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Current classification of *Peribunyaviridae* family: genetic diversity and contributing factors

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Abstract: The first detection of Bunyamweravirus in 1943 and many other antigenically related viruses identified in the following years led to the formal establishment of the *Bunyaviridae* family in 1975. This family became the *Bunyavirales* order in 2017 following proposals submitted to International Committee on Taxonomy of Viruses when the establishment of the *Peribunyaviridae* family was also approved for the *Orthobunyavirus* and *Herbertvirus* genera. The *Peribunyaviridae* family includes the *Orthobunyavirus, Herbertvirus, Pacuvirus, and Shangavirus* genera. Many types of viruses within this family can infect humans, mammals, plants, and insects. However, many of these viruses are transmitted by arthropod vectors without requiring mammals for their viability, which propose that Peribunyaviruses might have initially evolved as viruses that only infect insects. The leading factors contributing to genetic diversity in the *Peribunyaviridae* family are mutations and genetic reassortment. Mutations are generally detected in the M segment, which encodes the surface glycoproteins that enable viruses to avoid the immune response.
Keywords: Classification, genetic reassortment, mutation, *Peribunyaviridae*.

Peribunyaviridae ailesinin güncel olarak sınıflandırılması: genetik çeşitlilik ve etki eden faktörler

Özet: İlk olarak 1943 yılında Bunyamweravirus tespit edilmesi ve sonraki yıllarında pek çok serolojik olarak ilişkili virusun bulunması ile birlikte, 1975 yılında kurulan *Bunyaviridae* ailesi 2017 yılında *Uluslararası Virus Taksonomi Komitesi (International Committee on Taxonomy of Viruses)*'ne sunulan öneriler neticesinde *Bunyavirales* takımına yükseltilmiştir. Yine bu tarihten itibaren *Orthobunyavirus* ve *Herbertvirus* genusları için *Peribunyaviridae* ailesinin kurulması onaylanmıştır. *Peribunyaviridae* ailesi içinde *Orthobunyavirus, Herbertvirus, Pacuvirus ve Shangavirus* genusları bulunmaktadır. Bu aile içinde insan, memeli, bitki ve insektleri enfekte etme özelliğine sahip pek çok virus türü mevcuttur. Ailedeki pek çok virusun arthropod vektörlerle aktarılması ve virusların devamlılığı için memelilere ihtiyaç duymaması sonucu, bu virusların ilk başta sadece artropodları enfekte eden viruslardan evrimleştiği düşünülmektedir. *Peribunyaviridae* ailesindeki genetik çeşitliliğe katkıda bulunan faktörlerin başında mutasyonlar ve genetik reassortment gelmektedir. Bazı araştırıcılar tarafından bu ailedeki çoğu virusun genetik reassortment yoluyla oluştuğu düşünülmektedir. Mutasyonlara ise en çok yüzey glikoproteinleri kodlayan M segmentinde rastlanılmakta ve bu sayede viruslar immun yanıttan kaçabilmektedir.
Anahtar kelimeler: Sınıflandırma, genetik reassortment, mutasyon, *Peribunyaviridae*.

Introduction

The history of the classification of viruses is not as old as virus discovery. While Beijerinck was the first to detect a virus in 1898, viruses were not classified by virologists until the 1920s. The first system was generally based on pathological features. However the plant viruses were classified according to host reaction and differential host species using a binomial-trinomial nomenclature based on the name of the infected plant (Holmes, 1939). Detailed virus classification began only after the invention of the electron microscope in the 1950s. Hundreds of new viruses were then quickly discovered using this technology. Virologists at the International Microbiology Congress, held in Moscow in 1966, established the International Committee on Nomenclature of Viruses (ICNV) to develop a globally recognized nomenclature and taxonomy system for all viruses that detected so far. In 1974, this was later renamed the International Committee on Taxonomy of Viruses (ICTV) and accepted as an official organization in matters related to the nomenclature and taxonomy of viruses (Fauquet, 1999).

Viruses in the order *Bunyavirales* were named after Bunyamweravirus (BUNV), isolated from Aedes species in Uganda's Semliki forests during yellow fever research in 1943. Within 25 years, many other antigenically related viruses had been detected in laboratories in India, South America, and Africa (Rosenberg et al., 2013), which were first classified as Bunyamwera supergroup viruses. However, following advanced biochemical and structural analyzes, they were regrouped as the *Bunyaviridae* family in 1975 (Vaheri et al., 2013).

