



This form should be used for all taxonomic proposals. Please complete all those modules that are applicable (and then delete the unwanted sections). For guidance, see the notes written in blue and the separate document "Help with completing a taxonomic proposal"

Please try to keep related proposals within a single document; you can copy the modules to create more than one genus within a new family, for example.

MODULE1: **TITLE, AUTHORS, etc**

<b>Code assigned:</b>	<b>2014.005aV</b>	(to be completed by ICTV officers)
<b>Short title:</b> A new species in the genus <i>Tospovirus</i> isolated from <i>Citrullus lanatus</i> and other cucurbitaceous hosts. (e.g. 6 new species in the genus <i>Zetavirus</i> )		
<b>Modules attached</b> (modules 1 and 9 are required)	1 <input checked="" type="checkbox"/> 2 <input type="checkbox"/> 3 <input type="checkbox"/> 4 <input type="checkbox"/> 5 <input type="checkbox"/> 6 <input type="checkbox"/> 7 <input type="checkbox"/> 8 <input type="checkbox"/> 9 <input checked="" type="checkbox"/>	

**Author(s) with e-mail address(es) of the proposer:**

Rakesh Kumar Jain ([rakeshjain56@yahoo.co.in](mailto:rakeshjain56@yahoo.co.in)), Bikash Mandal ([leafcurl@rediffmail.com](mailto:leafcurl@rediffmail.com)), H R Pappu ([hrp@wsu.edu](mailto:hrp@wsu.edu)) and Somnath Kadappa Holkar([somnathk\\_holkar@yahoo.com](mailto:somnathk_holkar@yahoo.com))

**List the ICTV study group(s) that have seen this proposal:**

A list of study groups and contacts is provided at <a href="http://www.ictvonline.org/subcommittees.asp">http://www.ictvonline.org/subcommittees.asp</a> . If in doubt, contact the appropriate subcommittee chair (fungal, invertebrate, plant, prokaryote or vertebrate viruses)	Bunyaviridae study group Chair Elliot, Richard M., <a href="mailto:rme1@st-andrews.ac.uk">rme1@st-andrews.ac.uk</a>
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**ICTV-ECor Study Group comments and response of the proposer:**

The Bunyaviridae Study Group supports this proposal. RM Elliott, 19/06/2014

EC comment: This proposal was conditionally approved. The figure legend to the phylogenetic tree should be modified to describe the method used to build the tree. Please also ensure that the tree is properly rooted using Bunyawera virus as an outgroup.

Proposer Response: The tree has been properly rooted using Bunyawera virus as an outgroup. The method used to build the tree has also been indicated in the figure legend.

Date first submitted to ICTV:

May 8, 2014

Date of this revision (if different to above):

June 19 2014

MODULE2: **NEW SPECIES**

creating and naming one or more new species.

If more than one, they should be a group of related species belonging to the same genus. All new species must be placed in a higher taxon. This is usually a genus although it is also permissible for species to be "unassigned" within a subfamily or family. Wherever possible, provide sequence accession number(s) for one isolate of each new species proposed.

Code	<b><i>2014.005aV</i></b>	(assigned by ICTV officers)
<b>To create <i>Watermelon bud necrosis virus</i>, a new species within:</b>		
Genus:	<i>Tospovirus</i>	Fill in all that apply. • If the higher taxon has yet to be created (in a later module, below) write " <b>(new)</b> " after its proposed name. • If no genus is specified, enter " <b>unassigned</b> " in the genus box.
Subfamily:		
Family:	<i>Bunyaviridae</i>	
Order:		
<b>And name the new species:</b>		<b>GenBank sequence accession number(s) of reference isolate:</b>
<i>Watermelon bud necrosis virus</i>		GU584184; GU584185; GU735408

### Reasons to justify the creation and assignment of the new species:

An unusual disease characterized by leaf mottling, yellowing and necrotic streak of veins, shortened internodes and necrosis and dieback of the buds (Fig.1) was observed on watermelon (*Citrullus lanatus*) during 1991-92 at the Indian Institute of Horticultural Research experimental farm near Bangalore, India[1]. Since then the disease has become increasingly important on other cucurbitaceous hosts in other vegetable growing states in India [3]. Initial studies on the etiology revealed that the causal agent is a tospovirus serologically related to *Watermelon silver mottle virus* (WSMoV) serogroup [1] and is vectored by *Thrips palmi*. Subsequently, nucleocapsid protein (N) gene was sequenced [2]. The percentage of amino acid sequence identity in the N protein has been identified as a suitable descriptor for the taxonomy of tospoviruses and identity of less than 90% is considered a species demarcation criteria. Host range, vector specificity and serological relationships of the N protein are also considered. Based on these criteria, the watermelon tospovirus was suggested to constitute a distinct tospovirus species, i.e. Watermelon bud necrosis virus (WBNV)[2]. There are currently nine species in the genus *Tospovirus*; in addition a number of other tospoviruses have been partially characterized but not further classified (see Table 2). Comparative amino acid sequence homology revealed that N gene of WBNV shared 81-85% identity with GBNV, WSMoV, CaCV, GloxRSV and TNeV (for definition of abbreviations, see Table 2) and 25-64% identity with other tospoviruses (Table 1 and 2). Cluster dendrogram also revealed a similar trend and WBNV, GBNV, WSMoV, CaCV, GloxRSV and TNeV formed one cluster (Fig.2). The phylogenetic profiles of other virus encoded proteins, NSs, NSm, Gn/Gc and RdRp of WBNV are also consistent with that of N protein [4, 5]. Considering distinct differences in the N protein and other proteins and host range, we propose that WBNV constitutes a distinct tospovirus species.

