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Recent Advance Technology in Nipah Virus

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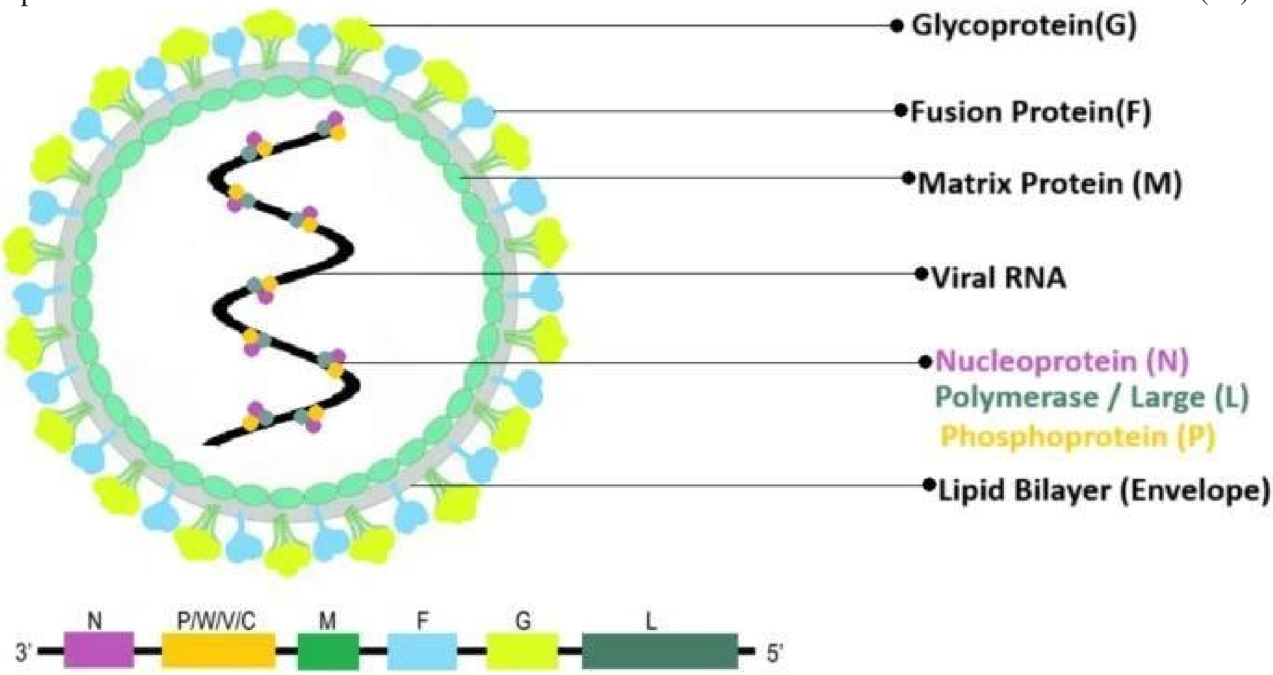
Abstract:

Nipah virus (NiV) is a highly fatal zoonotic pathogen belonging to the family Paramyxoviridae and genus Henipavirus. First identified during outbreaks in Malaysia and Singapore between 1998 and 1999, NiV remains a major public health concern in South and Southeast Asia. The virus has a negative-sense, single-stranded RNA genome encoding six structural proteins (N, P, M, F, G, and L) and exhibits pleomorphic morphology. Fruit bats (Pteropus spp.) are Its natural reservoir, with transmission occurring through bat-human contact, contaminated food (e.g., raw date palm sap), infected animals such as pigs and horses, and limited human-to-human spread. Clinical outcomes range from asymptomatic infection to severe respiratory disease and fatal encephalitis, with case fatality rates varying between 40% and 75%, significantly higher in India and Bangladesh than in Malaysia and Singapore. Epidemiological studies report outbreaks in Malaysia, Bangladesh, India, the Philippines, and Singapore, often linked to occupational exposure, animal contact, and cultural practices. Diagnosis relies on RT-PCR, virus isolation, serology (ELISA, IFA), and immunohistochemistry. Currently, no licensed antiviral therapy or vaccine exists; management focuses on supportive care, infection control, public awareness, and animal surveillance. Research into vaccines—including recombinant viral vectors (VSV, measles virus, canarypox), virus-like particles (VLPs), DNA vaccines, and immunoinformatics-based peptide vaccines—has shown promise in preclinical studies. Global initiatives, such as the Coalition for Epidemic Preparedness Innovations (CEPI), are advancing vaccine development. Given its high mortality, recurrent outbreaks, and pandemic potential, NiV requires strengthened surveillance, early detection, and integrated strategies combining public health measures, research, and vaccination efforts.

