Genome Comparison of Aggressive and Non Agressive Geminivirus Strain from West Sumatera

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Introduction

- Economic role of Chilli as vegetable and spices
- Price fluctuation contribute to 2nd factor in national inflation rate
- Price fluctuation is caused by PepYLCD

Introduction

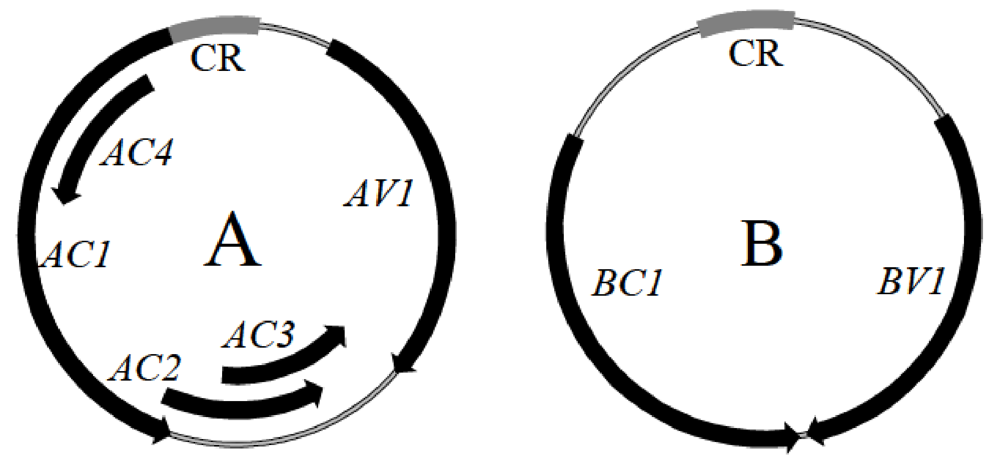
- PepYLCD caused by Gemini virus
- Decrease can reach up to 100%
- No effective control so far.
- High genetic variation.

Geminivirus genome

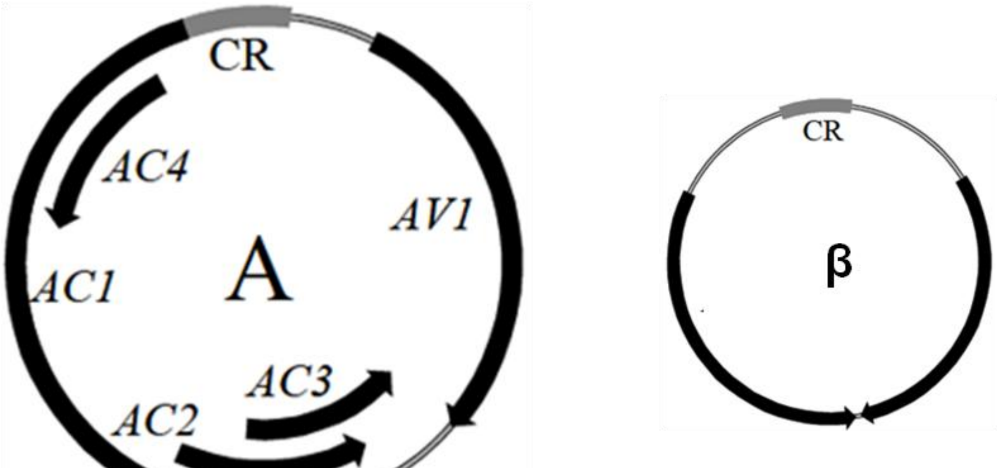
- GV is ssDNA virus.
- Belongs to genus of begomovirus
- Most have bipartite, genome characterized by Genom A and B-2,7 kb.
- Some are monopartite, have only A-like genome and β-satellite.

Gemini virus genome

Bipartite



Monopartite



Rojas, (2004)

Geminivirus genes and function

Monopartite	Bipartite	Protein and function
V1	AV1	*Movement protein* (MP), movement in the infected plant.
V2	AV2	*Coat protein,* spreading, and movement in the host, protection in the digestion system of the vector. (Whitefly).
C1	AC1	*Replication-associated protein* (Rep), virus replication.
C2	AC2	*Transcriptional activator protein* (TrAP), activating of promotor transcription of CP.
C3	AC3	*Replication enhancer protein* (REn), interact with C1-protein for virus particle accumulation.
C4	AC4	interacct with C1 and MP in symptom determination, cell division initiation, cell to cell movement, breaking plant defence system.
-	BV1	*Nuclear shuttle protein* (NSP), encode DNA B.
-	BC1	*Movement protein* (MP), virus movement in the infected plant.

