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Research Article

The Occurrence of Three Allexiviruses from Imported Garlic in Indonesia

Amanda Febriyanti Nurdevi[1], Sari Nurulita[1*], Didiet Rahayu Diana[2], Sri Hendrastuti Hidayat[1]

[1]*Department of Plant Protection, Faculty of Agriculture, IPB University, Kampus IPB Dramaga, Bogor, 16680, Indonesia*
[2]*Regional Quarantine Service, Indonesia Quarantine Agency, Tanjung Priok, North Jakarta 14310, Indonesia*

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ABSTRACT

International trade in fresh horticultural commodities poses a high risk of emerging pests and diseases, including plant viruses. Infections of potyvirus, carlavirus, and allexivirus cause mosaic disease on garlic and potential yield loss. Potyvirus and Carlavirus have been well studied in Indonesia, but not for Allexivirus. Therefore, this study aimed to detect and identify of allexivirus species from imported garlic (consumption garlic) using reverse-transcription polymerase chain reaction (RT-PCR). Samples of garlic bulbs were collected from the plant quarantine inspection site of the regional quarantine service, North Jakarta City, DKI Jakarta Province. Individual garlic cloves were then germinated in water media. Common symptoms observed include mosaic, yellow stripes, twisted leaves, and leaf malformation. Molecular detection was done by RT-PCR using specific primers for three allexivirus species, namely Garlic virus A (GarVA), Garlic virus B (GarVB), and Garlic virus E (GarVE). Specific DNA fragments for GarVA, GarVB, and GarVE were successfully amplified from all tested samples. Nucleotide sequence analysis using SDT Matrix revealed the highest similarity of GarVA, -B, and -E with Chinese garlic isolates, i.e., 99.5%, 99.1%, and 95.0%, respectively. Further analysis revealed that Indonesian allexiviruses demonstrated a high genetic diversity and have a close genetic relationship with isolates from China. This study provides information about the occurrence of three allexiviruses in Indonesia and suggests avoiding the use of imported garlic bulbs for vegetative propagation.

1. Introduction

Garlic (*Allium sativum*) is an economically important horticultural commodity and is primarily used as one of the main ingredients in Indonesian cuisines. Garlic has benefits as a medicinal remedy due to its bioactive compounds, including allicin, phenolic, saponins, and flavonoids (Gupta *et al.* 2015; Verma *et al.* 2023). However, domestic production of garlic is insufficient to meet national demand, and this situation has consequences for imports of this commodity. Since 2020, local garlic production has been decreasing, with the lowest recorded value in 2022, with total production of 30.5 thousand tons (BPS 2024). In 2024, the demand for garlic in Indonesia reached 345.5 thousand tons, with China, India, and Germany as major garlic exporter countries (BPS 2024).

International trading of fresh horticultural commodities poses a risk of introducing new or emerging pests and diseases, such as plant viruses. Previous studies reported main virus groups infecting *Allium* spp., including potyvirus, carlavirus, allexivirus, and orthotospovirus (Chen *et al.* 2001; Melo-Filho *et al.* 2004; Katis *et al.* 2012; Bag *et al.* 2015). Potyvirus and Carlavirus, as common viruses infecting allium, have been widely studied in Indonesia, while more research is needed for Allexivirus. The genus *Allexivirus*, family *Alphaflexiviridae* has the mos members among viruses infecting allium, namely *Garlic virus* A, B, C, D, E, X

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* Corresponding Author
E-mail Address: sarinurulita@apps.ipb.ac.id

(GarVA, -B, -C, -D, -E, -X) and *Shallot virus* X (ShVX) (ICTV 2024). Allexivirus infection caused a 14 to 32% decrease in bulb weight and a 6% to 11% reduction in bulb diameter (Cafrune *et al.* 2006). Mixed infections of these viruses resulted in complex symptoms, including chlorotic leaf streaks, stunted growth, and a significant reduction in bulb weight (Cremer *et al.* 2021). Allexiviruses can be transmitted via infected propagative material through dry bulb mites *Aceria tulipae* (Acari: Eriophyidae) in a semi-persistent manner during the vegetative period and bulb storage (Mansouri *et al.* 2021).

The presence of allexiviruses in Indonesia represents a new threat to domestic garlic cultivation. A previous study by Swari *et al.* (2015) reported the presence of allexivirus in shallot (*Allium cepa* var. *aggregatum*) cultivar 'Parangkusumo' from Bantul, with an incidence rate of 10%. Further study of allexivirus has not yet been carried out, especially on imported garlic (consumption bulbs) in Indonesia. Therefore, accurate detection of this virus is essential to obtain information on the presence of allexivirus in garlic imported into Indonesia.

2. Materials and Methods

2.1. Sample Collection

Imported garlic bulbs originally from China were obtained from the plant quarantine inspection site of the regional quarantine service, North Jakarta City, DKI Jakarta Province. Bulb germination and virus detection were conducted at the Plant Virology Laboratory of the Department of Plant Protection, Faculty of Agriculture, IPB University, from December 2024 until April 2025.

2.2. Bulbs Germination

Garlic bulb samples were selected randomly and separated into individual cloves. The individual cloves from one bulb were then germinated in a plastic tray measuring 36 × 24 cm using the floating method with distilled water as the medium. The leaves were then harvested at 2 to 4 weeks after planting (WAP) and made into composites. One composite sample consisted of five leaves from different bulbs, with a total leaf mass of 0.1 g, to be used for virus detection.

2.3. RT-PCR

Total RNA was extracted using a purification kit (Thermo-Fisher Scientific GeneJET RNA Purification, Waltham, USA). The RT-PCR cocktail consisted of 12.5 µL DreamTaq green PCR master mix (2×) (Thermo-Fisher Scientific, Waltham, USA); 2 µL each of 10 µM reverse and forward primers (Table 1); 2 µL of 0.1 M DTT (Smobio, Taiwan); 0.25 µL RT enzyme (200U/µL); 0.5 µL RNAse inhibitor (Bioline, London, UK); 2 µL RNA template; and 4.5 µL nuclease-free water for a total volume of 25 µL. GeneAmp PCR system 9700 machine (Applied Biosystems, Thermo-Fisher Scientific, US) was used to amplify virus sequences, with an initial stage of cDNA synthesis at 45°C for 60 minutes, followed by a pre-denaturation step at 95°C for one minute. This was followed by 35 cycles consisting of denaturation at 95°C for 30 seconds, annealing for 30 seconds at temperatures listed in Table 1 according to Nurulita *et al.* (2026), followed by elongation at 72°C for one minute, and final elongation at 72°C for 10 minutes.

The amplification results were visualized using 1% agarose gel with 0.3 g agarose powder dissolved in 30 mL of 0.5 × TBE (Tris-borate EDTA) buffer and 3 µL FluoroVue (Smobio, Taiwan). Electrophoresis was performed at 100 volts for 30 minutes, followed by visualization using a UV transilluminator. Samples with positive results were sent for Sanger sequencing to First BASE Malaysia through PT Genetika Science Indonesia.

2.4. Sequence Identity and Phylogenetic Analysis

Sequences of the samples were edited using BioEdit software to generate sample contigs. All the contigs were then checked for their similarity through the Basic Local Alignment Search Tool (BLAST). Selected sequences of each virus target were then aligned in the Molecular Evolutionary Genetics Analysis (MEGA) program to create a percentage identity matrix and construct a phylogenetic tree. The identity matrix to analyze

Table 1. Specific primers and annealing temperature for allexivirus detection

Viruses	Primer code	Primer sequences (5'–3')	Target size (bp)	Ta*(°C)
GarVA	GarVA-F	GCRCACTCRGARCTCAARGA	610	51
	GarVA-R	TCYRAYTGAGCRCGTGA		
GarVB	GarVB-F	TAGCCTGTYTACWATTTGCTCA	250	53
	AllexiNAB-R	CCYTTCAGCRTRTAGCTTARC		
GarVE	GarVE-F	GTCCAGGAAAGGCTACCA	360	56
	GarVE-R	AGYCTACGTAATTTACATTCAGT		

sequence homology was visualized using the Sequence Demarcation Tool (SDT matrix version 1.3) (Muhire *et al.* 2014). Phylogenetic tree analysis was performed with MEGA X using 1000 bootstrap replications by the Kimura 2 model (Kimura 1980).

