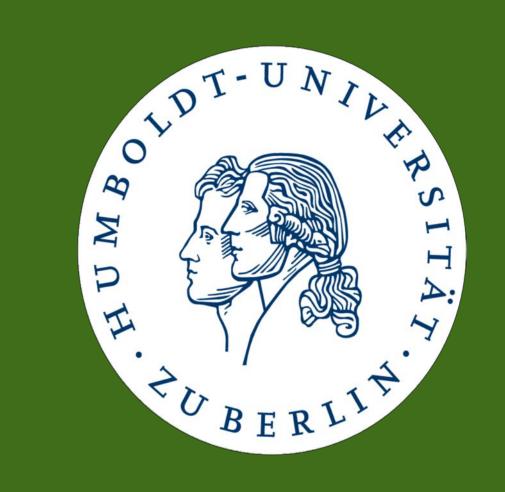
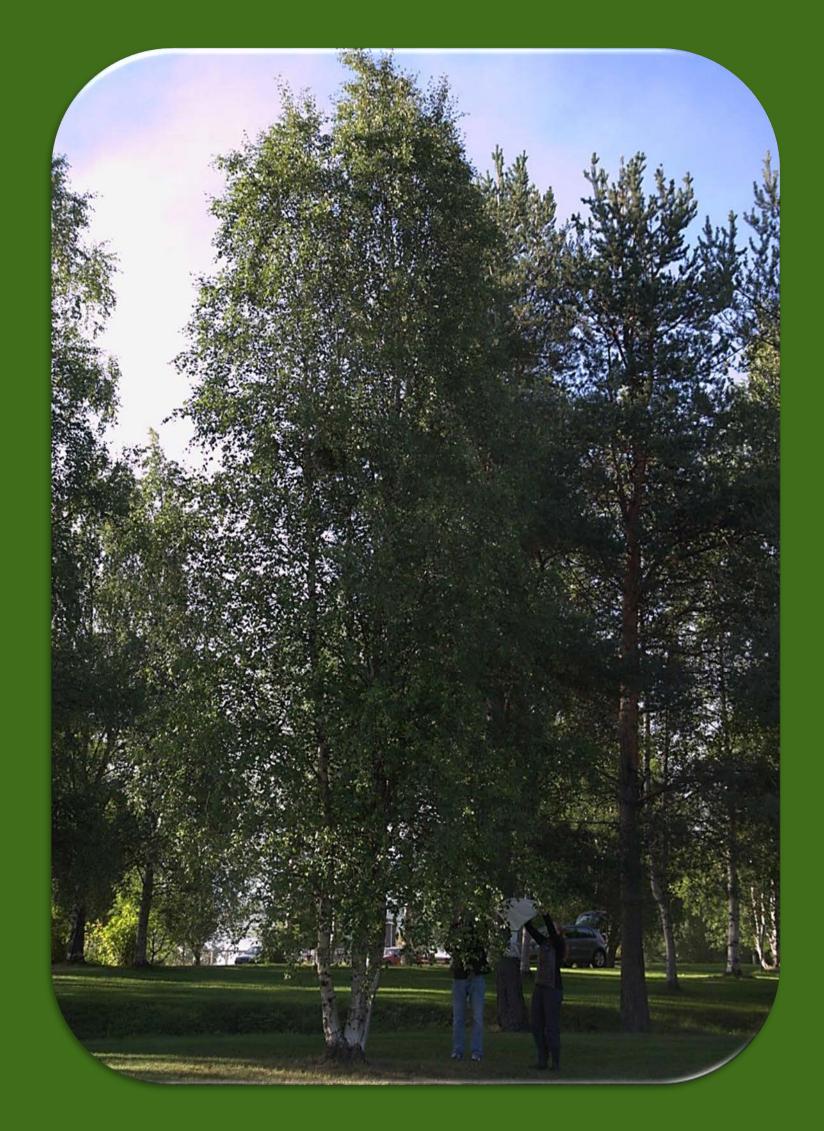
Cherry leaf roll virus in Betula spp. in Finland: what do we know about its population diversity?

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Characterization of diverse Cherry leaf roll virus (CLRV) isolates from birch (Betula spp.) has been of high interest as the virus is consistently detected in trees exhibiting strong symptoms in Finnish forests and urban areas (Fig. 1 & 2). Samples from 14 Betula pubescens trees growing in the urban parks of Rovaniemi, Finland were investigated for the presence of CLRV variants. RT-PCR fragments from three different genetic regions were cloned and sequenced in order to estimate the genotypic and genetic variability of the virus.



Fig.2 Chlorotic veinbanding and leafroll symptoms observed in CLRV-infected B. pubescens in the city of Rovaniemi, Finland.

Fig.1 One of the 14 CLRV-infected birches sampled in the summer 2012 from Rovaniemi.