Meliansyah, 2010

Objectives

Short term:

Understanding of relationship between genome variation and the aggressivity.

Long term:

Development of broad spectrum resistancy of chilli cultivar against gemini virus.

Material Source

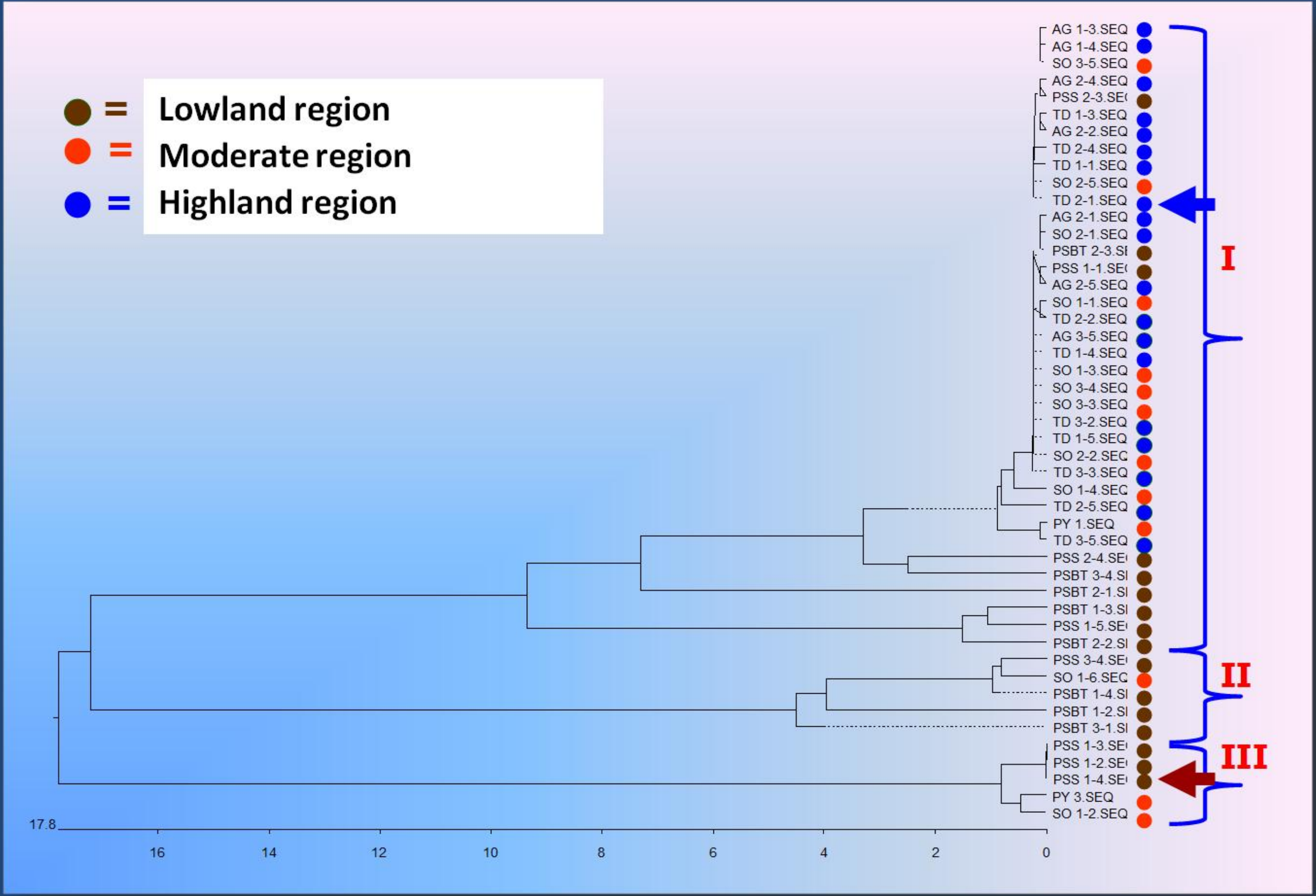