After further studies, the ICTV approved the promotion of the *Bunyaviridae* family to the *Bunyavirales* order. *Bunyaviridae* included five established genera of trisegmented negative-strand RNA viruses (*Hantavirus, Nairovirus, Orthobunyavirus, Phlebovirus, and Tospovirus*). Almost half of the currently known bunyaviruses have not been or cannot be assigned to these five genus based on established classification criteria. Besides, novel viruses have recently been discovered that cluster with classical trisegmented bunyaviruses in phylogenetic analyses of all their proteins, yet are bisegmented (e.g., Wǔhàn millipede virus 2, South Bay virus) (Li et al., 2015; Tokarz et al., 2014). Lastly, in a large number of plant viruses with more than three genomic segments, currently members of the unassigned genera Emaravirus and Tenuivirus, have long been referred to as clearly "bunyavirus-like" based on the clustering of encoded proteins with bunyavirus proteins (Elebaino et al., 2009; van Poelwijk et al., 1997).

The ICTV Bunyaviridae study group assured to take initial steps to clarify this taxonomic confusion in 2016. A thorough reconsideration/review of the "bunyavirus-like supergroup" was agreed upon a series of Taxonomic proposals: first, to classify recently

unassigned viruses to existing genera; second, to establish novel genera to assign typical bunyaviruses that cannot be assigned to the existing five genera (Junglen, 2016); third, to accept Tenuiviruses and Emaraviruses as official members of the bunyavirus supergroup; terminally, to reorganize the family taxonomically to adequately reflect the relationships of the various now included and classified bunyaviruses while at the same time establishing taxonomic "room" for further revisions in 2017.

This work demonstrated that the existing *Bunyaviridae* family should be upgraded to the *Bunyavirales* in order to reflect the evolutionary relationships of various bunyaviruses within a broader taxonomic framework in a better way. The phylogenetic analysis of the S, M, and L (small, medium, and large) segments of Bunyaviruses indicated that the genera within the *Bunyaviridae* should be reclassified as families. The creation of the *Bunyavirales* order also required a family for the remaining two genera (*Herbevirus and Orthobunyavirus*). This family was named *Peribunyaviridae* (ICTV, 2017).

When classifying viruses within the *Bunyavirales* order, S, M, and L segments open reading frame (ORF) full-length products (respectively nucleocapsid protein, surface glycoproteins, and RNA-dependent RNA polymerase [RdRp]) are analyzed separately with multiple sequence alignment (MAFFT) and classified according to their similarities. As a result, the Pacuvirus genus was added to the *Peribunyaviridae* family (Piet et al., 2018).

***Classification of the Peribunyaviridae family*:** According to the latest ICTV taxonomy data, there are 12 families in the *Bunyavirales* order, with four genera (*Orthobunyavirus, Herbevirus, Pacuvirus, and Shangavirus*) and 97 species in the *Peribunyaviridae* family (Table 1). Most of the Peribunyaviruses are transmitted by arthropod vectors, such as midges, mosquitoes, ticks and sandflies (Hughes et al., 2020). Most viruses in this group infect mammals, while the others infect only arthropods. Infection usually occurs during feeding by a blood-sucking arthropod. Arthropods can be persistently infected.

The infections that viruses in this family cause vary by virus type (Hughes et al., 2020). The most important species in the *Peribunyaviridae* family for veterinary medicine belong to the *Orthobunyavirus* genus. Many of them are transmitted by vectors that can cross the placental barrier in economically valuable animals and cause clinical symptoms, such as abortion, congenital anomalies, and stillbirths (Table 2).

Table 1. Comparison of former and current *Bunyaviridae* family classification (ICTV, 2020).

Former classification		**Recent classification**	
Family:	*Order:*	*Family:*	
Bunyaviridae	*Bunyavirales*	*Arenaviridae*	
Genus:		*Cruliviridae*	
Hantavirus		*Fimoviridae*	*Genus*
Nairovirus		*Hantaviridae*	
Orthobunyavirus		*Leishbuviridae*	
Phlebovirus		*Mypoviridae*	*Herbevirus*
Tospovirus		*Nairoviridae*	*Shangavirus*
		Peribunyaviridae	*Orthobunyavirus*
		Phasmaviridae	*Pacuvirus*
		Phenuiviridae	
		Tospoviridae	
		Wupedeviridae	

Table 2. *Peribunyaviridae* family members and the diseases that they cause (Amroun et al., 2017).