2.5. Genetic Diversity Analysis of Allexiviruses

Genetic diversity was analyzed using DNA sequence polymorphism software (DNAsp version 6) involving three parameters, i.e., the number of haplotypes (Hn), haplotype diversity (Hd), and nucleotide diversity (Pi) (Rozas *et al.* 2017). The analysis based on Hd values was classified as a low category at Hd value 0–0.5 and a high category at Hd value 0.5–1. Nucleotide diversity (Pi) was categorized as low (0.01–0.04), moderate (0.05–0.07), or high (0.08–0.1) (Nei and Kumar 2000). Fixation index values (Fst) as the parameter of population analysis were categorized to have high genetic differentiation at values greater than 0.33 (Luo *et al.* 2016). The haplotype network of sequences was visualized using Popart software (Leigh and Bryant 2015).

3. Results

3.1. Virus Symptoms and Detection of Allexivirus in Garlic Samples

The symptoms of virus infection were observed from the germination of garlic bulbs. In general, the germinated bulbs displayed typical symptoms, including mosaic, yellow stripes, twisted leaves, and malformation on the leaves (Figure 1).

Allexiviruses were detected in all samples and were commonly found in mixed infection (Table 2). GarVA was successfully amplified in all tested samples except CH-2 with the expected size of ±610 bp. GarVB and GarVE were detected in each composite sample with amplification sizes of ±260 bp and ±360 bp, respectively. GarVE infection was confirmed in only two samples, ‘CH-2’ and ‘CH-3’. The occurrence of multiple infections influences the symptoms of infection in plants. A synergistic effect was evidenced, resulting in more severe symptoms when compared to those caused by single infections.

3.1.1. Identity of Garlic Virus A

Nucleotide sequence of isolates from this study that have been aligned with GenBank accessions revealed similarity levels. Red squares indicate the highest similarity, while the lowest similarity is shown in blue (Figure 2). GarVA isolates in this study shared the highest nucleotide similarity (99.5%) with the Chinese GarVA isolate (MN059258.1) and an Australian isolate (JX997952.1) (98.4%). GarVC isolate from China (MN059141.1) was used as an outgroup and thus showed the lowest similarity with the other isolates.

Phylogenetic analysis revealed that the GarVA phylogenetic tree is divided into two major clades (Figure 3). The first clade, which includes the isolates

Table 2. Detection of three allexiviruses infecting garlic

Type of sample	Sample origin	Sample code	GarVA[a]	GarVB[b]	GarVE[c]
Imported garlic	China	4213 (CH-1)	+	-	-
		4554 (CH-2)	-	+	+
		4216 (CH-3)	+	+	+
		4649 (CH-4)	+	+	-

[a]Garlic virus A; [b]Garlic virus B; [c]Garlic virus E; [d]positive reaction; [e]negative reaction

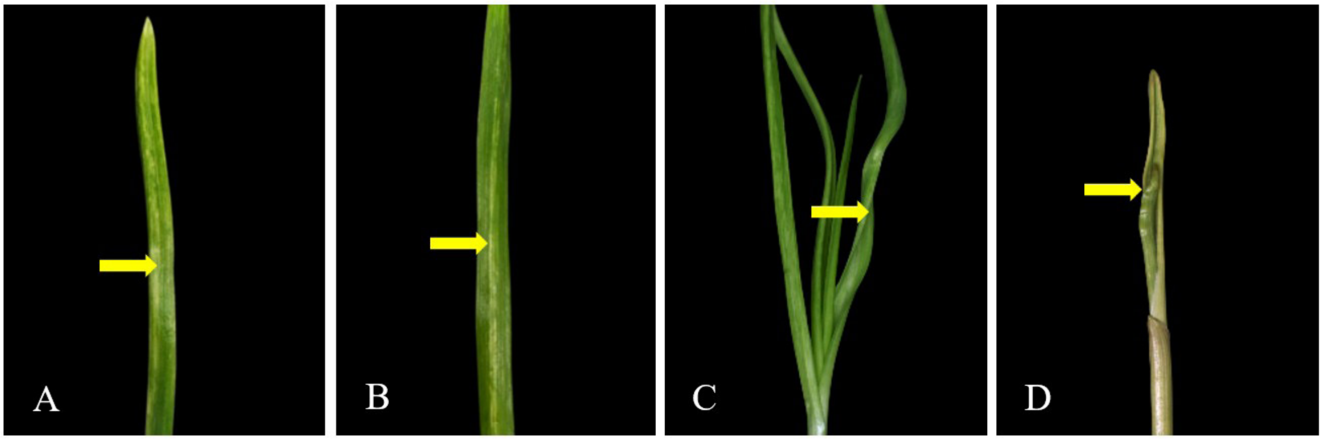

Figure 1. Symptom variations of virus-infected garlic. (A) leaf mosaic, (B) yellow striped on the leaf, (C), leaf twisted, (D) leaf malformation. Each symptom is indicated by yellow arrow