Introduction:

Nipah virus (NiV) is a highly fatal and contagious pathogen that causes severe outbreaks with high morbidity and mortality rates. It is a pleomorphic virus endemic to the Western Pacifc and Southeast Asia. It belongs to the family Paramyxoviridae and genus Henipavirus. Te genetic architecture of the virus comprises a single-stranded negative-sense, unsegmented RNA that encodes six genes: nucleocapsid (N), phosphoprotein (P), matrix (M), fusion protein (F), glycoprotein (G), and RNA polymerase (L) .The natural reservoir of the virus are bats of genus Pteropus; therefore, proximity with bats is one of the reasons for the occurrence of NiV and swal-lowing of raw date palm sap is another .Tis scenario has made NiV a condition of growing concern not only for health ofcials but also for researchers, especially because recent outbreaks continue to occur in certain regions of South and Southeast Asia.Te manifestations of Nipah virus infection vary greatly, ranging from subclinical to severe respiratory dis-eases and fatal encephalitis (3) Early detection and prompt response are crucial for controlling outbreaks; however, these efforts are hindered by diagnostic challenges and delayed rec-ognition, as NiV symptoms mimic those of many other febrile illnesses (3). The mortal-ity rate was approximately 40% in Malaysia and Singapore, compared to 70% in India and Bang-ladesh, where the outbreaks of NiV are frequent. Considering its degree of endemic in South Asia, systemic surveillance is required to overcome its recurrence and rising mortality rates (4).Af-ter its discovery from Sungai Nipah (Nipah River Village), where its name originally came from, the first cases characterized by severe febrile enceph-alitis in humans were reported in Singapore and Malaysia, between 1998 and 1999 (5)

