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3	Genetic Considerations in European Riparian Ecosystems Management:
4	Experts View on Status and Needs
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29 Abstract

30 1. Riparian vegetation supports high biodiversity providing many services and is, therefore, an important 31 landscape element. Riparian ecosystems are subject to numerous pressures leading to population decline and 32 genetic erosion of riparian plants. This may have cascading effects on various ecosystem levels, decreasing 33 services, so identifying the current status of riparian tree species genetic conservation needs is vital to improve 34 the effectiveness of restoration efforts.

- 35 2. We aimed to identify expert views on the status and importance of tree species genetic diversity, including
- 36 both adaptive (morphological) and neutral (molecular), and conservation needs across European riparian
- 37 ecosystems. Sharing such information among researchers, managers and policymakers have the potential to
- 38 enhance ecological restoration and management of riparian ecosystems.

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39 3. We identified experts in riparian genetic resources conservation and management across Europe. These 40 included stakeholders with different perspectives, ranging from researchers to practitioners. We designed a 41 set of questionnaires that asked our identified experts to answer questions related to riparian tree species 42 genetic conservation in their respective countries. Specifically, we asked about societal awareness, legislative 43 tools, good practices and conservation or restoration projects taking account of tree species genetic resources 44 in riparian ecosystems. Questionnaire responses were analyzed and discussed in light of scientific literature to 45 define needs and priorities related to the management and conservation of riparian tree species genetic 46 resources. 47 4. The experts recognized the ecosystem approach, as the most appropriate option for the conservation of tree 48 species genetic diversity in riparian ecosystems applied through a combination of *in-situ* and *ex-situ* measures 49 and/or integrative conservation. Simultaneous application of conservation measures at the level of priority

50 species, identified by experts, and protection of riparian areas are required.

51 5. Synthesis and applications. The results of this study revealed the importance of recognizing the ecological 52 processes that shape tree species genetic diversity in hydrographic networks (spatial patterns and gene flow, 53 fragmentation) but also the need to overcome socio-economic barriers, such as lack of policy priority, 54 deficiency in funding and weak legislation framework.

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56 Key words: genetic erosion, genetic resources conservation, knowledge conversion, riparian genetic diversity, 57 riparian vegetation

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60 **1 INTRODUCTION**

61 Riparian ecosystems have been recognized as critically important elements in the landscape, 62 providing multiple benefits to the environment and societies (Dufour et al. 2019). Riparian 63 vegetation in general, and specifically riparian trees, are of special significance, contributing to 64 multiple key ecosystem services, including filtering agricultural pollutants and improving river water 65 quality, regulating instream temperature, stabilizing river banks, sequestering carbon and producing 66 biomass, as well as providing cultural and recreational services (Riis et al. 2020). Whilst much 67 research has been devoted to the importance of species richness for riparian ecosystem functioning 68 and stability, relatively less is known about the potential ecosystem-level importance of genetic 69 diversity of riparian tree species. Genetic diversity within riparian plant populations differs from 70 other ecosystems in that downstream dispersal of propagules with flowing water leads to high gene 71 flow and downstream increases in genetic diversity. Riparian habitat continuity across river networks 72 leads to low genetic divergence among sites (Nilsson et al. 2010). Moreover they are subject to 73 multiple pressures that lead to genetic diversity reduction. For example, fragmentation by dams 74 (Werth et al. 2014) and hybridization with non-native populations threaten the genetic diversity and

75 integrity of riparian plant species. This is true also for important foundation species like Populus 76 nigra (Vanden-Broeck et al. 2005; Chenault et al. 2011; Vanden-Broeck et al. 2012), potentially 77 leading to low resistance to pathogens and increased extinction risk (Fady et al. 2020). This genetic 78 component is crucial for the many efforts to restore and recover riparian and floodplain vegetation. 79 For example, riparian vegetation has great potential for the deployment of nature-based solutions 80 which address the recovery of ecosystem structure and functioning (UNEP & IUCN, 2021), but the 81 provenance of plant material used in planting and seeding can be critical for the success of projects 82 (McKay et al. 2005; Breed et al. 2019). Despite these challenges, the genetic component is rarely 83 integrated into the management and conservation of riparian ecosystems and no transnational 84 vision has been developed on priorities in this domain.