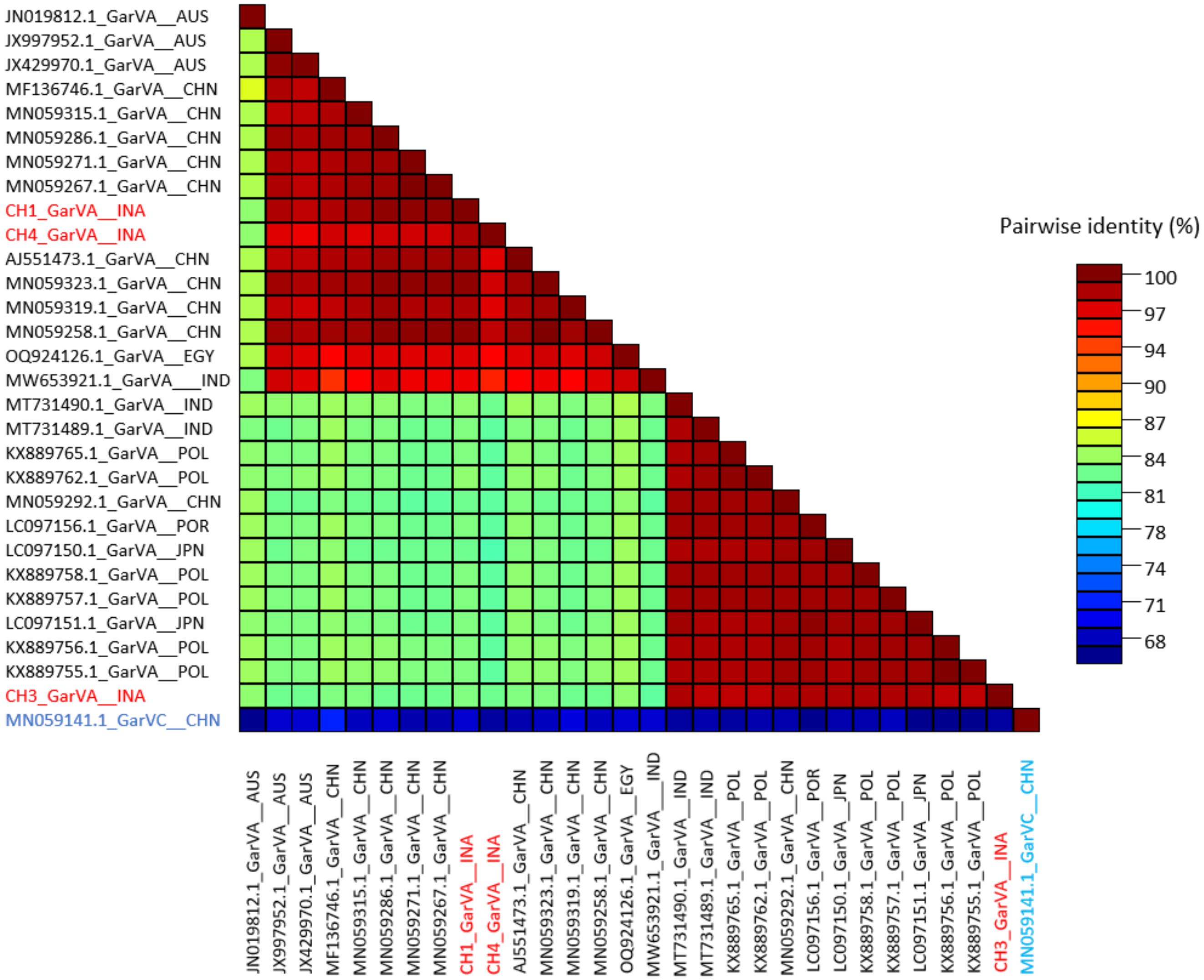

Figure 2. Sequence demarcation tool (SDT) matrix of Garlic virus A (GarVA) isolates based on pairwise sequence comparison. Garlic virus C (GarVC) isolate was used as an outgroup and highlighted with blue font. Sequences generated in this study are highlighted with red font

CH-1 and CH-4, was grouped closely with multiple isolates from China. CH-1 isolate was clustered with other isolates in the second clade. The significant distance between groups indicates a high level of diversity among GarVA isolates. In addition, the phylogenetic tree also showed that imported garlic isolates of GarVA in Indonesia had a close relationship with Chinese isolates, as their origin country.

3.1.2. Identity of Garlic Virus B

GarVB isolate from Indonesia exhibited low genetic variability, with pairwise nucleotide identity values ranging from 96.6 to 99.1% (Figure 4). This indicates that this isolate is very similar to other isolates in GenBank. GarVB isolates from this study shared the highest sequence identity (99.1%) with isolates from China (MN059150.1; KX889770.1) and Japan (LC097162.1). The minimum interspecific identity was 92.8%, further supporting the relatively conserved nature of the GarVB genome. Overall, the sequence similarity among GarVB isolates was consistently high, suggesting limited genetic diversity within this virus species.

The phylogenetic tree of GarVB revealed a close relationship among isolates, indicating a high degree of genetic similarity (Figure 5). GarVB isolates from this

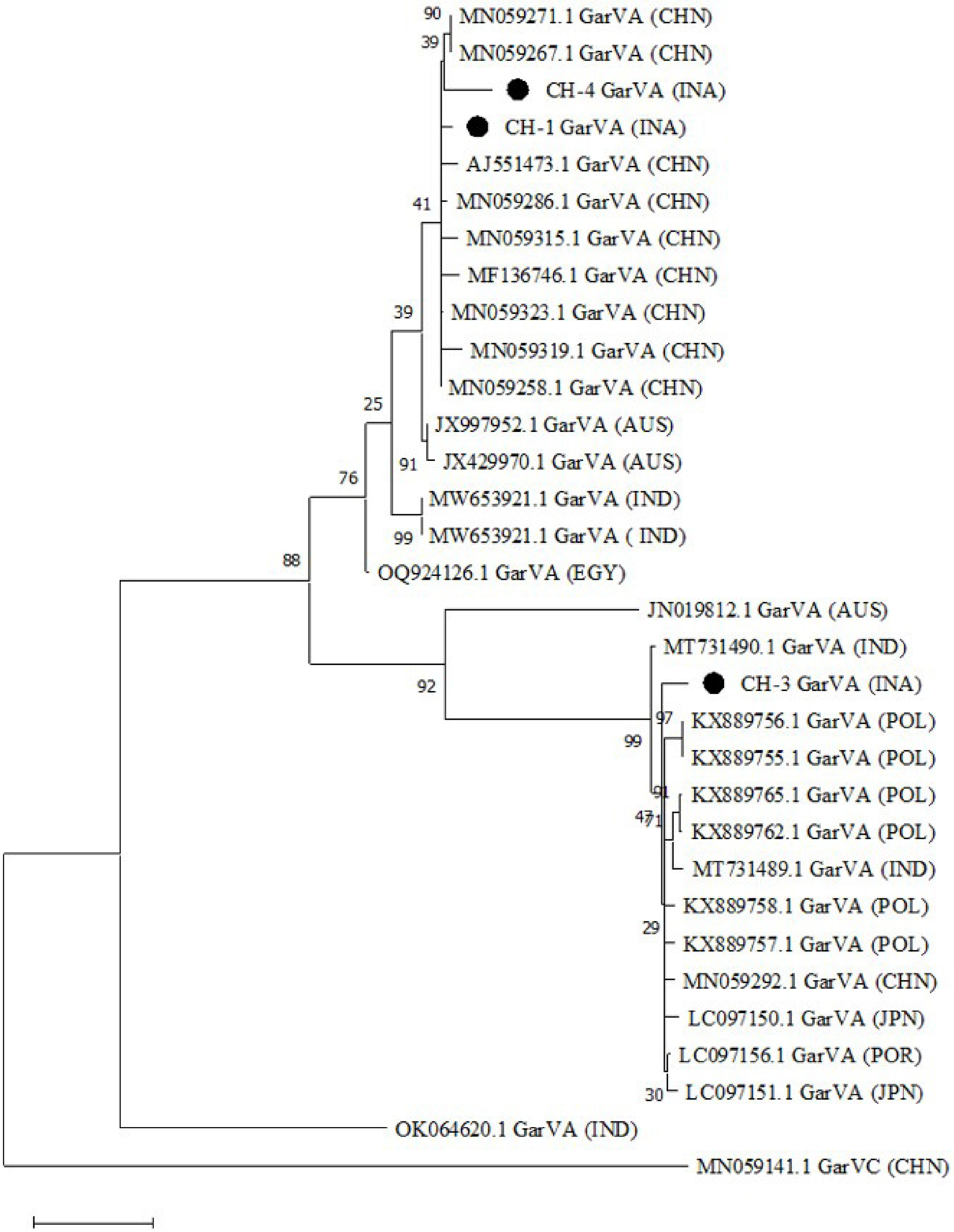

Figure 3. Phylogenetic tree of Garlic virus A (GarVA) based on nucleotide sequences showing the relationship of three different allexiviruses isolates identified in this study with selected allexivirus sequences in the GenBank. Garlic virus C (GarVC) isolate was used as an outgroup. Isolates from this study were marked with black dot colour the GenBank accession number for CH-1 GarVA is LC897734.1, CH-3 GarVA is LC897735.1, CH-4 GarVA is LC897735.1 and they were originally from China (imported garlic)