Explain how the proposed species differ(s) from all existing species.

- If species demarcation criteria (see module 3) have previously been defined for the genus, **explain how the new species meet these criteria.**
- If criteria for demarcating species need to be defined (because there will now be more than one species in the genus), please state the proposed criteria.
- Further material in support of this proposal may be presented in the Appendix, Module 9

MODULE9: **APPENDIX**: supporting material

additional material in support of this proposal

**References:**

1. Singh, S.J. and Krishnareddy, M. (1996). Watermelon bud necrosis: A new tospovirus disease. *Acta Hort.* 431: 68-78
2. Jain, R.K., Pappu, H.R., Pappu, S.S., Krishna Reddy, M. and Vani, M. (1998). Watermelon bud necrosis tospovirus is a distinct virus species belonging to serogroup IV. *Arch. Virol.* 143: 1637-1644.
3. Jain, R.K., Bag, S., Umamaheswaran, K. and Mandal, B. (2007). Natural infection by tospoviruses of cucurbitaceous and fabaceous vegetable crops in India. *J. Phytopathology.* 155: 22-25
4. Rakesh Kumar., Mandal, B., Geetanjali A.S., Jain, R.K. and Jaiwal P.K. (2010). Genome organization and sequence comparison suggest intraspecies incongruence in M RNA of Watermelon bud necrosis virus. *Arch.Virol.* 155(8): 1361-1365
5. Li, Ju.Ting., Yeh, Yi-Chun., Yeh, Shy-Dong., Raja, Joseph A.J., Rajagopalan, Prem.A., Liu Lu.Yuan. and Chen, Tsung.Chi. (2010). Complete genomic sequence of watermelon bud necrosis virus. *Arch. Virol.* 156(2): 359-362

**Annex:**

Include as much information as necessary to support the proposal, including diagrams comparing the old and new taxonomic orders. The use of Figures and Tables is strongly recommended but direct pasting of content from publications will require permission from the copyright holder together with appropriate acknowledgement as this proposal will be placed on a public web site. For phylogenetic analysis, try to provide a tree where branch length is related to genetic distance.

Table 1. Percentage of amino acids identity of nucleocapsid protein gene of WBNV aligned with other tospoviruses (for definitions of abbreviations, see Table 2)