Family	**Genus**	**Species**	**Diseases**	**Vector**	**Host**	**Geographical distribution**
Peribunyaviridae	*Orthobunyavirus*	Schmallenberg orthobunyavirus	Abortion, foetal malformation, stillbirth	*Culicoides spp.*	Cattle, sheep, goat	Europe
	Orthobunyavirus	Akabane orthobunyavirus	Abortion, Congenital abnormalities	*Culicoides spp.*	Cattle, sheep, goat	Africa, Asia, Australia
	Orthobunyavirus	Bunyamwera orthobunyavirus	Fever, headache, rash, rarely CNS diseases	Mosquito	Primary human	Africa, Asia, Australia
	Orthobunyavirus	Aino orthobunyavirus	Abortion, foetal malformation, stillbirth	*Culicoides spp.*	Cattle, Sheep, goat, horse Just ab +	Australia, Japan, Ethiopia, Israel
	Orthobunyavirus	Peaton Orthobunyavirus	Abortion, foetal malformation, stillbirth	*Culicoides spp.*	Cattle, sheep, goat, horse pig, cat, dog	Japan, Australia, Israel

Infection begins when the virus enters the body via the bite of infected arthropod. The virus first targets striated muscles, causing a high level of viremia due to replication. Then, the virus spreads to organs and can cross the blood-brain barrier to reach its main target – neurons (Taylor et al., 2014). Central nervous system (CNS) infection is age-dependent, with younger animals being more susceptible than adults. For example, malformed lambs and calves born from animals infected with the Schmallenberg virus (SBV) have high levels of viral antigens in their brain tissue. Analogous findings have been reported for other peribunyaviruses with teratogenic effects (Varela et al., 2013). The genus with the most species is *Orthobunyavirus*, with 88, including important mammal pathogens (ICTV, 2020), some of which are listed in Table 3.

Table 3. The classification of the *Peribunyaviridae* (ICTV, 2020).

Genus	Species	Viruses
Herbevirus	Herbert herbevirus	Herbert virus (HEBV)
Herbevirus	Kibale herbevirus	Kibale virus (KIBV)
Herbevirus	Tai herbevirus	Taï virus (TAIV)
Pacuvirus	Pacui pacuvirus	Pacui virus (PACV)
Pacuvirus	Rio Preto da Eva pacuvirus	Rio Preto da Eva virus (RPEV)
Pacuvirus	Tapirape pacuvirus	Tapirapé virus (TAPV)
Shangavirus	Insect shangavirus	Shuāngào insect virus 1 (SgIV-1)
Orthobunyavirus	Acara orthobunyavirus	Acará virus (ACAV) Moriche virus (MORV)
Orthobunyavirus	Aino orthobunyavirus	Aino virus (AINOV)
Orthobunyavirus	Akabane orthobunyavirus	Akabane virus (AKAV) Tinaroo virus (TINV) Yaba-7 virus (Y7V)
Orthobunyavirus	Anopheles A orthobunyavirus	Anopheles A virus (ANAV) Arumateua virus (ARTV = ARMTV) Caraipé virus (CPEV = CRPV) Las Maloyas virus (LMV) Lukuni virus (LUKV) Trombetas virus (TRMV) Tucuruí virus (TUCV = TUCRV)
Orthobunyavirus	Anopheles B orthobunyavirus	Anopheles B virus (ANBV) Boracéia virus (BORV)
Orthobunyavirus	Bunyamwera orthobunyavirus	Bunyamwera virus (BUNV) Germiston virus (GERV) Lokern virus (LOKV) Mboké virus (MBOV) Ngari virus (NRIV) Northway virus (NORV) Santa Rosa virus (SARV) Shokwe virus (SHOV) Stanfeld virus (STAV) Xingu virus (XINV)
Orthobunyavirus	Cache Valley orthobunyavirus	Cache Valley virus (CVV) Cholul virus (CHLV) Tlacotalpan virus (TLAV)
Orthobunyavirus	California encephalitis orthobunyavirus	California encephalitis virus (CEV) Morro Bay virus (MBV)
Orthobunyavirus	Gamboa orthobunyavirus	Brus Laguna virus (BLAV) Calchaquí virus (CQIV) Gamboa virus (GAMV) Pueblo Viejo virus (PVV) Soberanía virus (SOBV)
Orthobunyavirus	Guama orthobunyavirus	Ananindeua virus (ANUV) Guamá virus (GMAV) Mahogany Hammock virus (MHV) Moju virus (MOJUV)
Orthobunyavirus	Kaeng Khoi orthobunyavirus	Kaeng Khoi virus (KKV)
Orthobunyavirus	La Crosse orthobunyavirus	La Crosse virus (LACV)
Orthobunyavirus	Leanyer orthobunyavirus	Leanyer virus (LEAV)

Table 3 continued. The classification of the *Peribunyaviridae* (ICTV, 2020).

