

Hop Research Institute Co., Ltd.

## Genetic diversity of European wild hops in a historical context

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In the early Middle Ages, wild and cultivated hops (*Humulus lupulus* L.) were found in many parts of central and northern Europe and they have been used for the preparation of beer since the ninth century. The names of traditional landrace cultivars refer to the regions and places of origin from this time. Because of these landraces have been cultivated up to now, we would like to know about their influence on wild hop populations in Europe. DNA molecular genetic methods are the most appropriate technology for this purpose. We used 16 microsatellite EST-

SSR loci (Patzak et al., 2017) for cluster and PCoA genetic diversity analyses (Patzak and Henychová, 2018) of 218 wild hops and 44 landraces. Genetic diversity analyses (**Figure 1 and 2**) confirmed previous results that North American wild hops are different from European gemplasm (Patzak et al., 2010a, b) and their germplasm is in origin of old American cultivar Late Cluster. It was also proved differences for Caucasus wild hops (Patzak et al., 2010b). Saaz and Hersbruck landraces were historically the most important for genetic diversity of European wild hops. Our analyses showed that old cultivar Fuggle originated in wild hops from northern France. It could be supposed that active transfer of hop plants in Europe took place in the 14<sup>th</sup> century and subsequently also in the 19<sup>th</sup> century (**Figure 3**).

## **Materials and Methods**

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The young leaf or cone samples of 218 wild hops and 44 landraces have been collected since 2005 (Patzak et al., 2010a, b; Nesvadba et al., 2015; 2020; Ducrocq et al., 2022). DNA was isolated from samples by CTAB method and molecular analyses of 16 microsatellite loci were carried out according to Patzak et al. (2017). Dendrogram was based on Jaccard's similarity coefficient of 156 EST-SSR polymorphic molecular markers, determined by Neighbor-Joining (NJ) method of Unweighted Pair Group Method with Arithmetic means (UPGMA) in DARwin v. 5.0.155 (Dissimilarity Analysis and Representation for Windows, http://darwin.cirad.fr/darwin) software, visualized by Geneious Pro 4.8.2 (Biomatters Ltd., Auckland, New Zealand) software. The principal coordinate analysis (PCoA) was also conducted by DARwin software based on a genetic similarity/dissimilarity matrix. Three-dimensional graph was visualized with SigmaPlot for Windows v.10.0.0.54 (Systat Software Inc., San Jose, CA).

Figure 1. Dendrogram of genetic distances of 218 wild hops and 44 landraces revealed by unweighted pair group method with arithmetic means (UPGMA) and Neighbour-Joining (NJ) clustering based on the Jaccard similarity coefficient determined by 156 EST-SSR polymorphic molecular markers. Colours represent the country of origin: green – Czechia, blue – France, violet – Swiss, yellow – Spain, azure – Sweden, brown – Denmark, olive – Caucasus, orange – USA, pink – Canada, red – landraces.