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The aim of this paper is to identify expert views on the status and importance of tree species genetic diversity, including both adaptive (morphological) and neutral (molecular), and conservation needs across European riparian ecosystems. The research was conducted within the network of COST Action CONVERGES (<u>https://converges.eu/</u>), which aims to improve knowledge conversion and technology transfer among researchers, practitioners and policymakers for the improvement of riparian ecosystems management.

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94 2 METHODOLOGICAL FRAMEWORK

95 The main goal of CONVERGES is to synthesise existing knowledge about riparian vegetation across 96 countries and disciplines, covering topics from riparian research to restoration and management. A 97 subgroup named "Genetic conservation" (GC-WG2) was formed during the CONVERGES WG2 98 meeting (Iceland, 2018) to review the state-of-the-art in genetic conservation of riparian vegetation 99 at the European level and to identify the main knowledge gaps, conservation barriers and future 100 research and management needs.

Firstly, a *short questionnaire* (Appendix S1-A) was sent to relevant experts (researchers, stakeholders and practitioners) across the 39 COST participating countries, aimed at, collecting information on the research dimension (available literature, relevant projects), and identifying relevant riparian species. In total 22 responses were received from 15 countries (Appendix S2-A).

Secondly, *structured questionnaires* (Appendix S1-B) collected country reports covering strategies implemented so far, major barriers, current needs and potential solutions for improving the conservation of genetic resources across European riparian ecosystems. Analyses of structured questionnaires included numeric data and text data with open-ended questions (NVivo software

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- 109 <u>https://www.qsrinternational.com/nvivo-qualitative-data-analysis-software/home</u>). There were 30
- 110 responses from 19 countries (Appendix S2-B).
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112 3 STATUS OF GENETIC CONSERVATION OF RIPARIAN VEGETATION IN EUROPE

113 3.1 Research dimension

114 The genetic composition of riparian vegetation is influenced by several factors (biogeography, local 115 conditions and species interactions), but what sets it apart from other plant communities is water 116 flow as a vector for dispersal and the hierarchical structure of river networks (Nilsson et al. 2010). 117 Water flow is the main agent of dispersal of propagules and therefore influences downstream gene 118 flow among populations of riparian plants (Nilsson et al. 2010), while upstream gene flow mediated 119 by wind, or animals occurs less frequently (Wubs et al. 2016). As a consequence of downstream-120 biased gene flow, upstream populations may experience higher loss of alleles, while downstream 121 populations receive more alleles preventing loss due to genetic drift (Paz-Viñas et al. 2015). Yet, 122 other patterns may apply, as in Populus nigra L. where (i) no significant upstream/downstream 123 differences in genetic diversity occur (Imbert & Lefevre 2003) and (ii) clonality and sexual 124 regeneration can co-occur within the same natural stand (Tinschert et al. 2020). This results in high 125 complexity and requires extensive knowledge to implement adequate management measures.

126 In spite of the ecological importance and the recognized threats to riparian genetic diversity 127 conservation, the results of the structured questionnaires revealed both a lack of assessment of 128 genetic diversity status and changes in riparian tree species and the perception of a generally poor 129 status of riparian ecosystems. For example, the view of respondents about changes in riparian 130 genetic diversity in their country over the past ten years was that the situation is degrading. For this 131 question, three options were possible: "no significant changes", "improving status" or "degrading". 132 Apart from France and the Czech Republic, respondents from all countries (18/30) thought that 133 riparian genetic diversity was degrading. The respondent from France stated that a large number of 134 genetic conservation actions have been implemented and expanded knowledge about the genetic 135 diversity of riparian tree species (e.g. *Populus nigra*) has been achieved by using new DNA markers 136 (e.g. Single Nucleotide Polymorphism (SNP), Faivre-Rampant et al. 2016). According to the 137 informants, there was no coordinated plan of actions addressing the status of riparian tree species 138 genetic diversity.