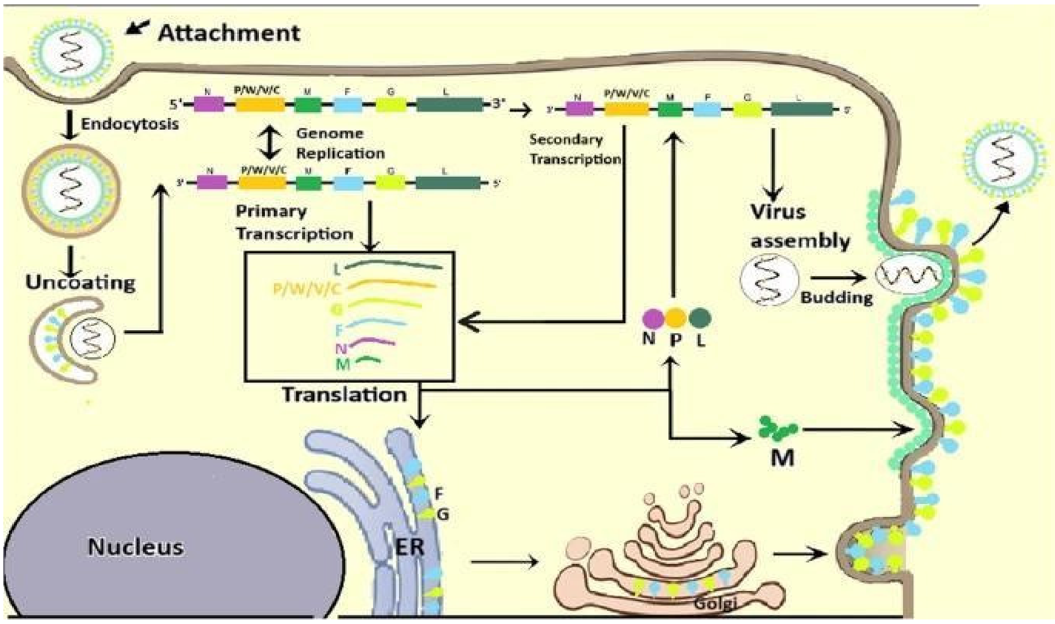
Discovered two decades ago as aetiolog-ic agent of a zoonotic disease, Nipah virus (NiV), is a member of genus Henipavirus in the Paramyxoviridae family. Unlike the Hendra Vi-rus, which is 18234 nucleotides long, NiV is 18246 nucleotides long, which accounts for them to be 15% longer than others in their family. (6)-ver, the genetic characteristics found in the genus Henipavirus, include the unique, 3' leader and 5' trailer sequences, which promote transcription and replication of genomic RNA, respectively (7)
Transmission of NiV has been reported by vari-ous ways which include human-to-human trans-mission and food-borne transmission (8) The virus particles are pleomorphic in shape, altering from spherical to filamentous, and have a Diameter of approximately between 40 and 600 Nm The novel RNA paramyxovirus (genus Henipavirus), closely related to the Hendra virus, was named after the outbreak in the village of Sungai Nipah in the State of Negeri Sembilan (close to the Federal Territory of Kuala Lumpur), Malaysia, in which the virus was first isolated from a human patient in 1998 (9)The
course of the disease can be very serious and lead to death. The disease is also widely described in pigs, with clinical signs involving the respiratory and nervous systems [3]. The natural reservoir of the virus is represented by frugivorous bats called "flying foxes" belonging to the Pteropus genus. The main outbreaks of NiV infection have so far occurred in the geographical area of the carrier bats, i.e., in Malaysia, Singapore, Bangladesh and India, but cases were also described in the Philippines. The emergence of the virus and the zoonotic potential of transmission to animals and humans seem to be related to losses in the bats' habitat (10)



Virus structure

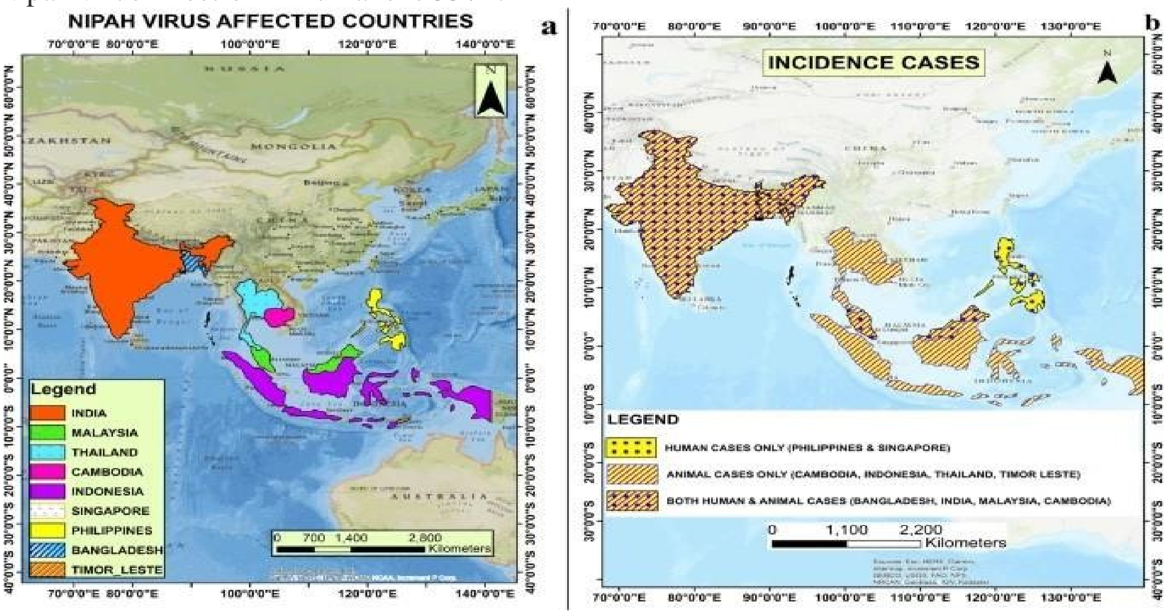
The G protein of the Nipah virus attaches to the host cells in a direct manner to the respective receptors that are found on the host cell membrane (Fig. 2).
The G protein of the Nipah virus specifically attaches to the EFNB2 and EFNB3 receptors .
There are also roles for the F protein in envelope-virion binding, which permits the virus envelope to fuse with the cell membrane. This primary fusion event brings the viral RNA into the cytoplasm of the host cell