TD -21-Isolate

PSS-14-Isolate

Strain grouping

48 isolates

Primer:
PAL1v1978
PAR1c715



Agressivity Testing

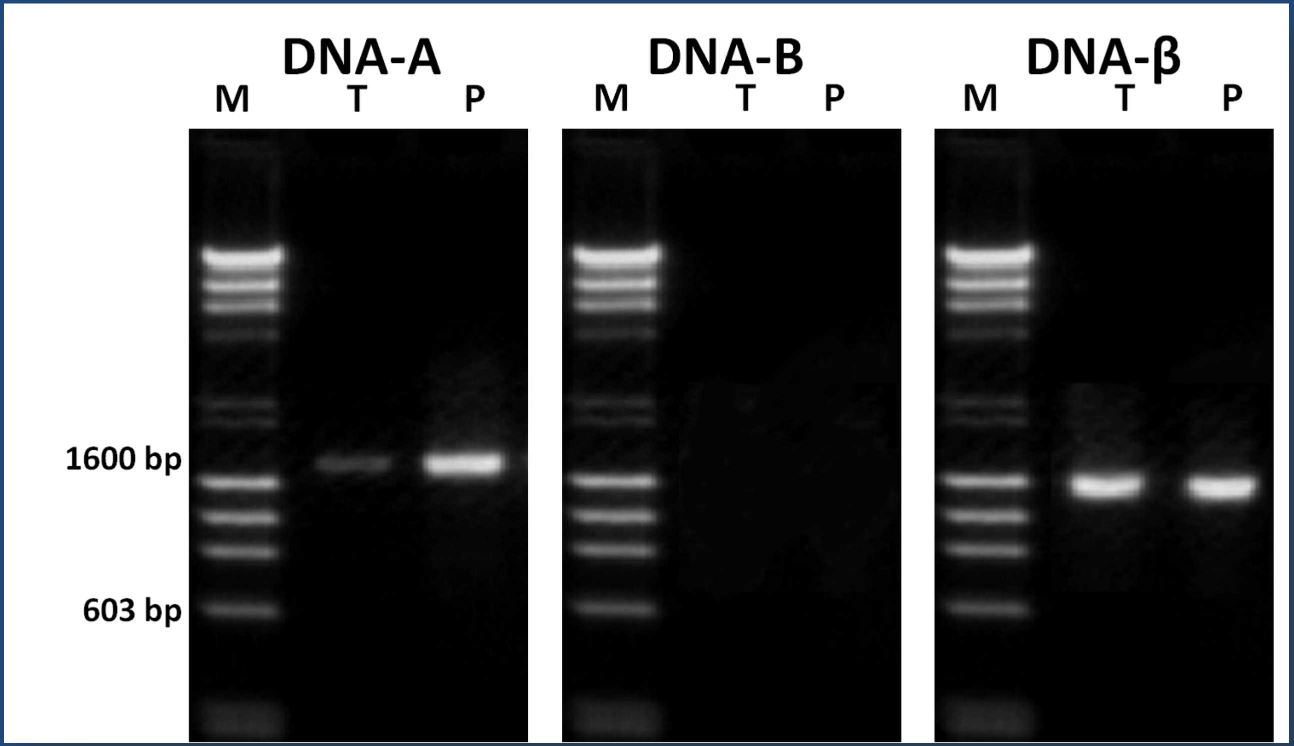
8 days after inoculation

TD -21-Isolate
(non aggressive)

PSS-14-Isolate
(agressive)

