The next generation of steps towards a phylogeny of Veronica subg. Pseudolysimachium

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Veronica subg. Pseudolysimachium is well-differentiated from other Veronica by both morphological and genomic characteristics. However, the phylogeny within the subgenus is still not completely resolved. We extracted DNA from 74 samples and used the Genotyping-by-sequencing (GBS) approach to infer the relationships within the subgenus. We merged overlapping paired-end reads and assembled the data with the assembly pipeline ipyrad to generate datasets for Maximum Likelihood (RAXML) analyses. In addition, we performed STRUCTURE analyses in order to delimit genotypically informative clusters. We obtained the following results: The earliest-branching clade consists of Eastern Asian samples, which form a well-delimited, yet unresolved group. The samples belonging to the subsection Pinnata form a well-supported clade. The remainder of the collections constitute two clades, one of them including samples belonging to the subsections Longifolia and Bachofenii, the other including collections belonging to the subsections Pseudolysimachium and Orchidae. The upper nodes show the limitations of species delimitation based on GBS data alone, especially among V. spicata, V. incana and V. barrelieri. We will discuss possible further steps to resolve the phylogeny of the subgenus.

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Two becomes one?: Genotyping-by-sequencing elucidates position of Ukrainian taxa within *Veronica* subg. *Pseudolysimachium*

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Species within *Veronica* subg. *Pseudolysimachium* have received divergent taxonomic treatments as species boundaries are blurred by frequent hybridization between groups. Here, we attempt to resolve the placement of Ukrainian *V. steppacea*, a taxon that has alternatively been placed within the more widespread *V. barrelieri*. To disentangle relationships within this complicated group, we analyzed GBS data from 130 individuals from Ukraine and adjacent territories. The data reveal no separation between *V. steppacea* and *V. barrelieri*. Albeit *V. incana* and *V. orchidea* possess distinctive morphologies, samples of *V. incana* group within *V. spicata*. Equally, *V. orchidea* groups with *V. spicata*, but is defined by a distinct admixture pattern in a structure analysis. Congruent with previous findings, *V. pinnata*, *V. spuria*, *V. longifolia* and *V. spicata* form separate groups, but hybridization between them appears to occur frequently. Assembly parameters such as the amount of missing data and the threshold for between-sample clustering had an influence on tree topologies.

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Historical biogeography of Australian chenopod flora

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Globally prominent in deserts and on coastlines, Australian chenopod flora shows a peculiar evolutionary pattern known as the littoral connection. This suggest that Australian arid flora, albeit extremely diverse, underwent similar evolutionary trajectory of coastal landfall, inland migration through declining palaeodrainage systems via marshy, sandy or saline habitats and subsequent rapid diversifications in the harsh Australian outback. The immense evolutionary success is thus associated with the necessary preadaptation of the coastal flora on physiologically similar inland habitats, assuming niche conservatism. Nevertheless, hybridisation, extensive incomplete lineage sorting associated with rapid diversification and overall inaccessibility of the Australian outback flora have hindered rigorous testing of this hypothesis. The recent advances in NGS approaches now allow for detailed biogeographic studies of widely inaccessible floras worldwide. Current studies suggest that MRCAs of Australian chenopods reached the Sahul coastlines via oceanic dispersal. From there, continuous Neogene serial invasions took place through continental drainage systems, driven by niche conservatism. I will give a short overview on the recent advances on the evolutionary history of Australian chenopod flora, its multiple origins and migration trajectories, repeated biogeographic patterns and the most prominent evolutionary differences within and among clades.

Molecular evolution of biotic stress responses in diverse populations of *Solanum chilense*

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Wild tomato species form an intriguing group of wild plant species. Each have their own specific habitats and corresponding ecological niches, though some overlap in their ranges does exist. In a patchwork of studies several dozens of different pathogen resistance properties have been identified in for a number of pathogens in several of the species. However, systematic analyses of such resistances and the underlying genetic variation within a single wild tomato species are sparse.

We are especially interested in understanding such variation and evolution of pathogen resistance properties in a true demographic and ecological context. Our work focuses on the wild tomato species *Solanum chilense*. The species grows on the southern edges of the species range for wild tomato and its very specific demography, resulting from two different expansions into extreme environments make it the ideal species to study phenotypic and genetic diversity of biotic defences. In this talk, I will highlight several aspects of recent research projects. These range from genomics analyses that indicate clear resistance gene family evolution to phenotypic and biochemical studies that show tremendous variations in defence hormone responses between and within populations of one wild tomato species.