Recently, several studies have demonstrated how anthropogenic barriers and habitat fragmentation affect the genetic structure of riverine plant populations (Werth et al. 2014). Such genetic isolation of rare and critically endangered plant populations can accelerate genetic erosion (the loss of genetic diversity within a species through a reduction in richness of alleles or alleles combination

over time) and lead to local extinctions (Labonne et al. 2008). Therefore, adequate assessments of
 genetic diversity in riparian ecosystems, especially within tree species, contribute to efficient
 conservation.

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148 3.2 Policy dimension

149 The long-term degradation of riparian ecosystems has continued to intensify since the 1950s, and 150 has contributed to reduced river health (Singh et al. 2021), low achievement of the objectives of the 151 Water Framework Directive, and the unfavourable status of floodplain forest habitats across Europe 152 (EEA 2018). Cortina-Segarra et al. (2021) identified genetic considerations as a pending crucial 153 element to enable the improvement of ecosystem restoration effectiveness. In this sense, riparian 154 tree genetic resources conservation lies in an undefined gap between the policies addressing nature 155 conservation and forestry. Genetic considerations are not treated as a separate issue in forestry 156 legislation. National-level forestry-planning documents refer to forest genetic resources in different 157 contexts, but without defining methods for their management in the context of genetic 158 conservation.

159 To the question, does your countries have plans or programs to assess the state of genetic diversity 160 of riparian tree species, 63% of respondents gave a negative answer. Respondents who gave a 161 positive answer (33%) cited plans and programmes concerning biodiversity protection and forest 162 genetic resources conservation in general, sometimes mentioning specific riparian tree species. 163 Indeed, for forest tree species, a step forward has been the adoption of the Global Plan of Action for 164 the Conservation, Sustainable Use and Development of Forest GR (2014), and its implementation 165 strategy (http://www.fao.org/3/i3849e/i3849e.pdf), an outcome of the EUFORGEN programme, that 166 encourages countries to upgrade the system of collecting genetic data and to establish a core 167 network of dynamic genetic conservation units (GCU) across Europe (www.eufgis.org). Moreover the 168 First Report on the State of the World's Forest GR (2014) calls for urgent action for better 169 management of forests and forest genetic resources to ensure their long-term sustainable use for 170 the local community that depends on forest products.

When it comes to riparian vegetation plant species (beyond other than trees), genetic issues are even more neglected. Riparian zones are subject to multiple legal instruments such as those related to land ownership, flood control or nature conservation. Yet, monitoring and assessment are not mandatory in European countries (González del Tánago et al. 2021), and the genetic component of riparian diversity is not addressed. Thus, when asked if there are procedures in place to moitor or measure genetic erosion within riparian tree species R 70% of respondents gave a negative answer.

177 These results reflect the neglect of riparian genetic resourcesand the disparity in planning the 178 assessment and monitoring Respondents (23%) who gave a positive answer provided only 179 information on procedures for monitoring the state of forests and forest genetic resources in 180 general No ilformation regarding tpecific monitoring of the state of riparian genetic resources was 181 nrovided. Riparian zones are subject to multiple legal instruments related to land ownership, flood 182 control or nature conservation. Yet, monitoring and assessment of riparian tree species genetic 183 diversity and connectivity are not mandatory in European countries (González del Tánago et al. 184 2021).