Virus transmission:-



Geographic distribution and endemic regions:-

Sun et al. documented 749 human cases of Nipah virus infection across Bangladesh, India, Malaysia, Singapore, and the Philippines (Fig. 3a). Of the patients with demographic data, 89% (358 of 402) were adults aged 15–59, with men making up 74% (391 of 530) and 26% (139 of 530) of the cases, respectively. Livestock practitioners were the most common occupational group, accounting for 68% (238 of 351) of cases. Among the 489 patients with known exposure, 69% (336) had animal contact, 26% (127) had exposure to infected individuals, and 5% (26) had exposure through date palm-related activities. Epidemiological patterns such as sex, exposure, and disease timing varied across the five countries. The crude case fatality rate of Nipah virus infection in humans is 55% .



Epidemiology :-

Malaysia: Human NiV infection was first identified in Malaysia from 1998 to 1999 . The name "Nipah" comes from Sungai Nipah (Nipah River village). Several cases presenting with fever, headache and reduced consciousness were reported from the state of Perak, Malaysia in September 1998. Initially, four cases tested positive for IgM antibodies against Japanese Encephalitis (JE) and a JE outbreak was declared. Despite the implementation of control measures, the outbreak intensified. By the end of the year, more clusters were reported in Port Dickson District, 300 km south. NiV, being less widespread as compared to coronavirus disease 2019 (COVID-19), still poses a significant threat to cause a pandemic due to its high mortality rate, infrequently sprouting outbreaks, and practically no treatment or vaccines as depicted in Table

Table 1. Comparative analysis of Nipah and coronavirus disease 2019.

Features	
Agent	Nipah virus
Transmission	Fruit bats to animals like
Human-to-human transmission	Limited, but
Incubation period	4 to 14 days
Symptoms	Fever, beada syndrome, e
Severity	Variable, ran symptoms
Case fatality rate	40% to 75%
Vaccination	No specific
Treatment	Supportive c available
Preventive measures	Avoiding exp infected ani

Features	
Global impact	Outbreaks, m

COVID-19: Coronavirus disease 2019; SARS-CoV-2: Severe acute respiratory syndrome coronavirus 2.

Bangladesh: The epidemiology of NiV is significantly different in Bangladesh. Since 2001, seasonal outbreaks of NiV have occurred in Bangladesh in the winter months, primarily in 20 districts.in central and north-western Bangladesh (the 'Nipah belt'), where the majority of spillover events occur.Though contact with pigs has been reported from a majority of patients in Bangladesh, close contact with pigs was found to be a risk factor in one outbreak.