Orthobunyavirus	Oropouche orthobunyavirus	Iquitos virus (IQTV)d Madre de Dios virus (MDDV) Oropouche virus (OROV) Perdões virus (PDEV) Pintupo virus (PINTV)
Orthobunyavirus	Peaton orthobunyavirus	Peaton virus (PEAV)
Orthobunyavirus	Schmallenberg orthobunyavirus	Douglas virus (DOUV) Sathuperi virus (SATV) Schmallenberg virus (SBV) Shamonda virus (SHAV)
Orthobunyavirus	Simbu orthobunyavirus	Para virus (PARAV) Simbu virus (SIMV)

Orthobunyaviruses are divided into more than 18 serogroups based upon the presence/absence or degree of serological cross-reactions using various analyses, such as hemagglutination inhibition (HI), complement fixation (CF) and neutralization assays (Calisher et al., 1983). These serogroups are Anopheles A, Anopheles B, Bakau, Gamboa, Guamá, Capim, Mapputta, Tete, Koongol, Turlock, Group C, Koongol, Nyando, Bwamba, California, Bunyamwera, Minatitlan, Simbu, Olifanstlei, Patois, Wyeomyia (Wikipedia, 2020). Within the Orthobunyavirus, the most important species for veterinary medicine are in the Simbu serogroup, particularly Akabane (AKAV), Aino (AINOV), Douglas (DOUV), Peaton (PEAV), Sabo (SABOV), Sango (SANV), Sathuperi (SATV), Schmallenberg (SBV), Shamonda (SHAV), Shuni (SHUV), and Simbuvirus (SIMV). Although some of these viruses, such as AKAV, SBV, AINOV, and PEAV, progress asymptomatically in ripe sheep, cattle, and goats, they cause various CNS symptoms in fetuses infected in the second trimester of pregnancy, including arthrogryposis, torticollis, hydranencephaly, and scoliosis. Although these viruses can cause abortion throughout pregnancy, they most commonly occur in the first trimester. As the immune system develops during the last trimester of pregnancy, offspring infected at this point are usually born healthy (Uchida et al., 2000).

Genomic features: Members of the *Peribunyaviridae* family contain an enveloped, negative-sense, and segmented RNA genome. These segments are Small (S), Medium (M) and Large (L), each of which encodes a different protein (Elliott, 2014). The most variable segment is M, which encodes the surface glycoproteins, whereas the most stable segment is S (Wernike et al., 2015).

Peribunyaviruses encode four structural proteins. The surface glycoproteins (Gn and Gc), N (nucleocapsid) and L (viral RdRp) integral proteins. Electron microscopic images proves that purified virions have from pleomorphic to spherical particles nearly 90 nm in diameter, a

pair of membrane envelopes, and pointed spikes predicted to be glycoproteins. Virions have an average diameter of 108 ± 8 nm and are pleomorphic. The glycoprotein spikes are about 18 nm long, projecting from their membranes. The spikes consist of trimers of Gn-Gc proteins with a tripod-like formation on the viral surface (Obijeski et al., 1976). Figure 1 shows the Virion structure of Peribunyavirus (Bowden et al., 2013).