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186 3.3 Management dimension

187 Since it is not included in most policies, riparian tree species genetic diversity has been mostly 188 disregarded in impact assessments, monitoring and restoration projects. When asked about the 189 most effective approaches to conserving riparian tree species genetic resources, participants were 190 given four options: *in-situ*, *ex-situ*, combination (*ex situ* and *in situ*) and integrative approaches (i.e. 191 conservation that includes stakeholder engagement to achieve sustainable management of natural 192 resources, González et al. 2017). Forty percent of respondents concluded that "in-situ + ex-situ" 193 approaches were most effective. A slightly lower proportion (37%) supported integrative 194 conservation. Ex-situ as a separate approach was not selected by any respondent which indicates 195 that the majority of the respondents (both from the scientific community and practitioners) agrees 196 upon that pure *ex-situ* is not effective for conserving riparian tree species genetic diversity. While 197 few riparian species have been studied genetically, the research on the genetic structure of black 198 poplar (Populus nigra), one of the most threatened tree species in Europe, provides a basis for 199 recommendations on management of black popular and other riparian tree species. Restoring and 200 re-instating the natural dynamics of floodplains, in combination with having sufficiently sized and 201 spaced natural populations as seed sources, is recommended for long-term conservation (Smulders 202 et al. 2008). Using local provenances for revegetation is vital to avoid introgression from exotic 203 cultivars (Chenault et al. 2011). This requires identification of natural populations as propagule 204 sources (e.g. 46 genetic conservation units across Europe for P. nigra; www.eufgis.org). These "in-205 situ" conservation actions should be supported by "ex-situ" measures to preserve indigenous genetic 206 material, in case of in-situ measures failure (Storme et al. 2004). For example, in Italy, the CREA 207 (www.crea.gov.it) long-term ex-situ collections (2500 poplars and willows), have been already used 208 in river restoration.

209 According to the results of the structured questionnaires, the main benefit of conserving riparian 210 tree species genetic diversity highlighted by 28 of the 30 respondents is freshwater ecosystem

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211 conservation. Also, 27 of them recognised scientific interest as another main benefit. Filtering water 212 pollutants, social importance, and economic benefits were also highly ranked. Six out of 30 213 respondents recognized food security as one of the main benefits.

214 The results overall showed that the ecosystem approach, defined by the COP 5 Decision V/6 as "a 215 strategy for the integrated management of land, water and living resources that promotes 216 conservation and sustainable use in an equitable way", is recognized as the most appropriate 217 option for the conservation of riparian tree species genetic diversity. Among the existing examples of 218 good practices in the integrative conservation of riparian tree species genetic diversity, mentioned 219 is the REFOCuS by the respondents, project 220 (http://www.interreg-danube.eu/approved-projects/refocus) that aims to boost riparian forest 221 resilience in the Mura-Drava-Danube biosphere reserve as ecological corridors.

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223 4 NEEDS AND PRIORITIES

224 The structured questionnaires revealed a general lack of an integrated strategy for the conservation 225 of riparian tree species genetic diversity across European countries. Existing efforts in specific 226 countries have focused on a few flagship species, often integrated into forestry programmes, but 227 offering limited progress from an ecosystem perspective. Thus, it is urgent to integrate the genetic 228 dimension in riparian vegetation management and to integrate riparian tree species in genetic 229 resource conservation strategies. Understanding ecological processes (e.g. gene flow) and pressures 230 (e.g. river fragmentation) in hydrographic networks is essential to conserve intraspecific genetic 231 diversity and adequately manage riparian tree genetic resources .

In the last part of the structured questionnaire, respondents were asked to identify specificities, strengths and weaknesses of riparian tree species genetic diversityconservation as well as to propose and describe the most beneficial project at the national level. According to the results, four groups of answers were identified as follows:

236 *Specificities* of conserving riparian tree species genetic diversity in comparison with other 237 ecosystems fell under eight categories: biodiversity, gene flow, regeneration, ecosystem functioning, 238 water regimes, legislation, water management and pressures.