India: In India, there was a large outbreak (66 probable cases and 45 deaths) in Siliguri, West Bengal in 2001 and another smaller outbreak (five cases, 100% fatality) in 2007 in Nadia district, West Bengal. These outbreaks were across the border from the Nipah belt in Bangladesh. In May 2018, an outbreak of NiV was declared in Kozhikode and Malappuram districts of Kerala, a southern state in the west coast, which is geographically disconnected from previously affected areas. Date palm sap consumption is not a common practice in this area. There were 18 confirmed cases and 17 deaths as of 1 June 2018 All cases belonged to the economically productive age group, with no sex differential

Philippines: An outbreak of NiV infection occurred in the Philippines in 2014. Seventeen cases were confirmed, the case fatality rate was 82%. Ten patients had a history of close contact with horses or of horse meat consumption. Deaths of 10 horses were reported in the same time period, of which nine showed neurological symptoms. However, samples from horses were not tested for NiV. Five patients, including two healthcare personnel, acquired the disease through person to person transmission. This strain was closely related to the Malaysian strain where definite person to person spread had not been previously identified

Pathogenesis:-The Henipaviruses are the only zoonotic paramyxoviruses. They are also exceptional in their broad host range and high case fatality rates. They have a nonsegmented negative-stranded RNA genome consisting of helical nucleocapsidsc encased in an envelope forming spherical to filamentous, pleomorphic virus particles. Both HeV and NiV have a significantly larger genome than other paramyxoviruses. The genome encodes six structural proteins, the nucleocapsid protein (N), phosphoprotein (P),

matrix protein (M), fusion protein (F), glycoprotein (G) and large protein (L) or RNA polymerase, in the order 3′-N-P-M-F-G-L-5′. There are three predicted non-structural proteins, C, V and W which are all encoded by the P gene

Diagnosis:- The diagnosis of Nipah virus (NiV) infection involves a combination of clinical evaluation, laboratory tests, and epidemiological information. Several diagnostic methods are used to detect NiV, including:

1) Real-time Reverse Transcription Polymerase Chain Reaction (RT-PCR): This technique detects the presence of NiV RNA in patient samples such as blood, respiratory secretions, cerebrospinal fluid, or tissue specimens
2) Virus Isolation: NiV can be isolated and cultured from clinical specimens, such as throat swabs, blood, or cerebrospinal fluid, using cell culture techniques.
3) Serological Tests: Serological assays, including enzyme-linked immunosorbent assay (ELISA) and indirect immunofluorescence assay (IFA), are employed to detect antibodies against NiV in patient serum samples. IgM antibodies indicate recent infection, while IgG antibodies suggest past exposure or immunity.
4) Immunohistochemistry: This technique involves the detection of NiV antigens in tissue specimens, usually obtained from autopsies of fatal cases, using specific antibodies .

Disease Control and Management:- The management and control of Nipah virus involves a combination of public health measures, surveillance, case management, and research efforts. Here are some key aspects of managing and controlling Nipah virus:

Early Detection: Establishing a robust surveillance system is crucial to detecting Nipah virus cases early. This includes active surveillance in healthcare facilities, monitoring of wildlife populations, and reporting of unusual illness patterns. Rapid identification and reporting of suspected cases allow for timely response and control measures.

Infection Prevention and Control: Outbreaks of NiV invoke costly emergency responses around the world. NiV poses a greater threat in regions associated with risk factors and with poor indicators of development With climate change and human encroachment into flying fox habitats, it is likely that outbreaks will occur in new locations Measures to prevent the access of bat to the sap stream of the date palm tree as well as shaved surface can minimize the risk of human exposure to NiV in such settings

Case Management**:** Providing supportive care to Nipah virus patients is an important aspect of management. There is currently no specific antiviral treatment available for Nipah virus, so symptomatic and supportive care, including respiratory and organ support, is the mainstay of treatment.

Public **Awareness and Education**: Raising public awareness about Nipah virus, its transmission, and preventive measures is vital. This includes educating the public about avoiding direct contact with infected bats, refraining from consuming raw date palm sap contaminated by bat saliva or urine, and practicing good hygiene measures.

Animal Surveillance and Control: Nipah virus is believed to originate from bats, with intermediate hosts such as pigs playing a role in transmission to humans. Surveillance and control measures targeting these animal populations, including culling infected or at-risk animals, can help prevent spillover events

Research and Vaccine Development: Continued research into

Nipah virus, including its epidemiology, transmission dynamics, and potential vaccines or therapeutics, is essential for better prevention and control. Development of effective vaccines and antiviral treatments can significantly enhance efforts to manage and control

Nipah virus outbreaks.