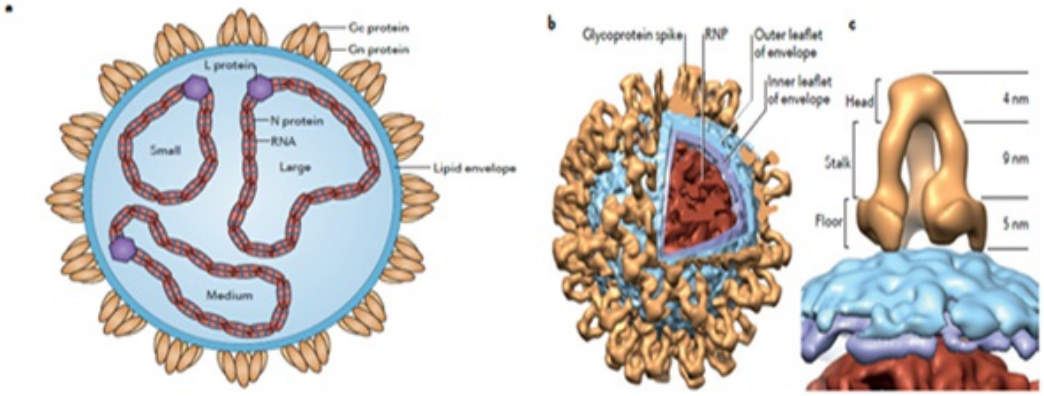

Figure 1. Virion structure of Peribunyavirus. a: Schematic diagram of orthobunyavirus virion. Segments of orthobunyaviruses (S, M, L) are encapsulated via the N protein to form RdRp-associated Ribonucleoprotein (RNP) complexes. RNPs are packaged in a lipid envelope originating from the host cell Golgi complex modified by the addition of viral glycoprotein Gn and Gc. b: Cross-sectional model of Bunyamwera virus (BUNV) virion. c: Structure of glycoproteins on the envelope surface of peribunyaviruses. This structure consists of two interrelated proteins: the floor region adjacent to the envelope and the head region distal to the envelope. These two protein contacts are separated from each other by a stalk region (Bowden et al., 2013).

Genome organization: The terminal nucleotides at the 3′ and 5′ ends of each segment are species-specific. They form a panhandle structure that functions as a promoter for both the replication and transcription of each segment (Barr et al., 2003). In spite of the length of each coding region and the size of the encoded proteins are conserved between different Peribunyaviruses, the length and sequence of UTRs are different. Also, UTRs are required for the genomic RNA encapsulation by the N protein, termination of mRNA transcription, and packaging of RNPs into virions. (Kohl et al., 2006). The integral sequences deletion in UTRs of BUNV causes mistakes in virus attenuation and replication and eliminates its cytopathic effect (CPE) in mammalian cells. However, the mechanism has not been elucidated yet, so further studies are required (Mazel-Sanchez & Elliot., 2012).

To translate genomic RNA segments (with negative polarity), they must first transcribe into positive polarity mRNA, which occurs instantly after infection. The S segment encodes the N protein, while the M segment encodes surface glycoproteins Gc and Gn, and the L segment encodes RdRp. (Elliott & Blakqori, 2011). Glycoproteins are encoded as a precursor polyprotein, including the non-structural protein NSm. The coding sequence of the M segment

mRNA is Gn-NSm-Gc, which is simultaneously translated by host proteases to produce these three proteins (Fazakerley et al., 1988). The S segment of most orthobunviruses encodes a second non-structural protein, NSs, translated from the same mRNA within the N protein-coding sequence while using an alternative AUG initiation codon (Fuller et al., 1983). Some Peribunyaviruses produce small amounts of NSs, while others even lack the gene region responsible for the production of this protein (Mohamed et al., 2009).

Structural and non-structural Peribunyavirus proteins:

L protein: The L protein is the RdRp responsible for catalyzing both transcription and replication. The L protein amino-terminal domains contain an endonuclease domain that separates capped oligonucleotides from the 5′ ends of the host mRNAs and is then used to initiate viral mRNA synthesis. That feature, called as 'cap snatching,' has been defined in the transcription of influenza viruses. In Peribunyaviruses, however, it occurs in the cytoplasm (Patterson et al., 1984; Reguera et al., 2010). L protein is the primary classification protein in the *Bunyavirales* order.

Gn-Gc proteins: The two proteins are type I integral membrane proteins and modified by N-linked glycosylation. Gn and Gc form heterodimers in the endoplasmic reticulum. Their carriage to the Golgi complex leans on the target signal, found in the transmembrane domain of Gn in Peribunyavirus. When only Gc is expressed, it remains in the endoplasmic reticulum, that proves it needs Gn to bud from the Golgi apparatus. (Shi et al., 2004). Gc appears to have a class II fusion area. In BUNV, residue mutations around the fusion peptide in Gc dramatically reduce membrane fusion, whereas deletion of the Gc ectodomain N-terminal domain has only minimally affected virus replication (Shi et al., 2009).

The Gc surface glycoprotein N-terminal variable region is extremely immunological and the main target region of neutralizing antibodies (Hellert et al., 2019). Wernike et al. (2021) developed a triplex ELISA based on the Gc proteins of SBV, AKAV, and SHUV. Compared to the neutralization test, which is the gold standard for diagnosing Simbu serogroup viruses, the respective specificities were 84.56%, 94.68%, and 89.39%, while the respective sensitivities were 89.08%, 69.44%, and 84.91%. Although these proteins are diagnostically important, their reliability is questionable since they have many variable regions.