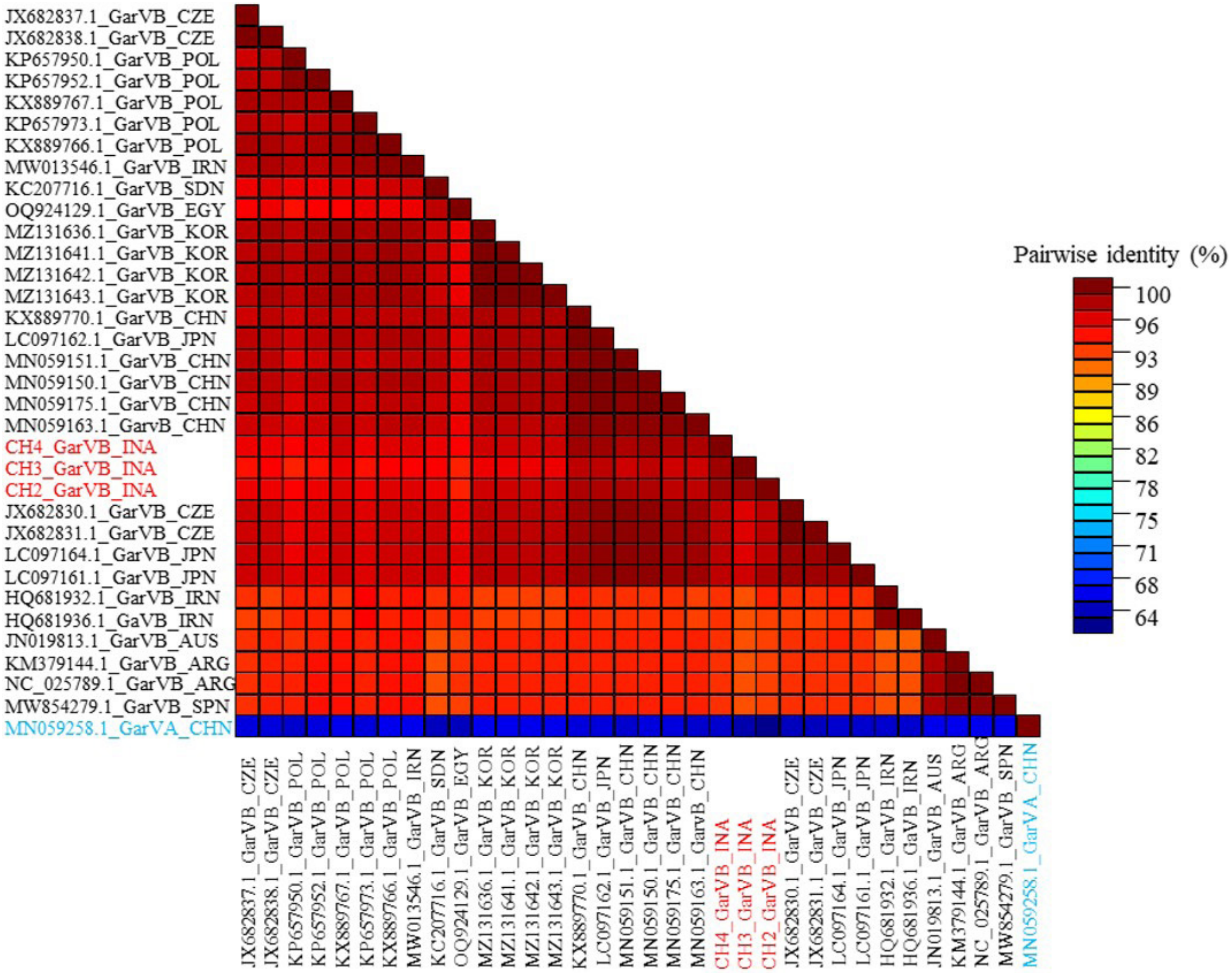

Figure 4. Sequence demarcation tool (SDT) matrix of Garlic virus B (GarVB) isolates based on pairwise sequence comparison. Garlic virus A (GarVA) isolate was used as an outgroup and highlighted with blue font. Sequences generated in this study are highlighted with red font

study were clustered closely with isolates from China and Japan. These findings suggest that the Indonesian GarVB isolate may have originated from a similar strain shared with those circulating in China and Japan, possibly introduced through the movement of infected plant materials or international trade routes.

3.1.3. Identity of Garlic Virus E

Comparative analysis of GarVE isolates revealed a high degree of sequence variability, with pairwise nucleotide identity ranging from 78.2 to 99.8% (Figure 6). GarVE isolate from Indonesia exhibited the highest sequence similarity (95%) with isolates from China (LC097187.1), Japan (LC097189.1; LC097188.1), and the USA (LC097184.1; LC097185.1). The lowest identity (88%) was observed with Indian isolates (OK064618–OK064620).

Indonesian GarVE isolates were positioned on a branch closely related to isolates from Japan (LC097189.1; LC097188.1), USA (LC097184.1; LC097185.1), and China (LC097187.1), supported by relatively high bootstrap values (Figure 7). The close genetic relationship between the Indonesian isolate and those countries suggests a common evolutionary origin, likely facilitated by the international movement of vegetative plant materials. Such trade-related activities may have contributed to the introduction and dissemination of genetically similar GarVE strains across the region.

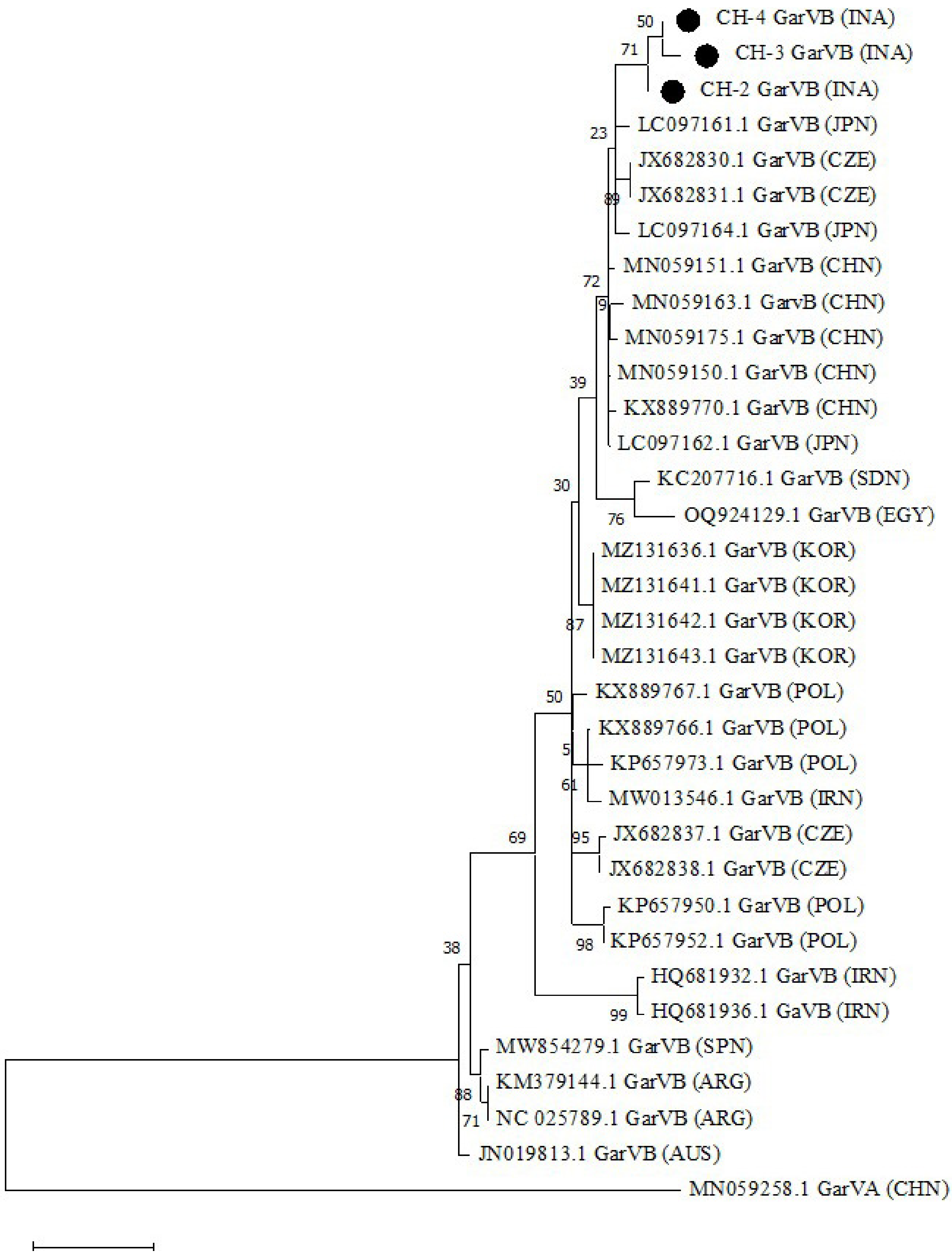

Figure 5. Phylogenetic tree of Garlic virus B (GarVB) based on nucleotide sequences showing the relationship of three different allexiviruses isolates identified in this study with selected allexivirus sequences in the GenBank. Garlic virus A (GarVA) isolate was used as an outgroup. Isolates from this study are marked in red dot colour the GenBank accession number for CH-2 GarVB is LC897737.1, CH-3 GarVB is LC897738.1, CH-4 GarVB is LC897739.1 and they are originally from China