	AF045067[WBNV]	U27809 [GBNV]	U78734 [WSMoV]	ABC86907 [CaCV]	ACZ18222 [ANSV]	AER23986 [BeNMV]	D00353 [BUNV]	AAW58115 [CCSV]	AAF04197 [CSNV]	AAQ83791 [GloxRSV]	AAE25255 [GRSV]	KC290943 [HCRSV]	AAA47944 [INSV]	AAF75556 [YSV]	EU275149 [MSMV]	BAB79455 [MYSV]	AAAC99405 [PCFV]	AAB94022[PYSV]	AAD34201 [PSMV]	EF445397 [PaRSV]	ADX96066 [SVNaV]	AAAG23654 [TCSV]	AAT68025 [TNeV]	ACK99533 [TNRSV]	BAA03025 [TSWV]	ABF59486 [TYRV]	EF552433[TZSV]	AAF04198 [ZLCV]
AF045067[WBNV]	ID	85	85	81	26	28	10	64	25	82	28	41	27	43	28	59	18	17	59	42	30	25	82	54	27	42	63	25
U27809 [GBNV]		ID	86	83	26	28	10	65	26	84	27	42	26	44	29	59	20	17	59	42	31	26	85	56	27	43	62	27
U78734 [WSMoV]			ID	87	26	28	10	63	26	87	29	41	28	43	28	58	19	18	58	42	29	26	85	55	28	43	62	27
ABC86907 [CaCV]				ID	26	27	8	64	25	98	29	42	28	46	27	59	19	18	59	42	30	27	92	57	30	43	62	26
ACZ18222 [ANSV]					ID	34	10	25	74	26	81	28	53	28	59	24	16	16	24	30	33	82	26	24	79	30	23	76
AER23986 [BeNMV]						ID	11	28	35	27	33	30	33	29	34	28	16	15	28	30	52	33	27	30	33	27	27	36
D00353 [BUNV]							ID	10	12	9	11	11	11	11	9	11	10	10	11	10	11	11	9	7	11	10	10	10
AAW58115 [CCSV]								ID	23	63	25	42	23	46	25	57	17	15	57	43	29	23	63	56	25	46	81	24
AAF04197 [CSNV]									ID	25	73	27	54	28	60	27	17	18	27	28	33	74	26	25	77	28	23	80
AAQ83791 [GloxRSV]										ID	29	41	28	46	28	58	19	18	58	42	30	27	93	57	30	43	62	26
AAE25255 [GRSV]											ID	28	53	29	59	25	16	17	25	29	33	85	28	26	79	31	25	76
KC290943 [HCRSV]												ID	25	67	28	46	17	16	46	86	31	27	43	44	28	82	41	28
AAA47944 [INSV]													ID	26	51	23	16	17	24	26	30	52	27	23	54	26	22	52
AAF75556 [YSV]														ID	29	50	17	17	49	67	30	29	46	43	30	69	45	27
EU275149 [MSMV]															ID	26	18	20	26	27	33	58	27	25	56	28	24	59
BAB79455 [MYSV]																ID	17	18	99	47	30	24	59	50	25	47	57	25
AAAC99405 [PCFV]																	ID	60	18	18	17	16	19	17	16	16	17	16
AAB94022[PYSV]																		ID	17	18	16	16	20	15	16	16	16	16
AAD34201 [PSMV]																			ID	46	30	25	59	50	25	46	57	25
EF445397 [PaRSV]																				ID	30	29	43	44	28	83	41	29
ADX96066 [SVNaV]																					ID	33	29	30	32	30	29	34
AAAG23654 [TCSV]																						ID	28	25	80	29	23	75
AAT68025 [TNeV]																							ID	57	30	44	62	27
ACK99533 [TNRSV]																								ID	28	45	55	24
BAA03025 [TSWV]																									ID	29	26	73
ABF59486 [TYRV]																										ID	41	29
EF552433[TZSV]																											ID	22
AAF04198 [ZLCV]																												ID

Table 2. GenBank accession numbers of the nucleocapsid amino acid sequences of tospoviruses used for comparison and phylogenetic analysis. The nine approved tospovirus species are listed first, in italics, followed by the unclassified tospoviruses. Bunyamwera virus N protein was used as an outgroup.

<b>Accepted Tospovirus species</b>	<b>Accession numbers</b>
<i>Groundnut bud necrosis virus [GBNV]</i>	U27809
<i>Watermelon silver mottle virus [WSMoV]</i>	U78734
<i>Groundnut ringspot virus [GRSV]</i>	AAB25255
<i>Impatiens necrotic spot virus [INSV]</i>	AAA47944
<i>Groundnut yellow spot virus [GYSV]</i>	AAB94022
<i>Polygonum ringspot virus [PoIRSV]</i>	EF445397
<i>Tomato chlorotic spot virus [TCSV]</i>	AAG23654
<i>Tomato spotted wilt virus [TSWV]</i>	BAA03025
<i>Zucchini lethal chlorosis virus [ZLCV]</i>	AAF04198
Watermelon bud necrosis virus [WBNV] proposed new species	AF045067
<b>Unclassified tospoviruses</b>	
Capsicum chlorosis virus [CaCV]	ABC86907
Alstroemeria necrotic streak virus [ANSV]	ACZ18222
Bean necrotic mosaic virus [BeNMV]	AER23986
Calla lily chlorotic spot virus [CCSV]	AAW58115
Chrysanthemum stem necrosis virus [CSNV]	AAF04197
Gloxinia ringspot virus [GloxRSV]	AAQ83791
Hippeastrum chlorotic ringspot virus [HCRSV]	KC290943
Iris yellow spot virus [IYSV]	AAF75556
Melon severe mosaic virus [MSMV]	EU275149
Melon yellow spot virus [MYSV]	BAB79455
Peanut chlorotic fan-spot virus [PCFV]	AAC99405
Physalis severe mottle virus [PSMV]	AAD34201
Soybean vein necrosis virus [SVNaV]	ADX96066
Tomato necrosis virus TD8 [TNeV]	AAT68025
Tomato necrotic ringspot virus [TNRSV]	ACK99533
Tomato yellow ring virus [TYRSV]	ABF59486
Tomato zonate spot virus [TZSV]	EF552433
<b>Outgroup sequence</b>	
<i>Bunyamwera virus</i> [BUNV] used as outgroup	D00353

Fig.1 Symptoms of Watermelon bud necrosis virus (WBNV) in watermelon under field conditions A: necrotic spots on leaves; B: bud necrosis; C: stunted growth of the plant D: stem necrosis; E: shortened internodes and F: deformed fruit with necrotic spots



Fig 2. Phylogenetic tree of the amino acid sequences of the nucleocapsid protein gene of tospoviruses.