Recently, inactive-live attenuated vaccines have been applied in endemic areas for SBV control. Since these vaccines do not differentiate vaccinated from infected animals (DIVA), Endalew et al. (2019) developed a subunit vaccine containing SBV surface glycoproteins. However, these vaccines were ineffective after challenging infection and unable to prevent viremia or disease.

***N protein**:* N protein is a highly immunogenic protein that is the main protein produced by the infected cell. N protein encapsulates genomic-antigenomic RNA. The region that generates the signal for encapsulation is located at the 5'end. N protein also interacts with Gc, Gn, and RdRp (Shi et al., 2006).

Since an antibody response occurs in every infection against the N protein, and the S segment encoding this protein is the most conserved region in the viral genome, ELISA kits, which have been used in diagnosis and surveillance programs since 2013, have been developed based on this protein (Bréard et al., 2013).

Non-structural proteins: Peribunyaviruses often encode one or two non-structural proteins. The BUNV NSm protein is localized in the Golgi apparatus independently of other viral proteins and interacts with the C terminus of the Gc protein (Nakitare & Elliott, 1993). This suggests that NSm may have functions like budding and virus release related to the localization of virion particles close to the maturation zone (Lappin et al., 1994). Also, electron microscopy has demonstrated that infected cells contain new tubular structures containing cellular proteins and NSm protein. Although the NSm protein is not necessary for virus viability, the viruses with deleted NSm protein grow more slowly and have lower titers than field strains (Shi et al., 2006).

Most Peribunyaviruses encode another 10kDa non-structural protein (NSs). Unlike the N protein, the amino acid sequence of NSs are disposed to vary more among distinct Peribunyaviruses and is localized to both cytoplasm and nucleus (Thomas et al., 2004). Although the NSs protein is not essential for viral replication, this protein contributes to viral pathogenesis by playing a role in vector/host immune system interactions. The NSs protein has also been associated with apoptosis (Eifan et al., 2013). They also inhibit cellular translation in mammalian cell cultures as deletion mutants with this protein removed cannot stop cellular translation. However, NSs do not effect on protein synthesis in mosquito cell cultures (Elliott et al., 2013).

Genetic reassortment: Similar to all negative-sense RNA viruses, Peribunyavirus RdRp lacks a proofreading function, leading to significant genetic heterogeneity in virus populations. Moreover, the reassortment of the genome segments observed during the co-infection of viruses belonging to the same family increases genetic diversity since they have a segmented genome. During co-infection with two different Peribunyaviruses, new reassortant viruses can occur with six distinct possibilities to those identified with parental viruses. However, there are some conditions for genomic reassortment. In particular, reassortment can only occur between closely related viruses (within the same serogroup). For Bunyamwera (BUNV) serogroup viruses, the

S and L segments appear to be genetically linked, hence commonly transmitted in pairs, whereas no such link has been observed between viruses in the California serogroup (Iroegbu & Pringle, 1981; Urquidi & Bishop, 1992).

Genetic reassortment is more frequent in arthropod vectors because, by sucking blood from different vertebrate hosts, such vectors make co-infection with two distinct Peribunyavirus species far more likely. The phenotypes of reassortant viruses vary (Beaty et al., 1983) because reassortment generally occurs in the M segment, which encodes surface glycoproteins (Figure 2). The vectors that transfer these viruses also vary. For example, if a new vector is different biologically (e.g., blood-sucking from distinct animal species), new viruses may be introduced to other hosts (Beaty et al., 1981). An instance of a phenotypic mutation is the Ngari virus. This newly formed virus is a reassortant virus with the S and L segments of BUNV but the Batai virus (BATV) M segment. BUNV and BATV only infect humans and cause febrile illness, whereas the Ngari virus is related with severe hemorrhagic disease (Briese et al., 2006).