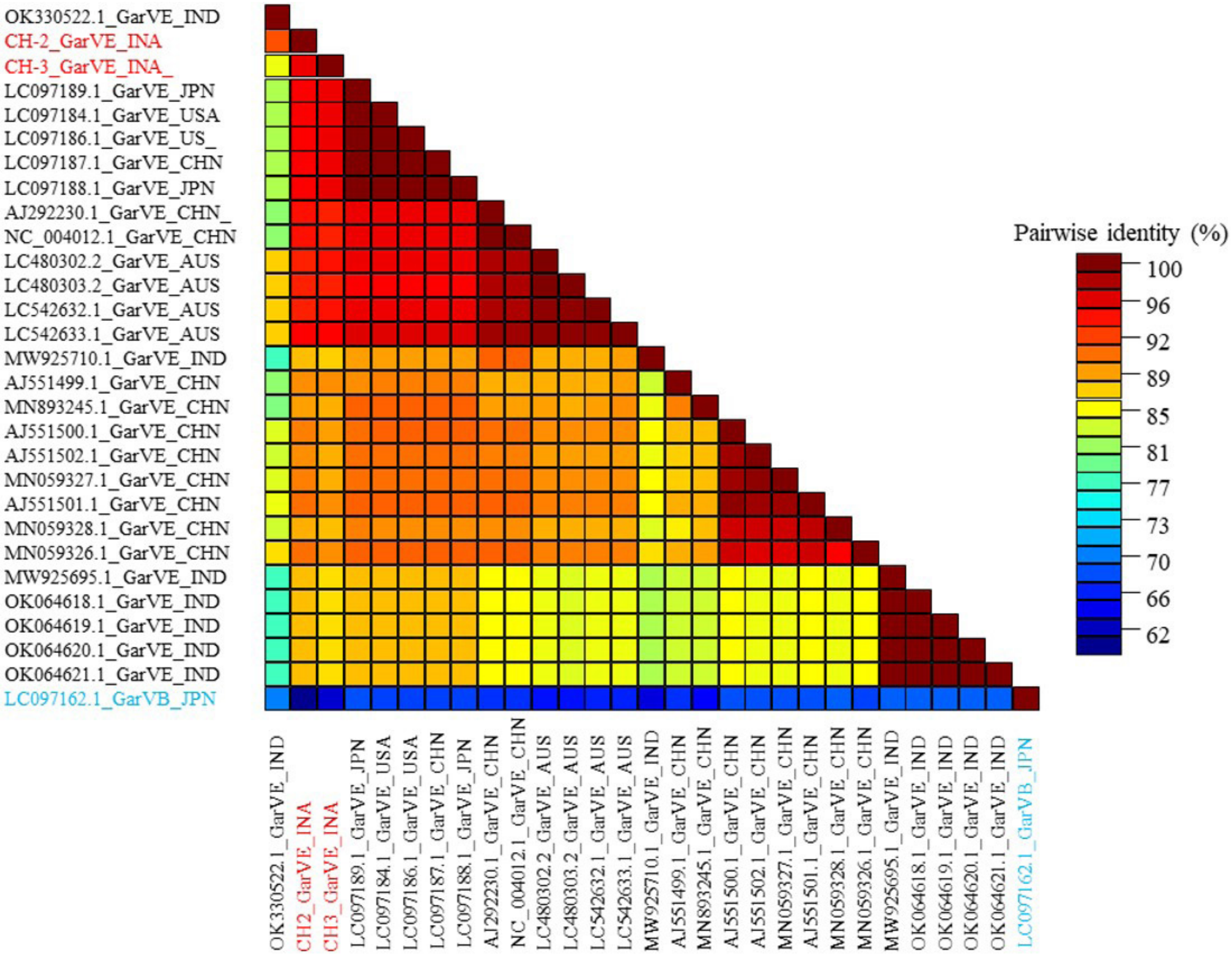

Figure 6. Sequence demarcation tool (SDT) matrix of Garlic virus E (GarVE) isolates based on pairwise sequence comparison. Garlic virus B (GarVB) isolate used as an outgroup and highlighted with blue font. Sequences generated in this study are highlighted with red font

3.2. Genetic Diversity of Allexiviruses

Genetic diversity reflects the extent of genetic variation within viral populations (Table 3). Indonesian GarVA population exhibited a high value of haplotype diversity (Hd = 1) and a relatively high nucleotide diversity (Pi = 0.113), indicating genetic differentiation among isolates. This is further supported by the number of haplotypes that is equal to the number of sequences, suggesting that each isolate possesses a unique haplotype. In contrast, the Chinese GarVA population exhibited a lower number of haplotypes (Hn = 27) compared to the number of individuals (N = 33), indicating the presence of identical haplotypes within the population. Consequently, the haplotype diversity of the Chinese population (Hd = 0.986) was slightly lower than that of the Indonesian population (Hd = 1), suggesting lower genetic differentiation within the Chinese population. These results underscore regional differences in the genetic structure and evolutionary dynamics of GarVA populations.

A low nucleotide diversity (Pi = 0.009), followed by a high haplotype diversity (Hd = 1.000), was observed in the GarVA population from Japan. A similar pattern was found in the GarVB population from Indonesia (Hd = 1.000; Pi = 0.005). These results suggest that even small nucleotide differences can significantly influence allele variation, leading to haplotype diversification within the population. Low genetic diversity was identified in the Korean GarVB population and GarVE from the United States and Japan, with both haplotype and nucleotide diversity values reported as zero. This indicates that the viruses within these