Non multa, sed multum: On the need for quality control in the generation and analysis of complete plastid genomes

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In contemporary botanical research, the sequencing and comparative analysis of complete plastid genomes has become *a routine procedure, with dozens*, if not hundreds, of new genomes generated per investigation. The number of publicly available plastid genome records has, consequently, skyrocketed: by September 2021, more than 13,000 unique and complete plastid genome sequences of flowering plants are available on NCBI GenBank, and thousands more are added every year. This large collection of plastid genomes is highly diverse taxonomically and represents nearly all major clades of flowering plants. Understandably, many researchers wish to utilize this genomic wealth and select specific genomes for reuse in their own investigations. When doing so, archived plastid genomes are often employed at face value, with little regard for their correctness in both sequence and annotation. Here, I take stock of the data quality among archived plastid genomes of flowering plants. Specifically, I illustrate that almost half of all plastid genomes archived on NCBI GenBank exhibit annotation errors in key sequence features. I also show that the assembly process, as well as the comparative analysis of these genomes, are much more capricious than widely acknowledged, up to the point of misleading subsequent phylogenetic analyses.

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No red leaves needed when you have root nodules

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Trees growing at a particular latitude in Eastern North America (ENA) receive significantly more autumnal solar irradiation than do trees growing at the same latitude in Eurasia (Renner and Zohner, 2019: Fig. 5a). This empirical difference provides a plausible explanation for the fact that a higher percentage of ENA trees and shrubs produce anthocyanin before shedding their leaves than observed in European and East Asian floras (Renner and Zohner, 2019, 2020). The proposed link between autumnal light intensity and anthocyanin deposition in leaves is photoprotection, allowing senescing leaves to cope for longer against photooxidative stress and, consequently, to sustain greater nutrient resorption, especially nitrogen, compared to anthocyanin-deficient (yellow) leaves. However, anthocyanins are costly, and for species with root nodules occupied by nitrogen-fixing symbionts their deposition in leaves that are about to be shed may not pay off. We tested this expectation with a worldwide sample of dehiscent species, among them 86 nitrogen-fixers from different families, and find that none of them produce anthocyanin in the fall. This finding disagrees with Pena-Novas and Archetti (2021) who found no correlation between nitrogen and fall colors because their sampling did not differentiate between species with nitrogen-fixing bacteria and those without.

- Pena-Novas, I., and M. Archetti. 2021. Nitrogen resorption efficiency in autumn leaves correlated with chlorophyll resorption, not with anthocyanin production. Journal of Evolutionary Biology 34: 1423-1431.
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Patterns of genetic diversity and bioactive constituents in Lumnitzera mangroves from Indonesia

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Indonesia is a mega-biodiversity country with the largest area and richest number of mangrove species. This archipelagic country is ideal for studying population genetics and potential new sources of bioactive compounds. In my presentation, I will give a brief summary of the results from our research on the biodiversity of Indonesia's mangrove genus *Lumnitzera*. We investigated the patterns of genetic diversity and wealth of active constituents of *Lumnitzera littorea* and *L. racemosa*. We employed 2,442 biallelic SNP loci using a ddRADseq approach and performed TLC, hyphenated LC-MS/MS, and isolation to analyze the constituent variety. Our results revealed low genetic diversity and strong genetic differentiation in both species. This pattern can be explained by sea surface currents and geographical distance limiting propagule dispersal. Our phytochemical characterization showed that both species possess unusual diversity in sulfated constituents not reported previously from mangroves. Molecular phylogenetic analyses using nuclear ITS rDNA supported the natural product differentiation in *L. racemosa* populations from different localities. This study advances the investigation of mangrove biodiversity by using a combination of genomics and metabolomics approaches and demonstrates the uniqueness and potential of *Lumnitzera* for human health.