TD21 and PSS14 have monopartite genome

PAR1c715/PAL1v1978 prBV1855/prBC656 Beta 01/02

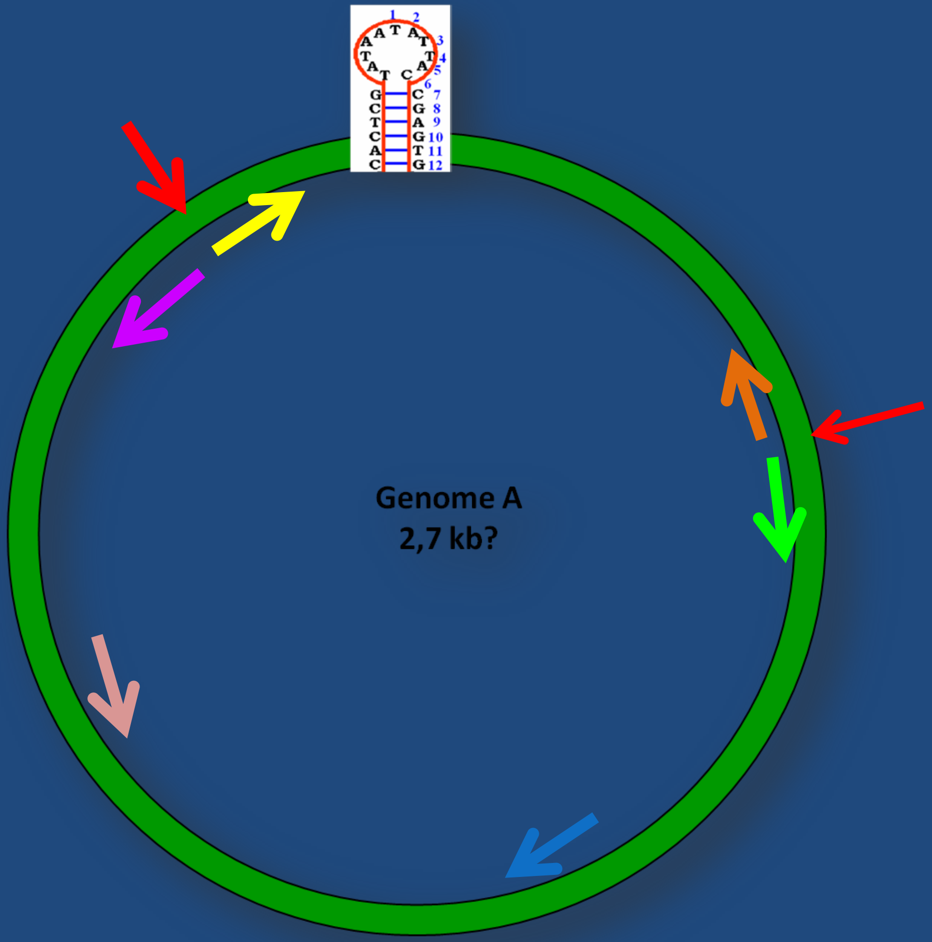


M = 1 kb ladder
T = TD21
P = PSS14

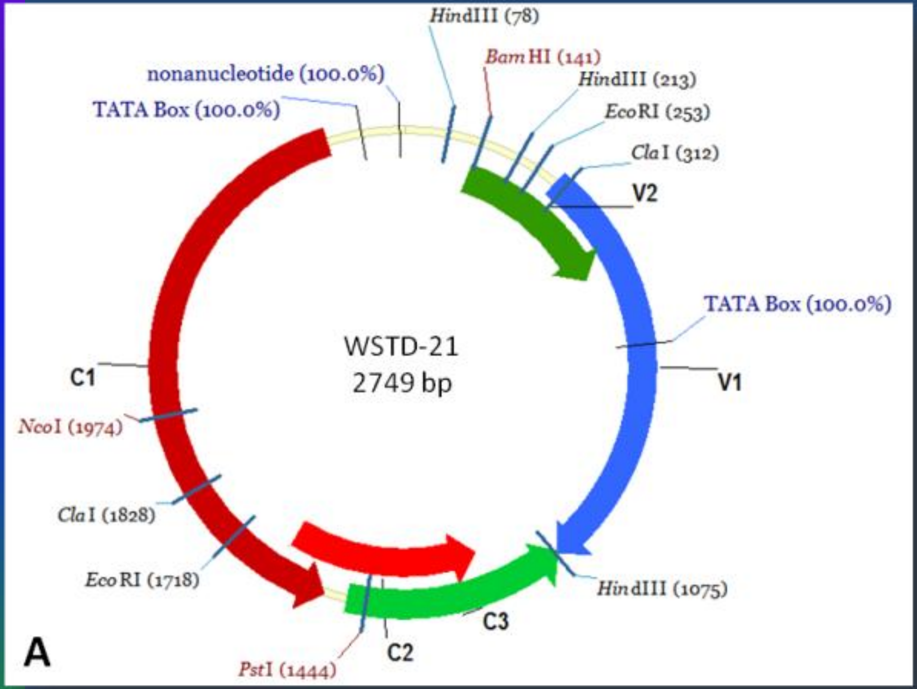
Primer walking for whole genome sequencing.