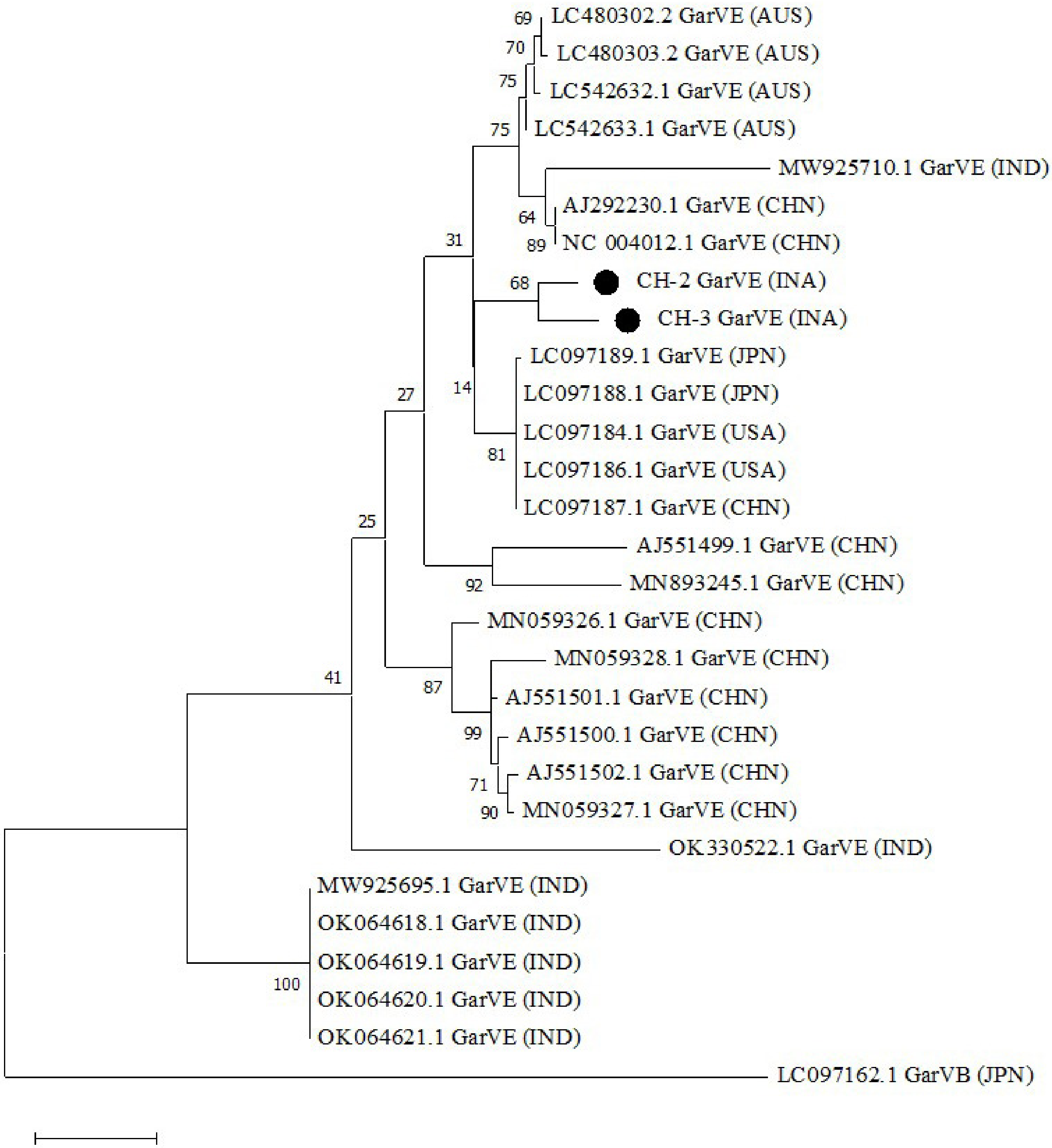

Figure 7. Phylogenetic tree of Garlic virus E (GarVE) based on nucleotide sequences showing the relationship of three different allexiviruses isolates identified in this study with selected allexivirus sequences in the GenBank. Garlic virus B (GarVB) isolate was used as an outgroup. Isolates from this study are marked in red dot colour the GenBank accession number for CH-2 GarVE is LC897740.1, CH-3 GarVE is LC897741.1 and they are originally from China

Table 3. Genetic diversity of *Garlic virus* A, -B, and -E infecting garlic

Virus	Population	N[a]	Parameters		
			Hn[b]	Hd[c]	Pi[d]
Garlic virus A	Indonesia[e]	3	3	1.000	0.113
	China	33	27	0.986	0.115
	Australia	5	5	1.000	0.134
	Poland	3	3	1.000	0.011
	Japan	2	2	1.000	0.009
	Vietnam	2	2	1.000	0.043
	All population	48	40	0.992	0.118
Garlic virus B	Indonesia	3	3	1.000	0.005
	Czech	4	3	0.833	0.018
	Poland	10	9	0.978	0.024
	China	17	8	0.882	0.026
	Japan	7	6	0.952	0.016
	Spain	2	2	1.000	0.016
	Korea	4	1	0.000	0.000
	All population	47	31	0.969	0.029
Garlic virus E	Indonesia	2	2	1.000	0.049
	China	11	10	0.981	0.084
	USA	2	1	0.000	0.000
	Japan	2	1	0.000	0.000
	Australia	4	4	1.000	0.008
	India	7	3	0.523	0.100
	All population	28	18	0.941	0.112

[a]number of sequences (population size), [b]number of haplotypes, [c]haplotype diversity, [d]nucleotide diversity, [e]samples origin from China

populations were not significantly differentiated and exhibited a high degree of genetic similarity. Genetic homogeneity within each population is supported by the number of haplotypes being lower than the number of sequences, suggesting that multiple sequences share identical haplotypes or are highly similar. In contrast, the Indonesian GarVE population exhibited distinct haplotypes among isolates, as evidenced by an equal number of haplotypes and sequences.

3.3. Genetic Population of Allexiviruses

Population structure is indicated by the fixation index (Fst), which reflects genetic differentiation among populations. Indonesian GarVA population exhibited the lowest Fst value with the Chinese population (Fst = 0.117), and the highest value with the Vietnamese population (Fst = 0.564) (Table 4). A lower Fst value indicates less genetic differentiation between populations, suggesting a higher level of genetic similarity between the Indonesian and Chinese GarVA populations. In contrast, a higher Fst value signifies greater genetic differentiation, indicating that gene flow between populations is more restricted. A greater genetic distance between populations leads to stronger genetic separation, resulting in lower genetic similarity among populations.

Indonesian GarVB population showed the lowest Fst value when compared with the Japanese population (Table 5). Likewise, the Japanese GarVB population had its lowest Fst value with the Chinese population. These results suggest that the Indonesian GarVB population may have originated from a genetic source similar to the Chinese GarVB population, which also shared genetic similarity with the Japanese population. This is further supported by the Fst value between Indonesia and China, which is categorized as low, indicating limited genetic differentiation (Fst = 0.311). The highest Fst value was observed between the Indonesian and Korean GarVB populations (Fst = 0.857), indicating a high level of genetic differentiation between these populations.

Population structure of Indonesian GarVE exhibited its lowest Fst value in comparison with the Chinese population (Fst = 0.357) (Table 6), although this value is classified within the moderate range. This suggests that the Indonesian GarVE population can be considered to have differentiated from the GarVE from China. In contrast, the highest level of genetic differentiation was observed between the Indonesian and Australian populations (Fst = 0.618), indicating limited gene flow and substantial genetic separation between these two populations. Furthermore, the fixation index between

Table 4. Population structure analysis (Fst) of *Garlic virus* A

Location	Indonesia	China	Australia	Poland	Japan	Vietnam
Indonesia[a]	*	*	*	*	*	*
China	0.117	*	*	*	*	*
Australia	0.162	0.074	*	*	*	*
Poland	0.431	0.490	0.312	*	*	*
Japan	0.448	0.502	0.316	0.030	*	*
Vietnam	0.564	0.493	0.450	0.860	0.866	*

*means identical, [a]samples were originally from China

Table 5. Population structure analysis (Fst) of *Garlic virus* B

Location	Indonesia	Czech	Poland	China	Japan	Spain	Korea
Indonesia[a]	*	*	*	*	*	*	
Czech	0.406	*	*	*	*	*	
Poland	0.559	0.298	*	*	*	*	
China	0.311	0.165	0.238	*	*	*	
Japan	0.282	0.118	0.386	0.116	*	*	
Spain	0.818	0.711	0.639	0.647	0.714	*	
Korea	0.857	0.526	0.486	0.452	0.588	0.857	*

*means identical, [a]samples were originally from China

Table 6. Population structure analysis (Fst) of *Garlic virus* E

Location	Indonesia	China	USA	Japan	Australia	India
Indonesia[a]	*	*	*	*	*	*
China	0.357	*	*	*	*	*
USA	0.608	0.519	*	*	*	*
Japan	0.608	0.519	0.000	*	*	*
Australia	0.618	0.454	0.918	0.918	*	*
India	0.543	0.427	0.682	0.682	0.687	*

*means identical, [a]samples were originally from China

the GarVE populations from the United States and Japan was zero, implying an absence of genetic differentiation and indicating that the viral genomes among these populations are genetically identical or highly similar.

The haplotype network illustrates relationships based on genetic similarity distances among populations. The haplotype network of GarVA (Figure 8) reveals three distinct groups, separated by hatch marks that represent mutational steps between groups. This network suggests that the central haplotype group serves as the origin from which mutations leading to other groups have emerged. The haplotype positioned at the centre of the network can be inferred as the ancestral haplotype, from which the surrounding derived haplotypes have evolved. Indonesian haplotypes are distributed across two haplotype clusters; two haplotypes of GarVA_INA are located on a branch derived from the Chinese haplotype, while the other haplotype of GarVA_INA is positioned on a separate branch originating from a Chinese haplotype that shares similarity with an Australian haplotype.