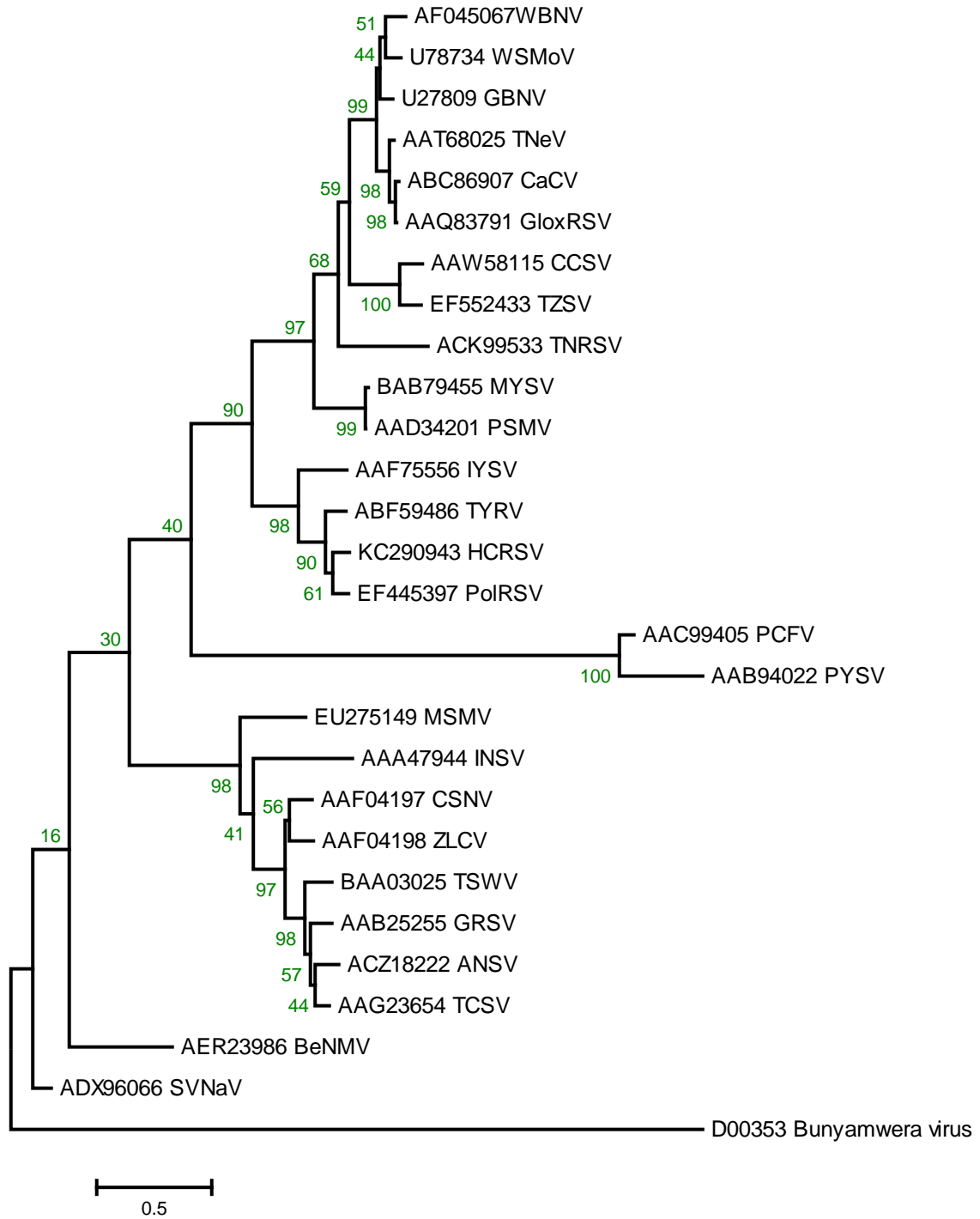


Figure 2. Cluster dendrogram showing the relationship among the deduced amino acid sequences of nucleocapsid protein of WBNV with those of known tospoviruses. The percentage of replicate trees in which the associated taxa clustered together in the bootstrap test (1000 replicates) is shown next to the branches. The scale bar shows the evolutionary distances in the units of base substitutions. Cluster dendrogram was generated using MEGA 5.2.2 (Tamura *et al.*, 2011)