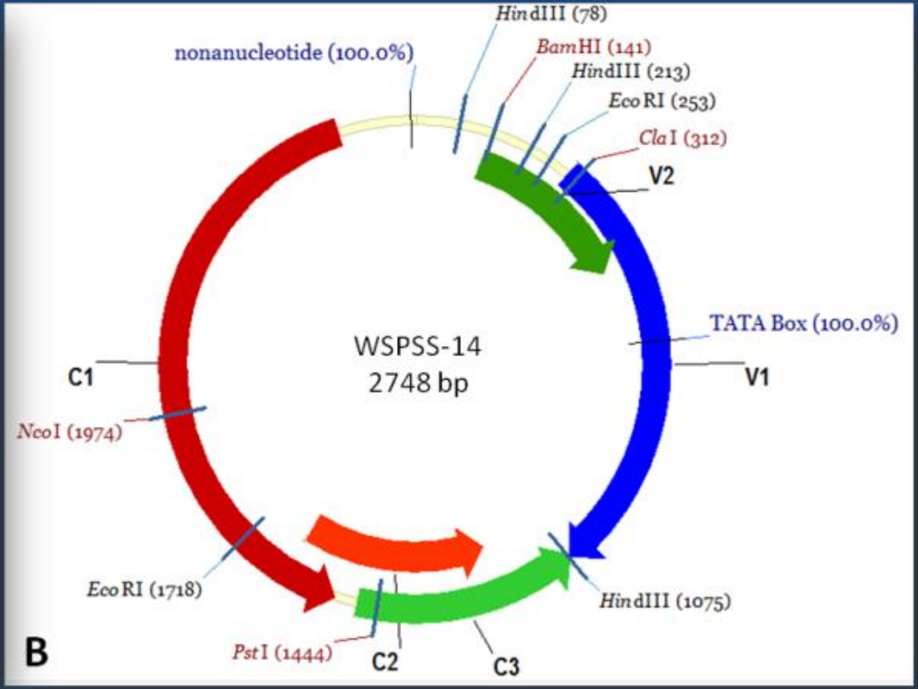
Primer:

PAL1v1978 / PAR1c715



Genome A of TD-21 and PSS-14 isolate





TD-21 ; 2749 bs

PSS-14; 2748 bs

Genome A size of TD-21 and PSS-14 isolates

TD21 : 2749
PSS14: 2748

Gene/ORF	TD21	PSS14
V1 (sense)	135-488	135-488
V2 (sense)	295-1071	295-1071
C1 (non sense)	1520-2608	1520-2608
C2 (non sense)	1213-1630	1213-1630
C3 (non sense)	1068-1478	1068-1478

1 InDel event in Common region (CR)

Movement protein (MP) gene/V1 is conserved

16 points;
96% similarity

```
PSS14-V1        ATGTGGATCCGCTTGTGCATCCGTTTCCAGAAACCCTACATGGATTTCGATGCATGTTG 60
TD21-V1         ATGTGGATCCGCTTATTCATCCTTTTCCTGAAACCCTACACGGGTTTCGATGCATGTTG 60
                ************** * ***** ***** *********** ** ****************

PSS14-V1        GCGATCAAGTATCTGCAAAGCTTAGAGGCTACGTATTCTCCTGATACGGTTGGAGGTGAA 120
TD21-V1         GCTATCAAGTATCTGCAAAGCTTAGAGGCTACGTATTCTCCTGATACGGTTGGAGGTGAA 120
                ** *********************************************************

PSS14-V1        TTCGTGAAGGATTTGATCTGTTTGCTGCGGTGTAAAAACTATGCCGAAGCGTTCCATCGA 180
TD21-V1         TTCGTGAGGGATTTAATTTGTCTGTTGCGGTGTAAAAATTATGCCGAAGCGTTCCATCGA 180
                ******* ****** ** *** ** ************* *********************

PSS14-V1        TACAGTGTCGTCGTTGCCAATGTCTATAACACGCCGGAGACTAAACTACGCGAGTCAGTA 240
TD21-V1         TACAGTGTCGTCGTTGCCAATGTCTATAACACGCCGGAGACTAAACTACGCGAGTCAGTA 240
                ************************************************************

PSS14-V1        CAGTCTCCCTGCTGCTGCCCCCACTGCCCCAGGCATGTCTTACAAACGAAGAGCGTGGGT 300
TD21-V1         CAGTCTCCCTGCTGCTGCCCCCACTGCCCCAGGCATGTCGTACAAACGAAGAGCATGGGT 300
                *************************************** ************** *****

PSS14-V1        AAATCGGCCTATGAATCGGAAACCCAGATTCTACAGGGGTCGAAGGACCAGTGA 354
TD21-V1         AAATCGGCCTATGAATCGGAAACCCAGATTCTACAGGGGTCGAAGGAGCAGTGA 354
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```
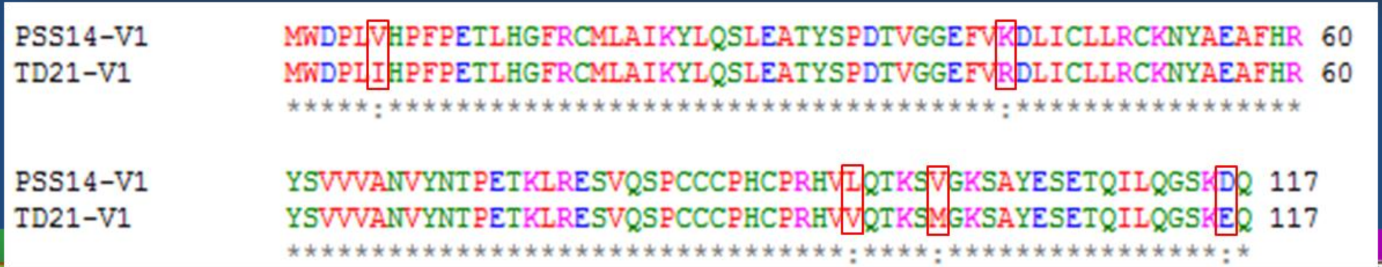

5 points
96% similarity