The haplotype network of GarVB (Figure 9) shows a centralized pattern with few mutational steps, as indicated by the small number of hatch marks. This suggests a close genetic relationship among the haplotypes. The GarVB_INA haplotypes are located on nearby branches and are derived from a Chinese haplotype that also shares similarities with a Japanese haplotype (GarVB_CHN_JPN inside the bracket). The haplotype network of GarVE (Figure 10) shows the absence of a central haplotype, indicating that there is no dominant haplotype that serves as the common ancestor of the others. The haplotypes of GarVE_INA are positioned on closely related branches and exhibited a close genetic relationship to a haplotype shared by isolates from China, the USA, and Japan, suggesting a common ancestral origin.

4. Discussion

This study reported the first specific molecular detection of three allexiviruses (GarVA, GarVB, and GarVE) from imported garlic (consumption bulbs) in

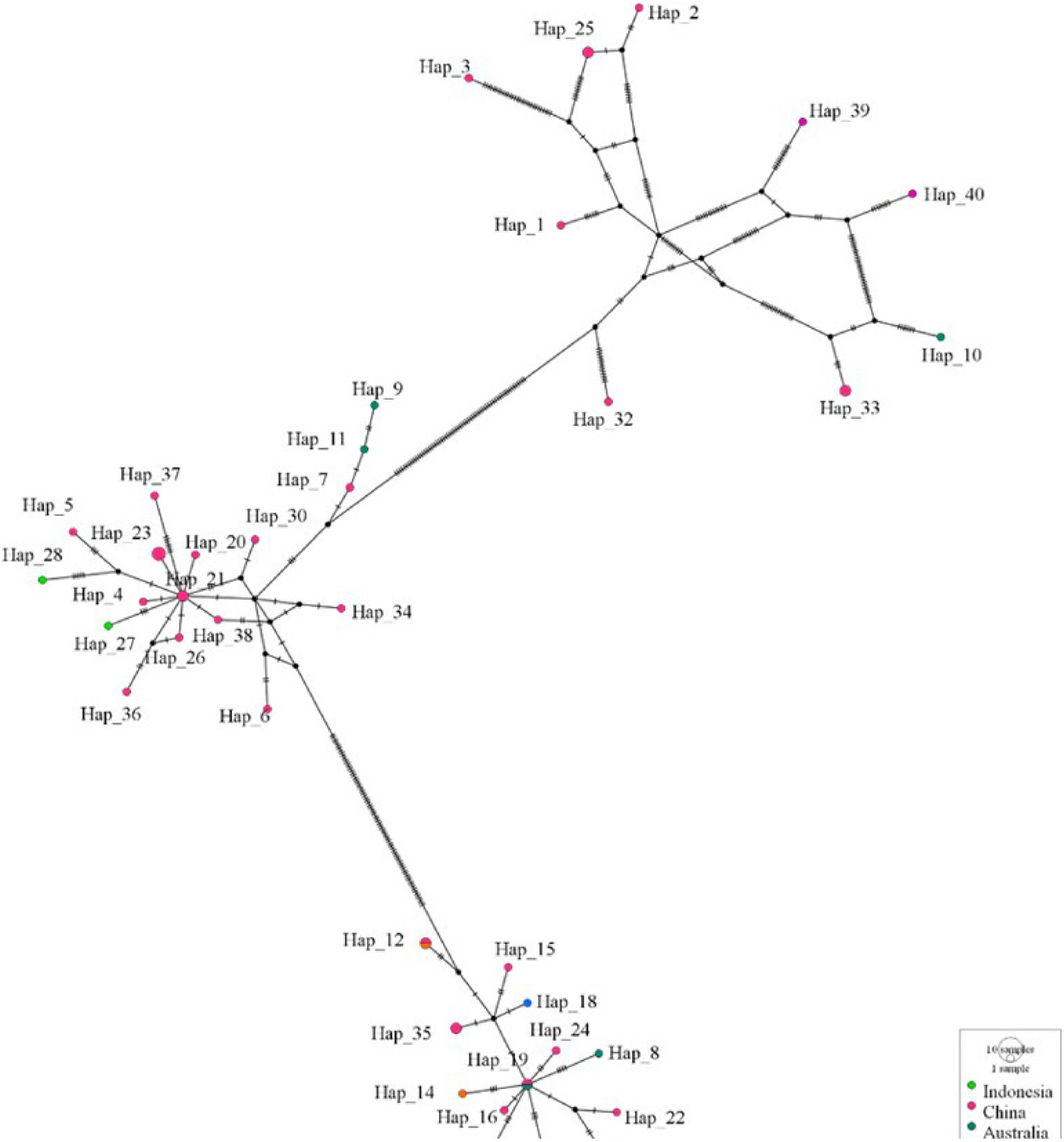

Figure 8. Haplotype network of Garlic virus A. The size of circle represents the sample size in the haplotype. Each hatch mark indicates the number of mutations. The samples of GarVA_INA were originally from China

Indonesia. The results of this study, particularly the optimization of RT-PCR conditions, will be used for further detection of local garlic cultivars, as farmers use them for planting material. To date, detection of viruses from various Indonesian garlic cultivars and imported bulbs has been done for poty- and carlaviruses (Hidayat *et al.* 2023; Nurulita *et al.* 2024). Further detection of allexiviruses will be projected on completion of the basis data of the main viruses infecting Indonesian garlic cultivars.

Typical symptoms of virus infection were present in all germinated bulbs, and specific RT-PCR confirmed virus presence. Allexivirus infection typically induces only mild symptoms in natural infection, such as