Identified *strengths* (*Figure 1*) that can help achieve effective riparian tree species genetic diversity
conservation, were: scientific knowledge, diversity status (inventories of the species),
institutional/organizational framework, community awareness, environmental
conditions/accessibility, legislation framework, financial support, policy priority and others.



FIGURE 1

245 Strengths identified by participants that can be used to achieve effective riparian GR conservation

Weaknesses (*Figure 2*) that should be alleviated and/or eliminated were classified in the following 248 categories: lack of policy priority, financial, legislation framework, institutional/organizational

249 framework, community awareness, diversity status/inventories of species, scientific knowledge level,

250 environmental barriers, and others.





legislation framework, which suggests a need for better governance to conserve riparian tree genetic
 resources. Respondents were of the opinion that policy needs to be given priority to riparian tree

259 species genetic diversity conservation to help design plans and develop projects that can be 260 implemented in short term.

The fourth question was to imagine the most beneficial project (Figure 3, Appendix S2-B) aimed at riparian tree genetic resources conservation at the national level. Their answers covered diverse topics: *in-situ* measures, research, knowledge transfer, vegetationinventories, monitoring, referencing hotspots, *ex-situ* measures, upgrading projects, funding scheme, genetic screening and education and professional training. The first four topics were most frequently mentioned by respondents as illustrated in Figure 3.



the conservation of intraspecific genetic diversity. The species that were most highly often cited bythe experts and practitioners are:

Alnus glutinosa (L.) Gaertner – mentioned in as many as 14 countries as a functionally and
 economically important species, but also due to conservation aspects. The conservation importance
 of this species is recognized in countries with very different environmental conditions throughout
 Europe (i.e. priority habitat 91E0*). An additional reason for prioritizing this species for conservation
 is its endangerment due to widespread population decline caused by the epidemic fungal pathogen
 Phytophthora x alni (Bjelke et al. 2016).
 Salix alba L. – mentioned in 10 countries. Similar to many other species in the Salicaceae family, it

is found living in the active channel and floodplains, mostly as a functionally important species

within in the typical dynamics of riparian ecosystems (Karrenberg et al. 2002; Degirmenci 2019).

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3. *Populus nigra* L. – mentioned by the experts from 9 countries (mostly the Central-South-East European countries), primarily as a conservation concern but also a functionally and economically important species. Genetic diversity of the species is highly endangered due to introgression and gene flow with both hybrid cultivars (especially *P. x euramericana* hybrids) and pure *P. nigra* varieties like the Lombardy poplar (Vanden-Broeck et al. 2005; Chenault et al. 2011; Vanden-Broeck et al. 2012).

4. Ulmus laevis Pall. – mentioned by the experts from 8 countries as a conservation concern, due to
reduction of its natural populations caused by Dutch Elm Disease and wetland degradation
(Devetaković et al. 2016).

295 Other species of importance for the genetic conservation of RE, according to respondents, are as 296 follows: *Quercus robur* L., *Populus alba* L., *Fraxinus angustifolia* Vahl, *Ulmus minor* Mill., *Salix* spp. (*S.* 297 × *rubens*, *S. fragilis*, *S. atrocinerea* Brot., *S. eleagnos* Scop., *S. salviifolia* Brot.), *Fraxinus excelsior* L., 298 *Populus* spp. (*P.* × *canescens*, *P. tremula*), *Platanus orientalis* L., *Alnus incana* (L.) Moench, *Betula* 299 *pubescens* L., *Carpinus betulus* L., *Liquidambar orientalis* Mill. and *Prunus padus* L.

Based on issues highlighted by the questionnaire respondents, we developed a framework for conducting surveys of the genetic status of species (Appendix S3), encompassing (1) Mapping of current status; (2) Species-level plans; (3) Funding; (4) Monitoring; (5) Knowledge transfer and dissemination; and (6) Evaluation. We envision that if the framework is applied to the species highlighted above, this can in turn inspire work on additional taxa.