```
PSS14-V1        MWDPLVHPFPETLHGFRCMLAIKYLQSLEATYSPDTVGGEFVKDLICLLRCKNYAEAFHR 60
TD21-V1         MWDPLIHPFPETLHGFRCMLAIKYLQSLEATYSPDTVGGEFVRDLICLLRCKNYAEAFHR 60
                *****:************************************:*****************

PSS14-V1        YSVVVANVYNTPETKLRESVQSPCCCPHCPRHVLQTKSVGKSAYESETQILQGSKDQ 117
TD21-V1         YSVVVANVYNTPETKLRESVQSPCCCPHCPRHVVQTKSMGKSAYESETQILQGSKEQ 117
                *********************************:****:****************:*
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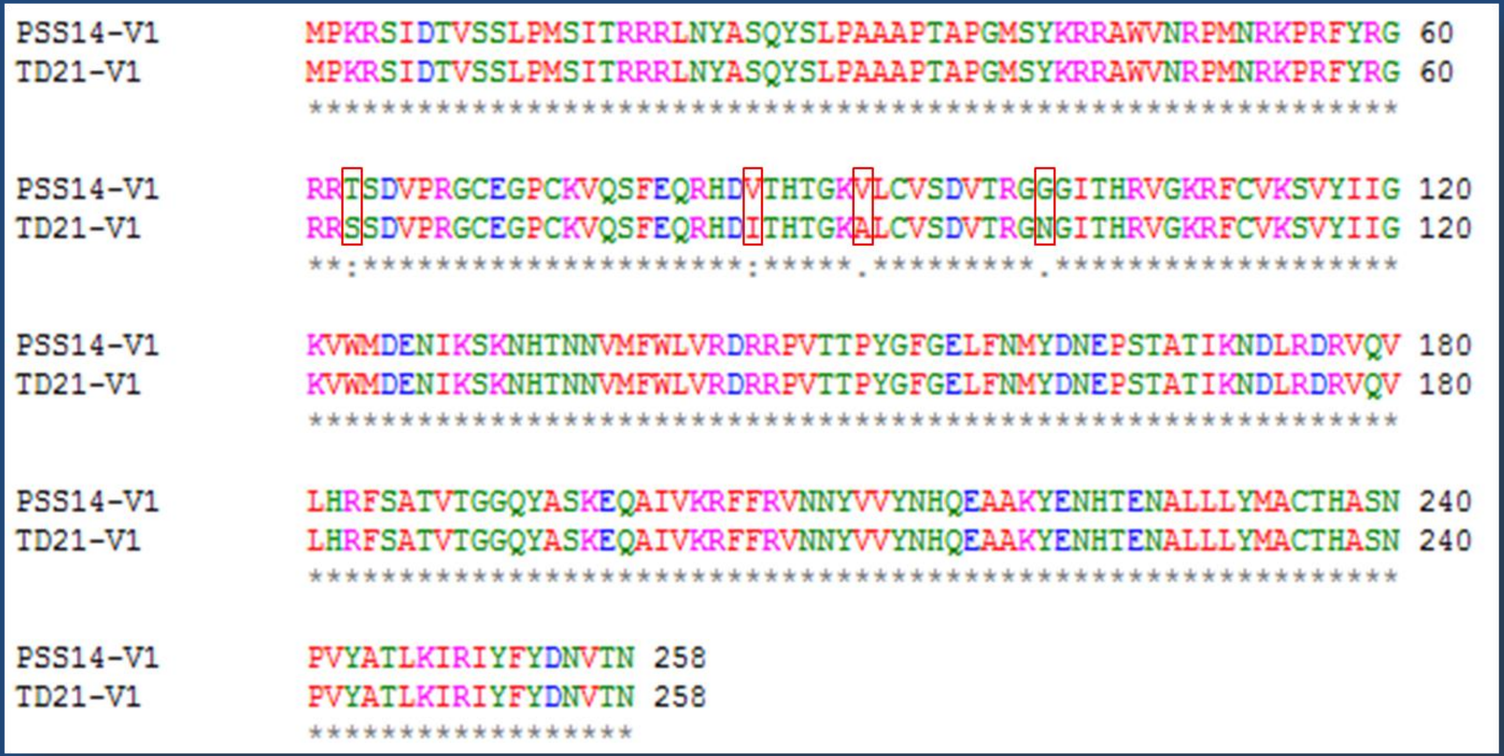
Coat protein (CP) /V2 gene is conserved



```
PSS14-V1        MPKRSIDTVSSLPMSITRRRLNYASQYSLPAAAPTAPGMSYKRRAWVNRPMNRKPRFYRG 60
TD21-V1         MPKRSIDTVSSLPMSITRRRLNYASQYSLPAAAPTAPGMSYKRRAWVNRPMNRKPRFYRG 60
                ************************************************************