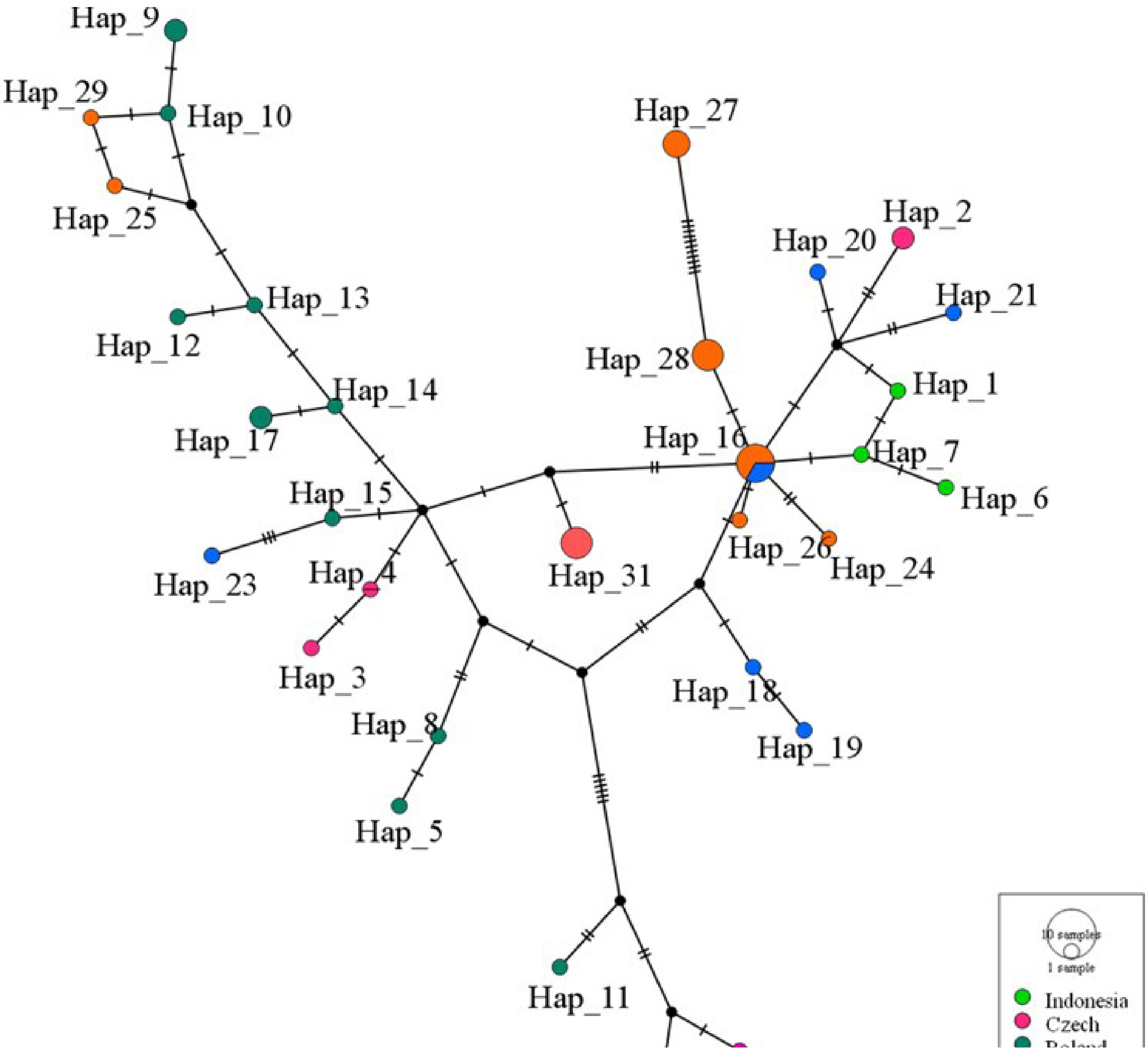

Figure 9. Haplotype network of *Garlic virus* B. The size of circle represents the sample size in the haplotype. Each hatch mark indicates the number of mutations. The samples of GarVA_INA were originally from China

mosaic patterns, leaf striping, and stunted growth (Kang *et al.* 2007; Koczor *et al.* 2024). Single infection of allexivirus in plants is rarely observed, making it difficult to distinguish the symptoms of these viruses specifically. Cafrune *et al.* (2006) reported that single infections of GarVC or GarVA led to reductions in bulb weight of up to 15% and bulb diameter by approximately 5%. Furthermore, latent infection of allexivirus is commonly found in garlic; therefore, it is possible to find viruses on both symptomatic and asymptomatic plants Abraham *et al.* (2019). Although allexiviruses commonly induce latent infections, coinfections with potyviruses, such as LYSV and OYDV, have a synergistic effect resulting in significant yield losses and bulb quality reduction (Perotto *et al.* 2010; Abraham *et al.* 2019).

Detection of allexiviruses in imported garlic indicates that the virus can be carried through vegetative plant materials originating from infected sources. The spread of allexivirus is also facilitated by its mite vector (Mansouri *et al.* 2021). This mite can act as a vector during both the vegetative growth phase and storage periods, making virus transmission possible during storage or transport of imported planting materials. Therefore, phytosanitary measures are crucial for regulating the importation of vegetative

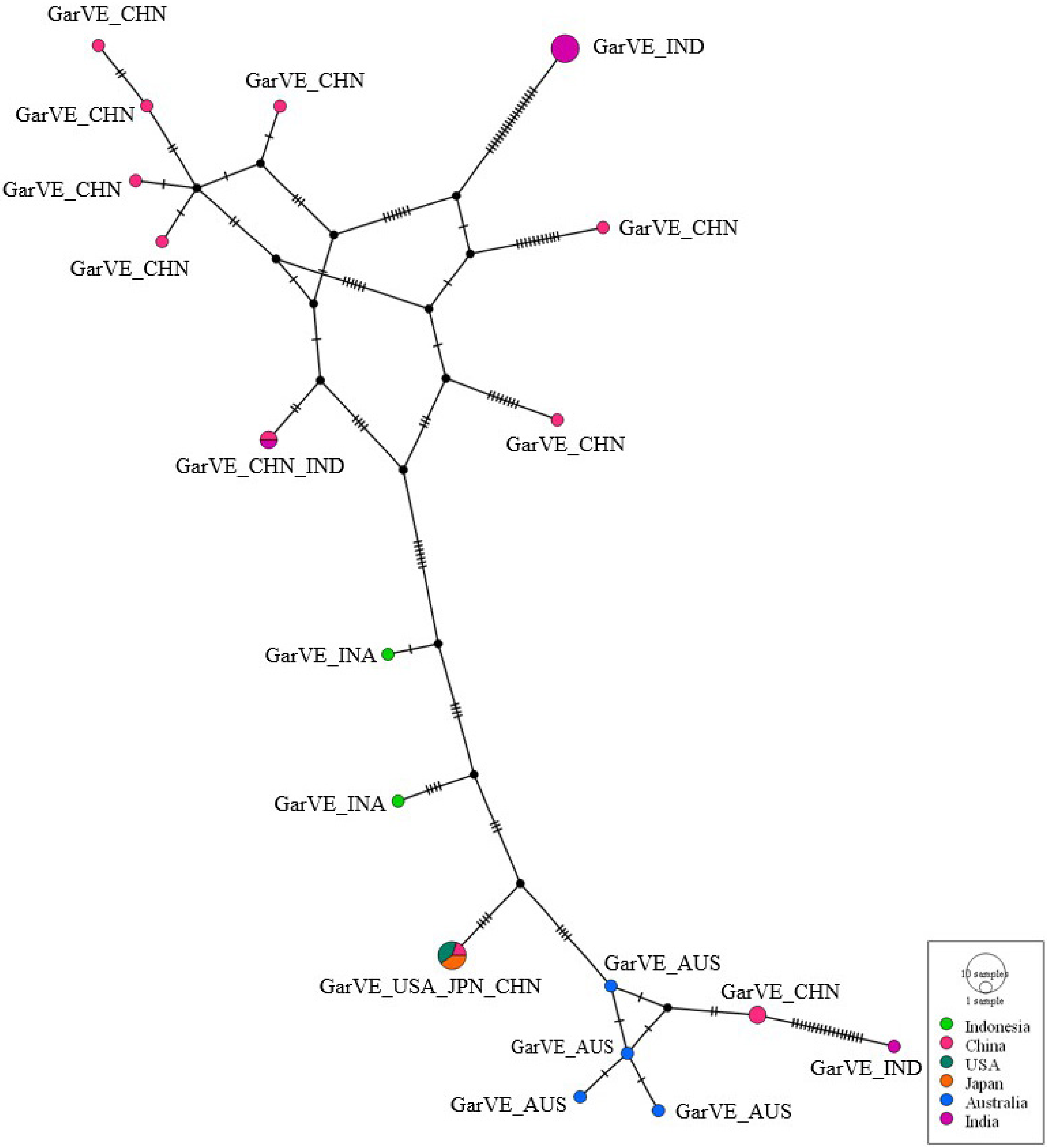

Figure 10. Haplotype network of *Garlic virus* E. The size of circle represents the sample size in the haplotype. Each hatch mark indicates the number of mutations. The samples of GarVA_INA were originally from China

plant materials. In this case, the Indonesian Quarantine Agency (IQA) should classify the quarantine status of both allexivirus and their vectors, *A. tulipae*.

Population genetics analysis, particularly for GarVA and GarVB, revealed a close genetic relationship between Indonesian and Chinese allexivirus isolates. These findings highlighted the crucial role of the Chinese haplotype in shaping the overall genetic structure and functioning as central nodes in genetic evolution (García-Arenal *et al.* 2001). According to Makyorukty *et al.* (2025), close genetic relationships among populations may be attributed to international trade, particularly involving garlic as the host plant of the virus, which facilitates the introduction of viral isolates through the importation of infected bulbs from source countries. This is relevant considering that the garlic bulbs analyzed in this study were imported from China, as the world's largest garlic exporter.

Variability in the genetic structure of virus populations has a significant role as the initial step in adaptation to new plants (host shifting), resistance breaking, and changes in symptoms and virulence (Acosta-leal *et al.* 2011). Allexivirus has been reported to expand its host range; for example, GarVD has been shown to infect plants outside the Allium genus, including species from dicotyledonous groups (Fidan *et al.* 2013). This demonstrates that in the case of a high disease epidemic, the virus has the potential to adapt to new hosts and evolve toward higher virulence. Therefore, genetic studies of allexivirus, as well as its detection in local garlic varieties, are necessary as preventive measures to mitigate yield loss and crop damage caused by allexivirus, particularly in local garlic production centers.

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