Results

- Variants identified: 5-6 variants were identified in each genomic region.
- Variant frequency: Based on their abundance in the trees, the variants were clustered in three groups: a. one predominant variant that was present in the majority of the trees (red, frequency \geq 40%), b. a few variants common among 3-4 trees (yellow, frequency = 10-40%) and c. rare variants detected once (green, frequency ≤ 10%) (Fig. 3).
- Within tree virus variability: up to three CLRV variants were found in each individual birch.
- Genetic diversity among trees: up to 16% for the UTR region, up to 23% in the CP and RdRp regions (Fig. 4).

Genetic diversity	3'- UTR	СР	RdRp
Within Finnish samples - On nucleotide level - On aminoacid level	1,9 - 16%	7,2 - 23 % 10,5 - 16,6 %	2,7 - 23,5 % 1,1 - 11,2 %
Within total <i>Betula</i> strains Among all CLRV strains in NCBI	0,9 - 16 % 0,3 - 17 %		

GENOME ORGANIZATION OF CLRV AND VIRUS VARIANTS IDENTIFIED IN THREE GENOMIC REGIONS

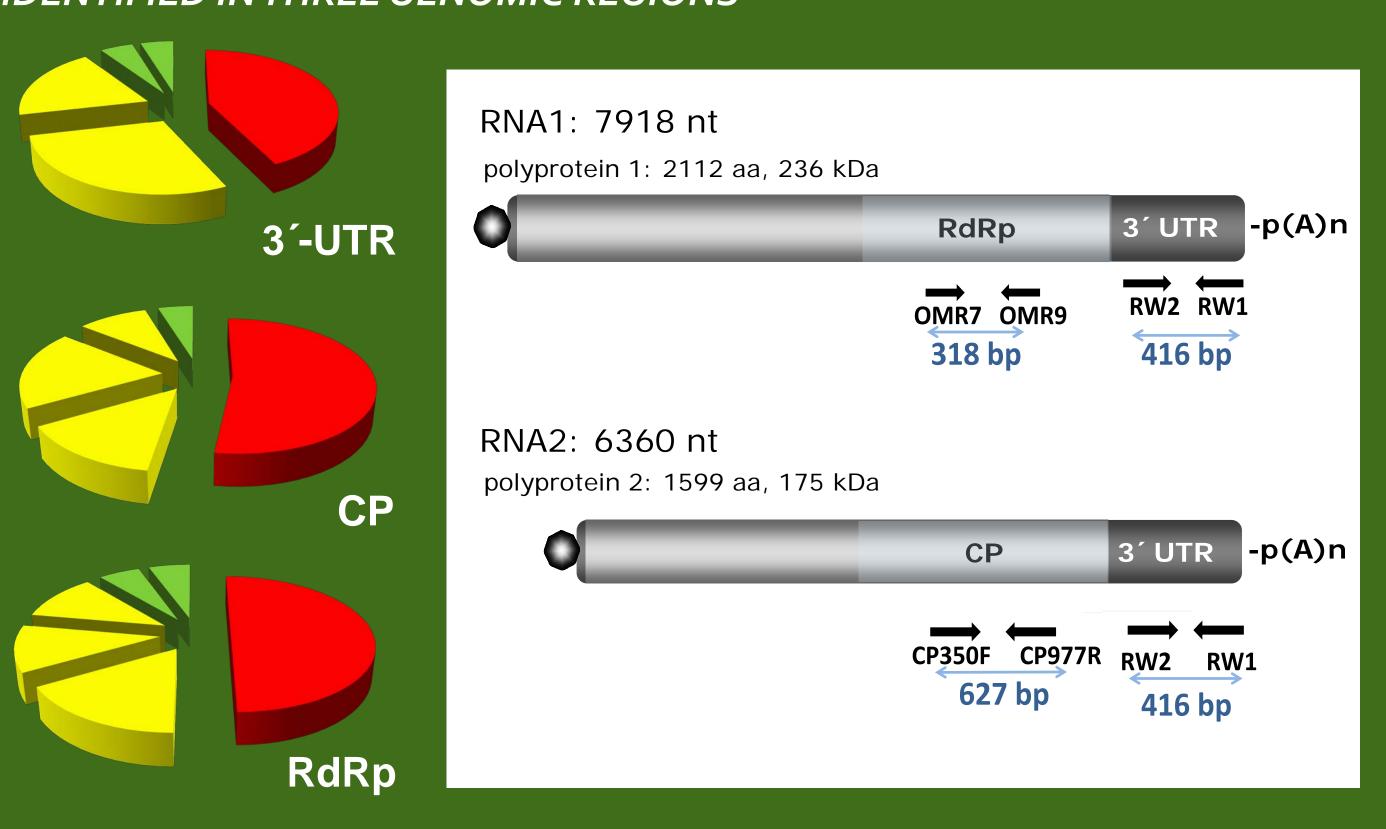


Fig.3 Variant distribution in each genome region with the variants clustered in three groups (predominant variant: red; common variants: yellow; rare variants: green) and display of the three partially sequenced genetic regions (3´- UTR: untranslated, CP: coat protein, RdRp: RNA-dependent RNA polymerase) in the two RNA genome segments of CLRV.

Fig.4 Genetic diversity based on nucleotide and/or aminoacid sequence data estimated in parts of the three genomic regions analyzed from 14 Finnish birches.