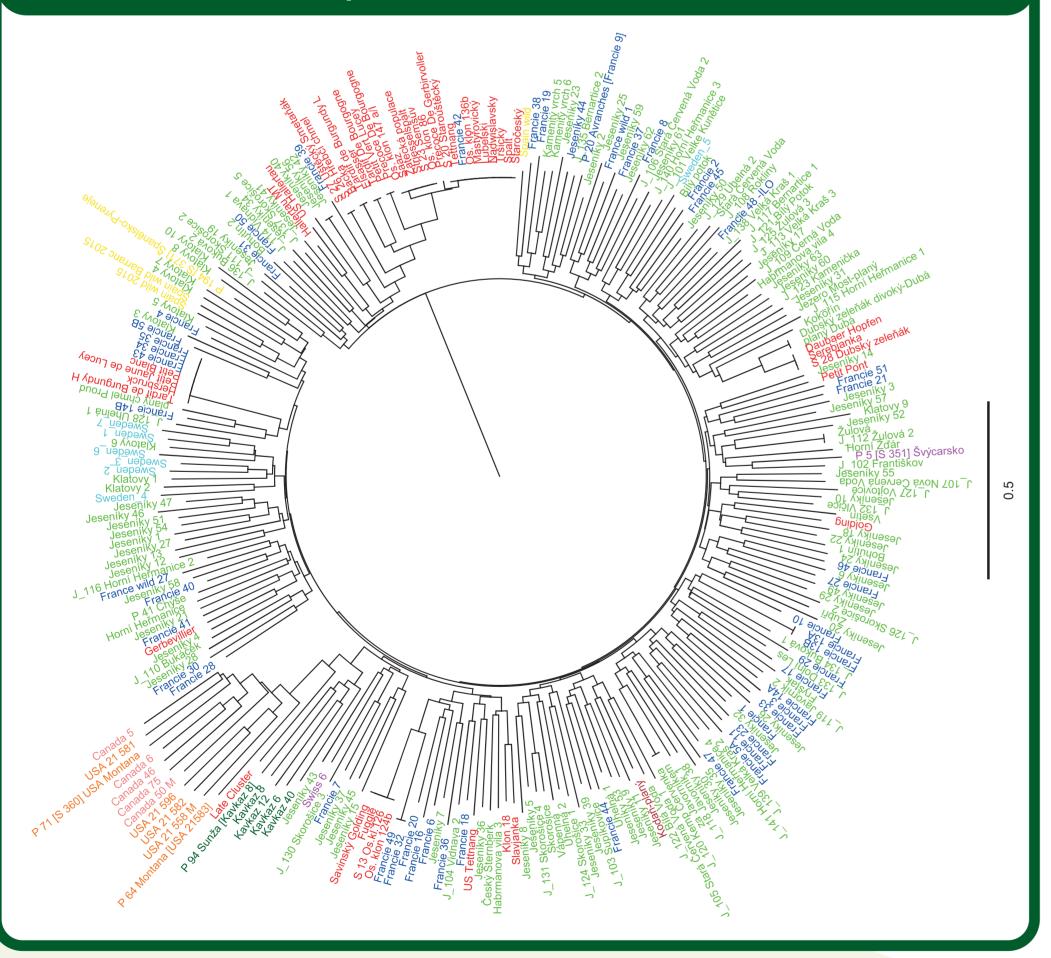
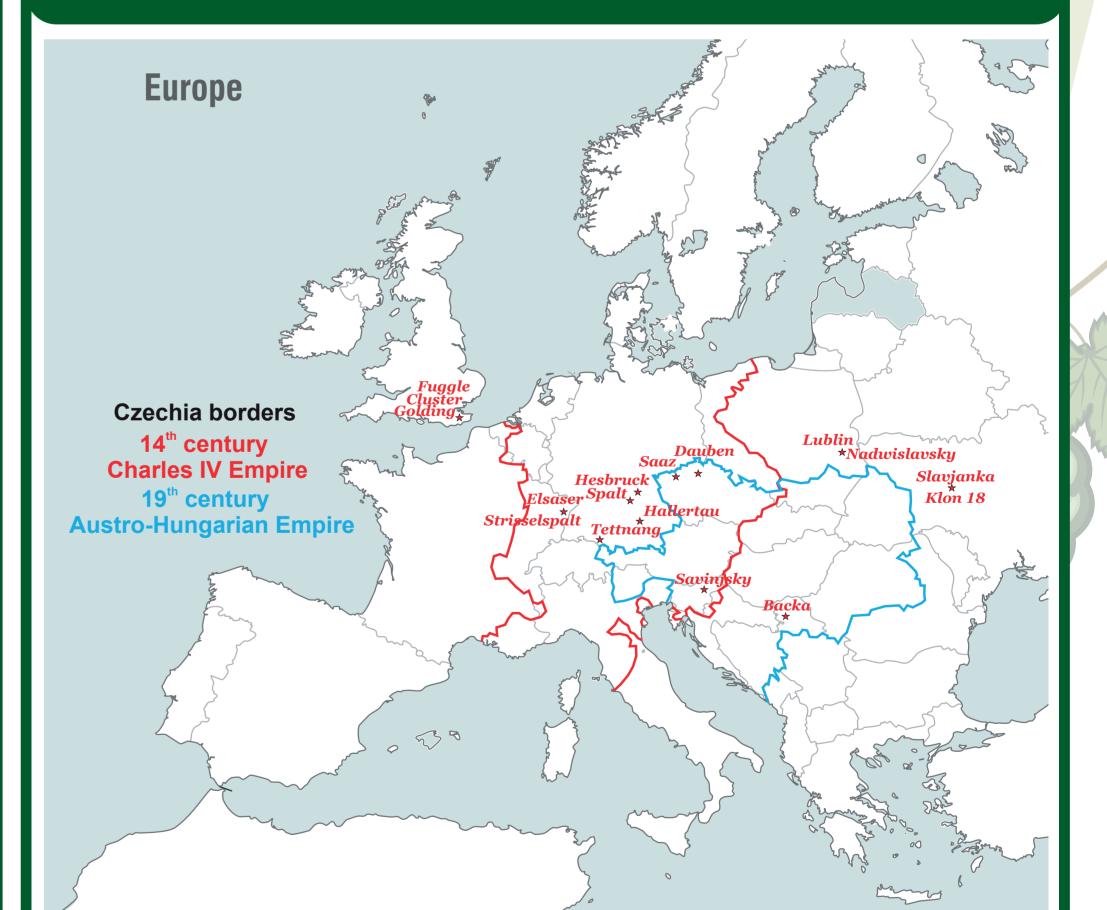