PSS14-V1        RRTSDVPRGCEGPCKVQSFEQRHDVTHTGKVLCVSDVTRGGGITHRVGKRFCVKSVYIIG 120
TD21-V1         RRSSDVPRGCEGPCKVQSFEQRHDITHTGKALCVSDVTRGNGITHRVGKRFCVKSVYIIG 120
                **:*********************:*****.*********.*******************

PSS14-V1        KVWMDENIKSKNHTNNVMFWLVRDRRPVTTPYGFGELFNMYDNEPSTATIKNDLRDRVQV 180
TD21-V1         KVWMDENIKSKNHTNNVMFWLVRDRRPVTTPYGFGELFNMYDNEPSTATIKNDLRDRVQV 180
                ************************************************************

PSS14-V1        LHRFSATVTGGQYASKEQAIVKRFFRVNNYVVYNHQEAAKYENHTENALLLYMACTHASN 240
TD21-V1         LHRFSATVTGGQYASKEQAIVKRFFRVNNYVVYNHQEAAKYENHTENALLLYMACTHASN 240
                ************************************************************

PSS14-V1        PVYATLKIRIYFYDNVTN 258
TD21-V1         PVYATLKIRIYFYDNVTN 258
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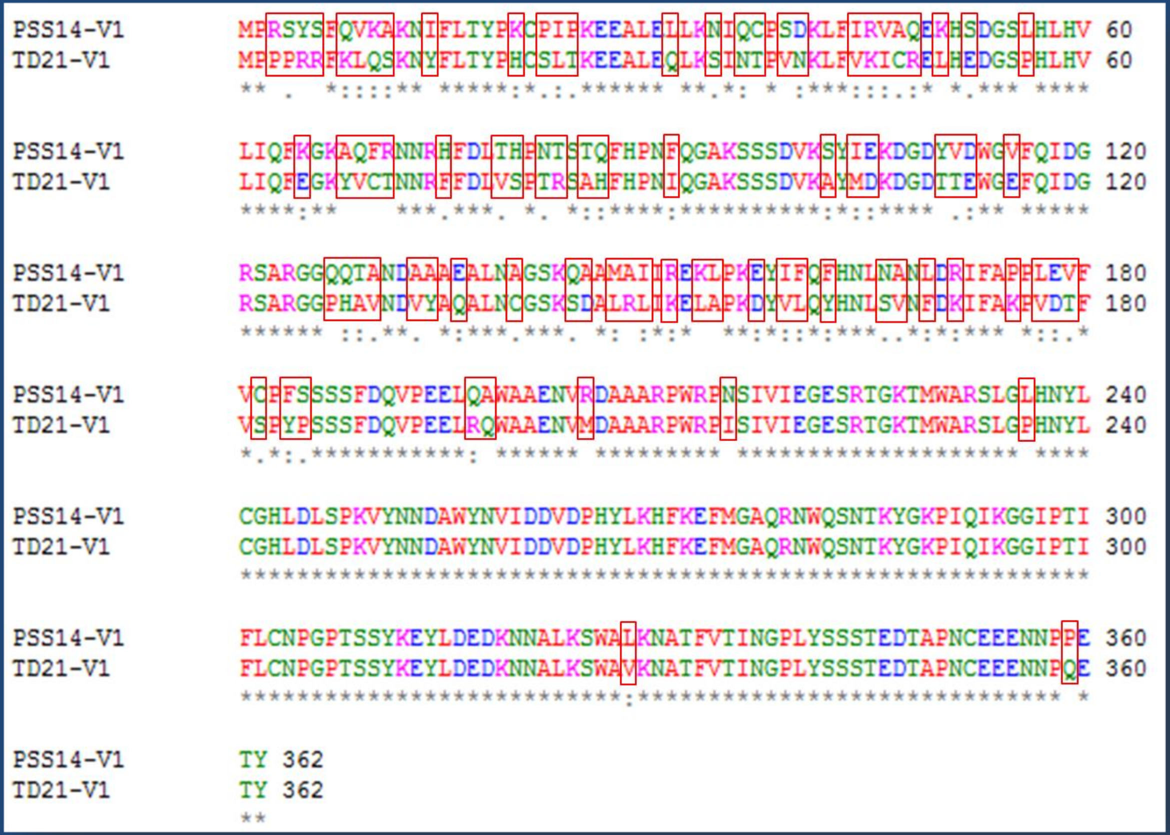
```