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306 **5 CONCLUSIONS/PROSPECTS**

The omission of genetic considerations in riparian management can potentially hamper efforts to manage river ecosystems to conserve biodiversity and ensure ecosystem service provision, given the key role of riparian vegetation and specific tree species at risk of genetic erosion. This omission needs to be addressed due to intensification of human [biotic and abiotic] pressures on riparian ecosystems, such as biological invasions, emerging alien diseases, and genetic pollution, that are leading to critical biodiversity loss.

313 Despite shortcomings in terms of representativeness of the respondents consulted, this study 314 represents (to our best knowledge) the first pan-European survey that offers a broad view of the 315 issues of genetic resources management for riparian tree species on a continental scale. Future 316 research would benefit from addressing riparian genetic patterns and ecological processes such as 317 gene flow, impacts caused by human alteration, such as agricultural use of riparian zones, 318 fragmentation of hydrographic networks, as well as global change, including potential geographic 319 range shifts or maladaptation of populations at the geographic range limits. In riparian restoration 321 schemes involving seeding or planting it is vital that genetic diversity is ensured, and that locally 322 adapted, native species are used. Furthermore, an understanding of the ecological processes that 323 shape riparian ecosystems (Rodríguez-Gonzalez et al. 2019) is required.

324 Research needs to be followed by adequate knowledge transfer both to managers and decision-325 makers to better inform legislation and implementation. Incorporating and improving policies on 326 genetic considerations are especially crucial as most species are non-commercial and binding 327 regulations are required to control/certify the origin of the plant material that is made available in 328 nurseries. In the context of increasing promotion of ecosystem restoration, guidance on best 329 practice in the management of plant reproductive material from local native populations (e.g. 330 cuttings propagation in the field with an adequate and large genetic diversity; seedlings with 331 identified and certified origin) is crucial. Inclusion of genetic diversity in assessments of the native 332 riparian tree species population status and recognition of trajectories in long-term monitoring are 333 needed to ensure ecological integrity of riparian genetic resources in the future.

334 Genetic diversity is important both to ensure current population fitness and to maintain their 335 adaptive potential to respond to environmental change (Fady et al. 2020). Restoration guidelines 336 strongly recommend considering among-habitat genetic differentiation, and using local propagule 337 sources to ensure evolutionary potential (Broadhurst et al. 2008), which requires identifying natural 338 populations to serve as propagule sources. Due to the comparatively rapid dynamics of riparian 339 forest species (rate of founding and extinction of populations), genetic conservation measures need 340 to be streamlined to enable their rapid adaptation. Resistance to emerging pathogens will be critical, 341 requiring selection of multiple individuals with resistance to pathogens, such as in elms, ash and 342 alder (Bjelke et al. 2016). In-situ conservation and restoration should be supported by ex-situ 343 conservation in seed orchards, clone collections, seed collections, arboreta, botanical gardens and 344 gene banks, that adequately represent the genetic diversity of natural populations (Storme et al. 345 2004).

346 Improved integration of ecological processes that shape genetic resources in hydrographic networks 347 (spatial patterns and gene flow, fragmentation) into management is required. Overcoming socio-348 economic barriers, such as the lack of policy priority and funding, and the weak legislation 349 framework would help adoption of an ecosystem approach for sustainable conservation of riparian 350 genetic resources within present and future climatic and land-use scenarios.

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363 AUTHORS' CONTRIBUTIONS

JM, PMRG, RP, GH and RJ conceived the study. JM and PMRG led the writing of the manuscript. FA
analysed data and prepared figures. All authors FA, JM, RP, GH, RJ, SD, MB, NB, LSB, GB, DB, PMC,
ND, YSG, MI, MKQ, NLP, MN, EN, EP, MŠN, LV, MV, PZ and PMRG contributed data and reviewed the
manuscript.

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369 DATA ACCESSIBILITY

- 370 "Data is accessible upon request to the corresponding author"
- 371

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