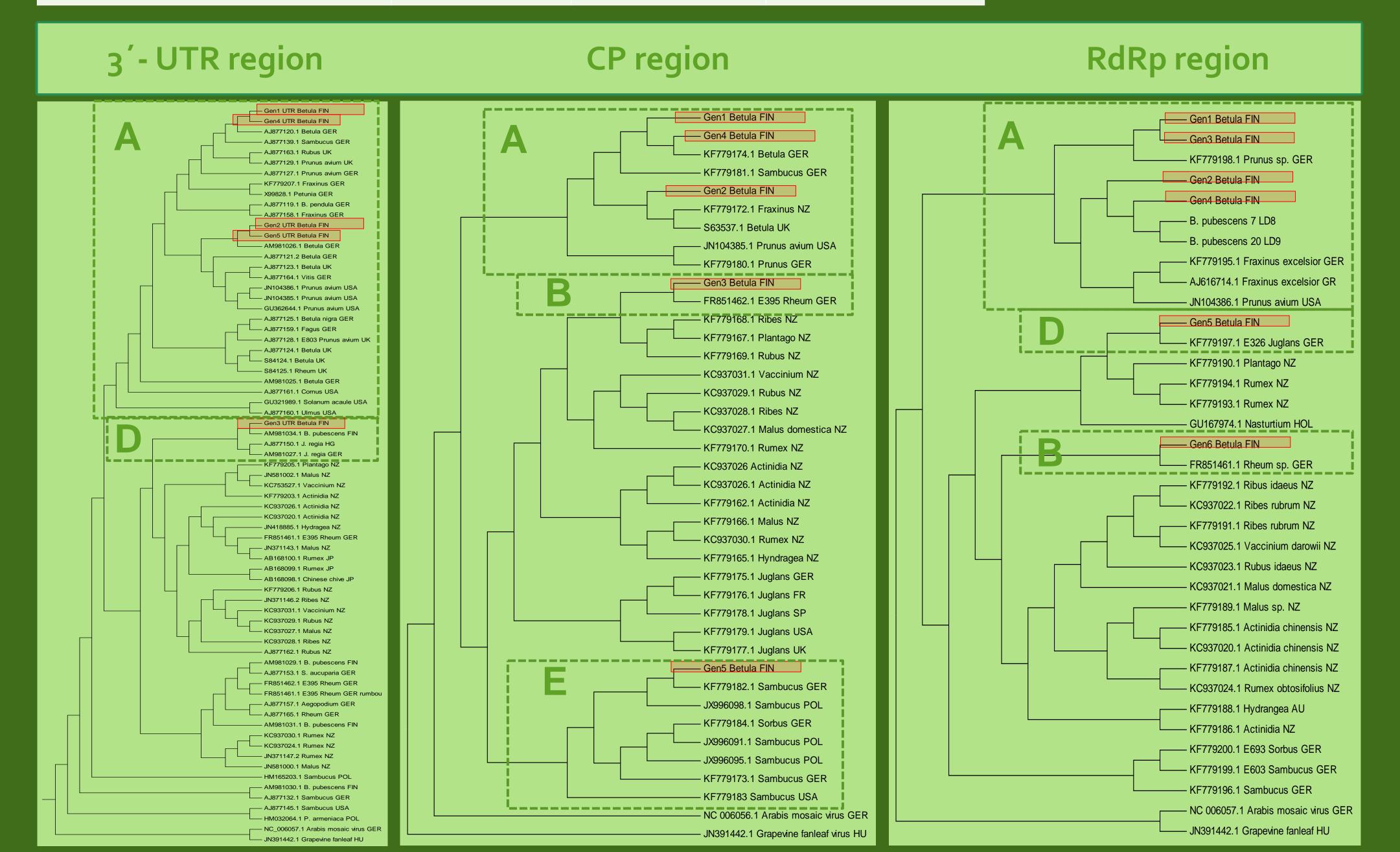


Fig.5 Phylogenetic trees generated by MEGA 6.06 applying the Maximum Likelihood method based on the Tamura-Nei model. Bootstrap analysis was performed with 1000 repetitions. Phylogenetic clusters (A to E) as established by Rebenstorf et al. (2006) by analysis of the 3 UTR fragments of CLRV are indicated in striped line-framed boxes. Samples derived from Rovaniemi are in red boxes.

Conclusions

- Each individual birch sampled was mixed-infected variants.
- High diversity of variants is found in Betula species in the urban parks from Rovaniemi, Finland.
- Although the analyzed CLRV population is restricted in sample size and geographic distribution, identified variants highly genetically variable belonging to 4 out of the 5 phylogenetic groups defined to date.
- CLRV variability in individual trees is expected to get higher when more samples of individual birches are included in the analysis.

Literature

REBENSTORF K., CANDRESSE T., DULUCQ M.J., BÜTTNER C. & OBERMEIER C., 2006. Host species-dependent population structure of a pollen-borne plant virus, Cherry leaf roll virus. Journal of Virology, **80**, p. 2453–2462.

Acknowledgements

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