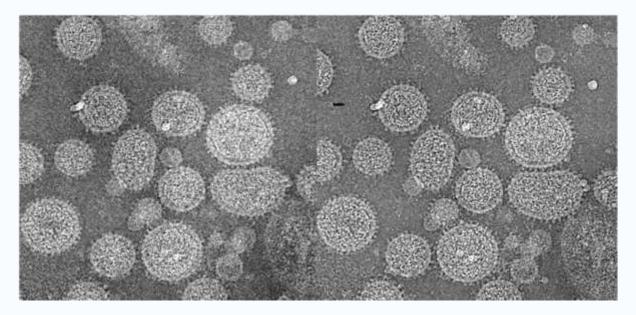


Figure 7. Cryo-EM images of TCRV.

JUNIN VIRUS (JUNV):

Junin is a member of the genus arenavirus, and characteristically causes Argentine hemorrhagic fever (AHF). AHF leads to major alterations within the vascular, neurological and immune systems and has a mortality rate of between 20 and 30% (37)(see fig 8). Symptoms of the disease are conjunctivitis, purpura, petechia and occasional sepsis.

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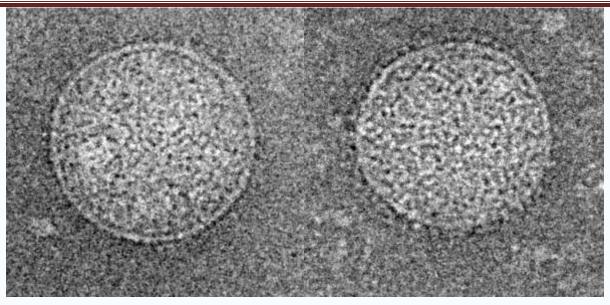


Figure 8. Cryo-EM images of Junin virus strain Candid 1 particles.

Junin virus was discovered in 1958. Its geographical distribution is still confined to Argentina. In 1958 it was distributed to around 15000km but in 2000 the distribution has risen to the area of around 150,000km (37). The natural hosts of Junin virus are rodents, particularly *Mus musculus*, *Calomys spp* and *Akodon azarae (Vesper Mouse)*. Direct rodent to human transmission only transpires when contact is made with excrement of an infected rodent. Hazardous group is 4. This commonly occurs via ingestion of contaminated food or water, inhalation of particles within urine or via direct contact of broken skin with rodent excrement. As described in material method about CTFit an image generated after Contrast transfer function is given in fig 9 showing clear difference of image before and after CTFit.

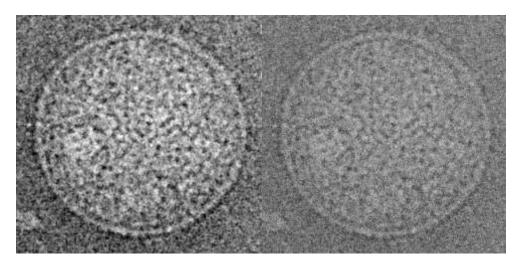


Figure 9. Cryo electron microscopy image of JUNV before (right) and after (left) CTF correction and filtering the images.

Conclusions

Pichinde virus (PICV) belongs to the New World arenaviruses (8) of clade A (see fig 6). Its natural host is *Oryzomys albegularis* (Tomes's Rice Rat) and it was discovered in Columbia in 1967 (36). It is a hazardous group 2. It does not cause any disease in humans but pathogenic in guinea pigs and hamsters.

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