Figure 2. Principal coordinate analysis of 218 wild hops and 44 landraces revealed by DARwin v. 5.0.155 (Dissimilarity Analysis and Representation for Windows, http://darwin.cirad.fr/darwin) based on 156 EST-SSR polymorphic molecular markers. Colours and signs represent the country of cultivar origin. x, y and z axes are the first, the second and the third principal coordinates.

0,2 Dauben hopfen USA Canada  $\cap$ 0.0 axis Caucasus Spain Swiss Sweden Denmark France Fuggle Czechia -0,4  $\Delta$ Landraces 0,2 axie 0,0 -0,4 -0,2 0,0 0,2 0,4 0,6 0,8 1,0 -0,2X axis

Figure 3. Hop landrace places (stars) on Europe map with borders of Czechia within Charles IV Empire in the 14<sup>th</sup> century and within Austro-Hungarian Empire in the end of 19<sup>th</sup> century.





Ducrocq, F., Laflotte, A., Girardeau, L., Patzak, J., Hehn, A., Piutti, S.: Analysis of genetic and metabolomic diversity in wild hops collected in the French North-East region. Proceedings of Scientific-Technical Commission of IHGC, Lugo, Spain, July 3-7, 116, 2022.

Nesvadba, V., Charvátová, J., Henychová, A., Patzak, J.: Evaluation of historical original clones of hops (Humulus lupulus L.). Kvasný průmysl 2020, 66, 382-391.

Nesvadba, V., Pšenáková, I., Ürgeová, E.: Variabilita planých chmelů a jejich využití ve šlechtění chmele. In: Zedek, V., Mládková, A., Holubec, V. eds. Genetické zdroje rostlin a změna klimatu. Praha, MZe, 2015, 63-66. ISBN 978-80-7434-249-3.

Patzak, J., Henychová, A.: Evaluation of genetic variability within actual hop (*Humulus lupulus* L.) cultivars by enlarged set of molecular markers. Czech Journal of Genetics and Plant Breeding 54: 86-91, 2018. Patzak, J., Krofta, K., Henychová, A.: Metody a markery pro prokázání autenticity českých odrůd chmele. Certifikovaná metodika, Žatec, Chmelařský institut s.r.o., 2017, 86 pp. ISBN 978-80-86836-18-8 Patzak, J., Nesvadba, V., Henychová, A., Krofta, K.: Assessment of the genetic diversity of wild hops (*Humulus lupulus* L.) in Europe using chemical and molecular analyses. Biochemical Systematics and Ecology 2010a, 38, 136-145. Patzak, J., Nesvadba, V., Krofta, K., Henychová, A.; Marzoev, A.I.; Richards, K. Evaluation of genetic variability of wild hops (*Humulus lupulus* L.) in Canada and the Caucasus region by chemical and molecular methods. Genome 2010b, 53, 545-557.

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