An NGS approach to unraveling the phylogeny and biogeography of section *Pruinosae* within the SE Asian ant-plant genus *Macaranga*

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The dynamic climatic and geological past of Sundaland, Southeast Asia, has had a major role to play in the evolution and diversification of its biota. *Macaranga*, a genus of pioneer trees very commonly associated with ants, represents a system that enables a good understanding of this phenomenon. The majority of speciation and diversification events in this genus are known to have occurred during the late Pliocene and Pleistocene. In our study, we use next generation sequence (NGS) data (Genotyping-by-Sequencing) for the very first time to evaluate the phylogeny and phylogeography of a specific taxonomic section within this genus: section *Pruinosae*. Phylogenetic analysis suggests that the myrmecophytic trait evolved once and was later lost once within this section. Genetic data also revealed a marked distinction between the western (Malay Peninsula and Sumatra) and eastern (Borneo) populations within all broadly distributed species indicating the presence of a barrier to gene flow across the Sunda shelf. Populations from the island of Bangka, although geographically in the vicinity of Sumatra, genetically clustered with those from Borneo, thus bringing us closer to visualizing this barrier on a map.

New insights into the phylogeny and character evolution of Podocarpaceae inferred from transcriptomic data

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As the second largest family of conifers, Podocarpaceae comprise 19 genera and approximately 187 species. Phylogenetic relationships among the Podocarpaceae genera have not been well disentangled in previous studies, even when genome-scale datasets were used. In this study, 38 transcriptomes of Podocarpaceae and its sister group Araucariaceae were newly generated, and the phylogeny of Podocarpaceae was reconstructed from multiple datasets and methods. Using 993 nuclear orthologous groups and 54 chloroplast genes, we obtained a robust phylogeny of Podocarpaceae and revealed that topological conflicts of different phylogenetic trees could be attributed to synonymous substitutions and long-branch attraction. Several analyses including molecular dating, diversification rate estimation, and ancestral state reconstruction of characters were further performed. The results showed that two morphological traits, fleshy seed cones and flattened leaves, might be important for Podocarpaceae to adapt to angiosperm-dominated forests and thus could have promoted its species diversification.

Morphological and genetic inconsistency in the hybrid zones of alpine willow species pairs

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Species interactions are influencing the recolonization of the Alps since the Last Glacial Maximum and have contributed to the current species ranges. In secondary contact zones of two closely related but previously isolated species, homoploid hybrids can either generate adaptive combinations in the parental species by introgression or create a separate species by hybrid speciation. Hybrids may therefore block the range expansion of the parents. Although the hybrid zones influence the recolonization dynamic of species, only few studies focused on secondary contact hybrid zones so far.

Among the 35 willow species (*Salix* L.) occurring in the European Alps, some closely related diploid species pairs (e.g. *Salix alpina/Salix breviserrata*) show a distinct biogeographical vicariance pattern and hybridize. Next generation RAD (Restriction site Associated DNA) sequencing allows us to address the following questions: Are the morphologically hybrid phenotypes observed in the contact zones supported by the genetic analyses? Are the contact zones characterized by introgression or tendencies to hybrid speciation?

We will present first results that indicate low divergence of the two analyzed species (pairs) and their hybrids illustrated by a low amount of genetic structure, contradicting the morphological and geographical separation.

Highly diverse willows share highly similar plastomes

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Plastome phylogenomics is used in a broad range of studies where single markers do not bear enough information. Phylogenetic reconstruction in genus *Salix* is difficult due to the lack of informative characters and reticulate evolution. To overcome this lack we used a genome skimming approach to reconstruct 41 complete plastomes of 32 Eurasian and North American *Salix* species representing different lineages, different ploidy levels, and separate geographic regions. We combined the data with available plastomes from Genbank. The results revealed a highly conserved structure of the observed plastomes. Within the genus, we observed a variation of 1.68%, most of which separated subg. *Salix* from the subgeneric *Chamaetia/Vetrix* clade. Non-coding DNA regions were responsible for most of the observed variation within subclades. Coding regions showed a variation of 0.72%, and 5.6% of the analyzed genes showed signals of diversifying selection. A comparison of nuclear RAD sequencing and plastome data on a subset of ten species showed discrepancies in topology and resolution. We assume that a combination of (i) a very low mutation rate due to efficient mechanisms preventing mutagenesis, (ii) reticulate evolution, including ancient and ongoing hybridization, and (iii) homoplasy has shaped plastome evolution in willows.

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