Vaccines:-Vaccination of humans is an integral part of preventing infection due to NiV. Prevention also includes vaccination of livestock (especially pigs and probably horses) in endemic areas Of note, outbreaks cannot be prevented amongst the livestock population in areas where contamination of date palm sap acts as major contributor to the spread of NiV infection. However, if vaccination of livestock is made cheap it may prove to be successful in certain regions. Extensive research involving

preclinical studies in a number of animals and nonhuman primates have identified multiple vaccine candidates, including vectored and subunit vaccines, offering protective immunity Among vectored vaccines, one employing vesicular stomatitis virus has shown protection inferrets, African green monkeys, as well as hamstersDespite these developments, funding for human clinical trials of candidate vaccines remains a problem for academic community.The pharmaceutical companies are hesitant to invest in research on development of vaccines for diseases like Nipah, which are rare occurrences, despite the high fatality.

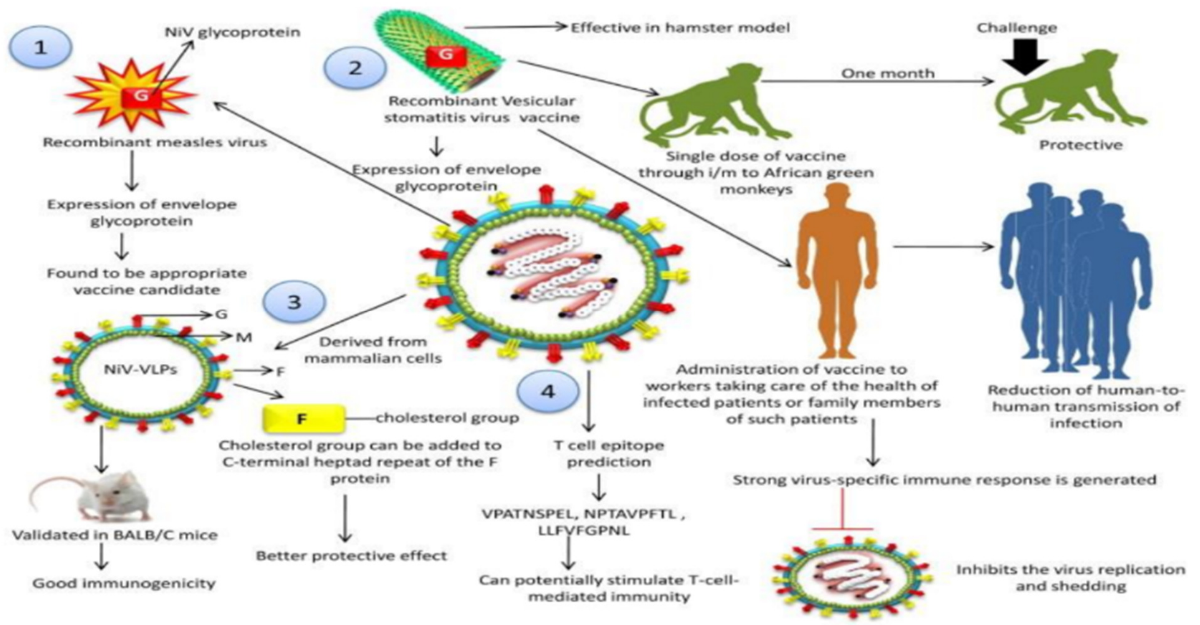
A collaborative effort has been undertaken by both government and pharmaceutical companies, known as the Coalition for Epidemic Preparedness Innovations (CEPI). It was formed in January 2017 for developing safe, efficacious and affordable vaccines against diseases associated with pandemic potential, like Nipah DNA vaccines, virus-like particles, virus vectors(live and recombinant), and other advanced vaccines have been developed as strategies of immunization against both HeV and NiV Experimental vaccines based on the several viral vectors, including the canarypox virus, vesicular stomatitis virus glycoprotein (VSVΔG) and rhabdovirus have been evaluatedA recombinant measles virus (rMV) vaccine that expresses envelope glycoprotein of NiV has been found to be promisingfor use in man.A replication-competent, recombinant VSV-vectored vaccine encoding NiV glycoprotein was reported to show high efficiency in a hamster model. A single intramuscular dose of the vaccine conferred protective immunity in African green monkeys one month after vaccination.Healthcare workers and family contacts attending Nipah cases should be considered for Nipah vaccination, in order to limit human-to-human transmission and curb outbreaks.Nipah virus-like particles (NiVVLPs) composed of three NiV proteins G, F and M derived from mammalian cells have been produced and validated as vaccine in BALB/c mice. The immunogenicity of the NiV-VLP vaccine was high because the VLPs possess the native characteristics of the virus including the