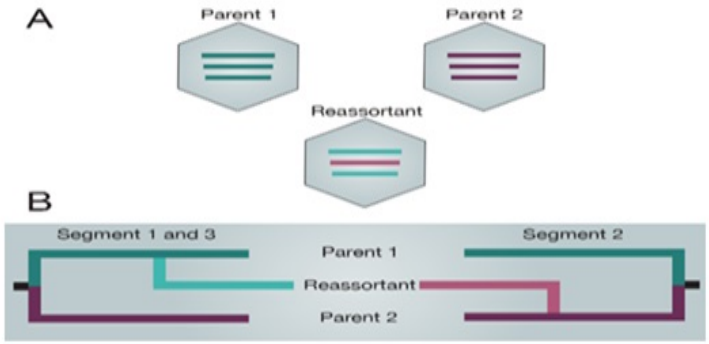

Figure 2. Peribunyaviruses genome reassortment (Vijaykrishna et al., 2015).

Table 4 shows from which virus the genome segments of various Peribunyaviruses originated from and cited the relevant studies. Most peribunyaviruses are thought to result from genomic reassortment. For example, the M segment of the SBV is thought to originate from the Sathuperi virus and Douglas virus meanwhile, the S and L segments are taken from the Shamonda virus (Yanase et al., 2012).

Table 4. Possible genome segment organization of the heterotypic reassortant *Bunyavirales* order (Briese, 2013).

Family / Serogroup	Virus	S segment	M segment	L segment	Reference
Grup C	İtaqui	S_{CARV}	M_{ORIV}	L_{NA}	Nunes et al. (2005)
Simbu	Iquitos	S_{OROV}	M_{unique}	L_{OROV}	Aguilar et al. (2011)
Simbu	Aino B7974	$S_{PEAV\ CSIRO110}$	M_{AINOV}	$L_{PEAVCSIRO110}$	Yanase et al. (2010)
Simbu	Shamonda	$S_{SATV/DOUV}$	M_{Y7V}	$L_{(SATV/DOUV)}$	Goller et al. (2012)
Simbu	Schmallenberg	$S_{\{SHAV(SATV)}$	$M_{(SATV/DOUV)}$	$L_{\{SHAV/SATV\}}$	Yanase et al. (2012)
Bunyamwera	Ngari	S_{BUNV}	M_{BATV}	L_{BUNV}	Gerrard et al.(2004)
Bunyamwera	Macaua	S_{unique}	$M_{(TAIAV/WYOV)}$	L_{unique}	Chowdhary et al.(2012)
Bunyamwera	Tucunduba	S_{TAIAV}	M_{WYOV}	L_{TAIAV}	Chowdhary et al. (2012)
Bunyamwera	Cholul	S_{CVV}	$M_{POTV/KRIV}$	$L_{\{POTV/CVV\}}$	Blitvich et al. (2012)
Phlebovirus	Granada	S_{MASV}	M_{unique}	L_{MASV}	Collao et al. (2010)

Goller et al. (2012) compared the nucleotide sequences of SBV segments and the amino acid similarities of the proteins encoded by these segments with those in other Peribunyaviruses. The N gene nucleotide sequence was furthest from Oropouche virus (OROV) (67.8% and 67.9% aa) and closest to SHAV (97.7% and 100% aa). The L gene nucleotide sequence was furthest from OROV (60.4% and 57.4% aa) and closest to SHAV (92.9% and 98.4% aa). The M gene nucleotide sequence was closest to SATV (82.1% and 90.1% aa) and furthest from SHAV (48.2% and 36.5% aa). In general, the nucleotide sequence of the M gene region of SHAV is not similar to other Simbu serogroup viruses. For example, it is 45.6% similar to OROV on a nucleotide basis and 33.4% as aa, 55% similar to Sangovirus on a nucleotide basis and 47.9% as aa. These results prove that SHAV inherited its M segment from another virus and indicating that SATV and SBV can be classified under the same species since they are closely related.

The life cycle of the peribunyaviruses in vectors: Most Peribunyaviruses are transmitted by mosquitoes and Culicoides as vectors, while Kaeng Khoi Orthobunyavirus is transmitted by bed bugs. Generally, peribunyaviruses are transmitted by one or very few arthropod vectors, even in regions with a wide variety of vector and virus species and a tight connection between vector and virus (Beaty & Calisher, 1991). Contrary to the generally persistent infection of arthropod cells, peribunyavirus infection of vertebrate cells causes a lytic

infection. The cellular interactions between the virus and the host that allow viral replication without damaging the vector are not known, even though persistent infection studies of mosquito cell cultures conducted with many Orthobunyaviruses have provided some data on the infection mechanism (Borucki et al., 2002). More specifically, the production of defective particles created by the L segment, the encapsulation of the mRNA formed by the S segment, and the self-restriction of the N protein contribute to viral persistence. In infected mosquito cells, transient morphological changes and widespread phyllopod-like structures occur in the cell in the early stages of infection. Furthermore, in virus-infected mosquito cells, the virus passes from cell to cell without damaging the cell membrane (Szemiel et al., 2012).