4 points; Similarity: 98,5%

Rep (C1) gene showed high level of polymorphims

Similarity: 77%



```
PSS14-V1        MPRSYSFQVKAKNIFLTYPKCPIPKEEALEILKNIQCPSDKLFIRVAQEKHSDGSLHLHV 60
TD21-V1         MPPPRRFKLQSKNYFLTYPHCSLTKEEALEQLKSINTPVNKLFVKICRELHEDGSPHLHV 60
                ** .  *::::** *****:*.:.****** **.*: * :***:::.:* *.*** ****

PSS14-V1        LIQFKGKAQFRNNRHFDLTHPNTSTQFHPNFQGAKSSSDVKSYIEKDGDYVDWGVFQIDG 120
TD21-V1         LIQFEGKYVCTNNRFFDLVSPTRSAHFHPNIQGAKSSSDVKAYMDKDGDTTEWGEFQIDG 120
                ****:**    ***.***. *. *::****:**********:*::**** .:** *****

PSS14-V1        RSARGGQQTANDAAAEALNAGSKQAAMAIIREKLPKEYIFQFHNLNANLDRIFAPPLEVF 180
TD21-V1         RSARGGPHAVNDVYAQALNCGSKSDALRLIKELAPKDYVLQYHNLSVNFDKIFAKPVDTF 180
                ****** ::.**. *:***.***. *: :*:*  **:*::*:***..*:*:*** *::.*

PSS14-V1        VCPFSSSSFDQVPEELQAWAAENVRDAAARPWRPNSIVIEGESRTGKTMWARSLGLHNYL 240
TD21-V1         VSPYPSSSFDQVPEELRQWAAENVMDAAARPWRPISIVIEGESRTGKTMWARSLGPHNYL 240
                *.*:.***********: ****** ********* ******************** ****

PSS14-V1        CGHLDLSPKVYNNDAWYNVIDDVDPHYLKHFKEFMGAQRNWQSNTKYGKPIQIKGGIPTI 300
TD21-V1         CGHLDLSPKVYNNDAWYNVIDDVDPHYLKHFKEFMGAQRNWQSNTKYGKPIQIKGGIPTI 300
                ************************************************************

PSS14-V1        FLCNPGPTSSYKEYLDEDKNNALKSWALKNATFVTINGPLYSSSTEDTAPNCEEENNPPE 360
TD21-V1         FLCNPGPTSSYKEYLDEDKNNALKSWAVKNATFVTINGPLYSSSTEDTAPNCEEENNPQE 360
                ***************************:****************************** *

PSS14-V1        TY 362
TD21-V1         TY 362
                **
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Big question: does C1 gene play important role in the agressivity of PepYLCV?

Conclusion

- Agressivity is not determined by genome type
- Both agressive and non agressive strain have similar gene (ORFs) components, which are almost similar except C1 (replicase gene)
- Seemed, that C1 (replicase gene) determine the aggressivity, however this assumption have to be proved by further functional analysis.

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Thanks a lot