size, morphology and surface composition .NiV-VLPs derived from mammalian cells transfected with plasmids containing NiV G, F and M genes have also been produced yielding VLPs with the three proteins. These VLPs are composed of G, M and F proteins of the virus. Golden Syrian hamsters immunized with these VLPs developed high titres of neutralizing antibody in serum, and showed complete protection upon viral challenge.Immunoinformatic advances have been utilized for developing peptide-based NiV vaccine by prediction and modeling of T-cell epitopes of NiV antigenic proteins. Specific epitopes, viz., VPATNSPEL, NPTAVPFTL and LLFVFGPNL of N, V and F proteins, respectively, showed substantial binding energy as well as score with HLA-B7, HLA-B*2705 and HLA-A2 MHC class-I alleles, respectively Such predicted peptides can potentially stimulate T-cell-mediated immunity and could have utility in developing epitopebased vaccines to counter NiV. In silico epitope prediction tools which evaluated G and F protein of NiV indicated that either GPKVSLIDTSSTITI or EWISIVPNFILVRNT peptides could formulate an effective universal vaccine component, inducing both humoral and cell-mediated immunity (Sakib et al. 2014). A more recent in silico analysis using bioinformatics tools indicated that the epitopes from G (VDPLRVQWRNNSVIS) and M (GKLEFRRNNAIAFKG) proteins can be helpful for designing common B- and T-cell epitope-based peptide vaccinesagainst HeV and NiV, and this approach needs to be evaluatedFrom another epitope-based immunoinformatics and prediction study on the NiV associated RNA-dependent RNA polymerase protein complex, best-predicted T-cell epitopes identified are ‘ELRSELIGY’ (peptide of phosphoprotein) and ‘YPLLWSFAM’ (nucleocapsid protein). Such approach identified B-cell epitope sequences in phosphoprotein (421 to 471), polymerase enzyme gene (606 to 640) and nucleocapsid protein (496 to 517). These studies are oriented for the validation of potential vaccine candidate protein portions from Nipah virus which could then spearhead towards the development of fruitful subunit vaccines The development of animal models of NiV disease is another priority, in order to evaluate the preventive and therapeutic approaches. This will help in employing successful immunization strategies (both active as well as passive) by targeting the envelope glycoprotein of the virusAn overview on different vaccine strategies available for Nipah virus (NiV) is presented in Table 1 and few important vaccine platforms are depicted in Figure 5.

Table 1.
Different vaccine strategies available for Nipah virus (NiV).

Different vaccine strategies available for Nipah virus (NiV).