Female arthropods acquire the virus during blood-sucking from infected hosts. Infected arthropods vary their feeding behavior, such as sucking blood from different host species (Reese et al., 2009). Arthropods are persistently and systematically infected. Vertical (transovarial) transmission has also been reported in some peribunyavirus infections. Transovarial transmission is an significant mechanism for the survival of some peribunyaviruses, especially during the winter months. Many peribunyaviruses are transmitted by arthropod vectors and do not require mammals for their viability. This propose that they initially evolved as the viruses that only infect insects. The recent confirmation of this is that viruses isolated from different biting midge species (such as Herbert virus-Herbert associated viruses) do not replicate in vertebrate cells and are phylogenetically classified as Peribunyavirus (Hughes et al., 2020).

The mechanism for evading innate immunity in vertebrate cells: The foreign molecules known as pathogen-associated molecular patterns (PAMPs) are identified by Pattern recognition receptors (PRRs). In the case of viruses, these pathogen-related molecules include different forms of nucleic acids, such as dsRNA and 5′ triphosphorylated RNA. Melanoma differentiation-associated protein 5 (MDA5) and retinoic acid-inducible gene I (RIG-I) which are RNA helicases recognize these foreign molecules intracellularly (Randall & Goodbourn, 2008). The linking of an RNA ligand to these PRRs activates the signal chain that stimulates the expression of several transcription factors [nuclear factor-κB (NF- κB), IFN regulatory factor 3 (IRF3)] and cyclic AMP-dependent transcription factor (ATF2). This stimulates interferon-(IFNβ) transcription which causes to the upregulation of hundreds of IFN-induced genes (ISG) (Schoggins & Rice, 2011). Recently, RIGI was reported to recognize the La Crosse encephalitis virus (LACV) nucleocapsid containing 5′ triphosphate and terminal panhandle, and recognition precedes transcription. This situation shows that the innate immune response is triggered at the first stage of infection, namely, after viral entry (Weber et al., 2013). There is minimal information about that the antiviral activities of most ISG products but but they

presumably restrict the viral replication at the many stages. For example, MxA protein binds to the newly synthesized N protein and inhibits LACV and BUNV replication, which blocks replication. Interferon (IFN) released from mammalian cells is extremely potent in controlling virus replication. The cause of the replication restriction of Orthobunyaviruses in cells pretreated with IFN has been reported. NSs proteins of BUNV, LACV, and SBV reduce IFN release in mammalian cells (Carlton-Smith & Elliott, 2012).

Hofmann et al. (2015) demonstrated the genetic stability of SBV by comparing the nucleotide sequence of the field isolate identified during the Swiss epidemic in 2012 with SBV isolates in GenBank. They found 101 mutations, mostly randomly distributed along the L and M segments, specifically in sequence between nucleotides 2100 and 2300 of the M segment and nucleotide 2000 of the L segment. The S segments were conserved, while changes in the amino acid level were seen in the highly variable region at the center of the M segment. In contrast, mutations in the L segment did not change amino acid sequences. Coupeau et al. (2013) demonstrated that the greatest genomic variability in viruses isolated during the SBV epidemic in Belgium was in the region encoding the N-terminal domain of the Gc protein in the M segment (1394–2562 nucleotide sequences). They concluded that mutations in the Gc protein might help the virus evade the immune response. Given that this protein is highly immunological and stimulates the neutralizing antibody response in the host when its structure changes, the virus can evade the immune response because previously formed antibodies cannot fully recognize the new structure.

Conclusion

The classification of viruses is constantly changing due to the detection of new viruses, mutations that change amino acid levels, and genetic reassortment in segmented viruses as new viruses are formed. The most important factor contributing to the genetic diversity of Peribunyaviruses is genetic reassortment, which is observed in most segmented viruses. Segment reassortment usually takes place in arthropods co-infected with two distinct peribunyaviruses. As in all viruses, another factor contributing to genetic diversity is mutations. Most mutations in Peribunyaviruses occur in the M segment, particularly in the highly variable region of the Gc surface glycoprotein coding sequence, which helps the virus evade the immune response.

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Ethical Statement

This study does not present any ethical concerns.

Conflict of Interest

The authors declared that there is no conflict of interest.

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