Vector	Antigen used	Dose for immunization	Animal model	Route of vaccination	Administration frequency	Challenge wi titer
Vesicular stomatitis virus (VSV)	rVSV expressing NiV G	10^5 plaque forming units (PFU)	Hamsters	Intraperitoneal	Single	6.8 × 10^4 TCID LD_{50})
	rVSV-ZEBOV-GP-NiVG	10^7 PFU	African Green monkey	intramuscular	Single	10^5 $TCID_{50}$ (Ma strain)
	rVSV-ΔG-NiVB/G	10^7 PFU	Ferrets	Intramuscular	Single	~5 × 10^5 PFU
	Replication-defective VSV	10^6 infectious particles	Female Syrian golden hamsters	Intramuscular	Single	10^5 $TCID_{50}$ per hamster (>10(times LD_{50}- Malaysian stra
Canarypox virus (ALVAC) vaccine vector	vCP2199, carrying the NiV-G and vCP2208, carrying the NiV F	10^8 PFU	Landrace female pigs	Intramuscular	Boosted 14 days postvaccination	2.5 × 10^5 PFU
Adeno-associated virus (AAV)	NiV G	2.1010/1.1010 genome particles	Balb/c male mice	Intra muscular or intra-dermal	One Booster	10^4 PFU
		6.1011 genome particles	Golden hamsters	Intra muscular		
Vaccinia virus	NiV G and NiV F	10^7 PFU	BALB/c female mice	Subcutaneously	Boosted with the same dose	1,000 PFU
Measles virus based-vectors (HL strain or Edmonston B strain)	NiV G	1 × 10^5 TCID50	African green monkeys	Subcutaneously	One booster	1 × 10^6 TCID5(
		2 × 10^4 TCID50	Hamsters	Intraperitoneal		1 × 10^8 TCID5(
Venezuelan equine encephalitis virus	NiV G	3.1 × 10^5 IU	C3H/He mice	Foot pad inoculation	Single	Serum neutra assay with pN 3.luc.E-R-repo gene encodin

Vector	Antigen used	Dose for immunization	Animal model	Route of vaccination	Administration frequency	Challenge wit titer
replicon particles						retroviruses ps typed with NiV F + NiV G glycoproteins
Newcastle disease virus (NDV), LaSota strains	NiVG and NiVF	108 EID50 2 × 109 EID50	Mice Pig	Intramuscular Intramuscular	Single	Serum neutral assay with VSVDGnGFP-N pseudo- typed
Nipah virus-like particles (NiV-VLPs)	NiV G, F and M adjuvanted with Alum (50 μg - Alhydrogel 2%); MPLA (15 μg) and CpG ODN (40 μg)	30 μg VLP	Golden Syrian hamsters	Intramuscular	Three-dose vaccination schedule	~16,000 PFU



Conclusion:- The emergence of the Nipah virus presents a formidable challenge to public health, characterized by severe clinical manifestations and posing a significant threat through both outbreaks and sporadic cases. Timely detection, surveillance, and heightened awareness among healthcare providers and the general population are critical for early identification and effective management. Continued research efforts are imperative to unravel the complexities of the virus, develop diagnostic tools, and explore potential treatments. International collaboration and information sharing are indispensable for a unified global response, while strengthening public health infrastructure and implementing proactive measures, including vaccination strategies, are crucial elements of a comprehensive approach to mitigate risks. As we navigate the complexities of emerging infectious diseases, a sustained commitment to preparedness, research, and collaborative action remains paramount for safeguarding global health and averting future crises. The 'One Health' approach is

also the utmost importance. There is requirement of coordination between institutes as well as at the international level among virologists from both medical and veterinary fields as well as ecologists for understanding to the fullest the period and mechanism involved in excretion of the virus by the bats. At the same time, the common people should be educated about food hygiene as well as hygiene at personal level. Inspection of all the imported livestock at the time of arrival and also before travel at the point of origin is essential. Proper isolation, quarantine and disinfection protocol including infrastructure facilities and trained personnel with protective clothing should be in place to respond quickly upon identification of any new case. There should be maintenance of proper hygiene at maximum level for slaughtering such livestock. For preventing future NiV outbreaks, a continuous surveillance in the area of human health, animal health, and reservoir hosts should be carried out to determine the prevalence and to predict risk of virus transmission in human and swine populations. Successful accelerated development of preventive vaccines and therapeutic antibodies or antivirals are need of the hour to control the spread and treat the infected patients during an outbreak. Collaborative efforts such as
CEPI and biotech companies will accelerate the vaccine or therapeutic development for NiV.

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