









136 established (Patoka et al. 2016b). The target area was defined as the territory of Hungary  
137 containing 14 climatic stations from the database of the WorldClim project (Hijmans et al.  
138 2005). Where the climate match between the source area and the climatic station in the target  
139 area reached a score of  $\geq 7.0$ , this was interpreted as there is no environmental barrier to  
140 survival (Kalous et al. 2015).

141

#### 142 *Risk assessment*

143 To evaluate the invasive potential of decapod crustaceans pet-traded in Hungary, we used the  
144 Freshwater Invertebrate Invasiveness Scoring Kit produced by the UK Centre for  
145 Environment, Fisheries & Aquaculture Science (FI-ISK, v.1.19, Tricarico et al. 2010). Based  
146 on the FI-ISK score, each evaluated species was subsequently classified as (i) low-risk (score  
147  $<1$ ), (ii) medium-risk (score  $\geq 1$  but  $<16$ ), or (iii) high-risk (score  $\geq 16$ ) from a Hungarian  
148 perspective.

149

#### 150 *Field sampling*

151 Selected localities were sampled for potential occurrence of pet-traded decapods from January  
152 2015 to March 2017. It covered a range of both thermal localities (warm-water ponds and  
153 their outflows, sometimes connected with thermal spas;  $n = 11$ ) and those possessing regular  
154 temperature conditions ( $n = 17$ ) in the Danube river, its side arms and tributaries. Since then,  
155 several new populations of exotic aquatic species and potential habitats were identified  
156 (Takács et al. 2017, Weiperth et al. 2015, 2016). We sampled 1 km long section of each  
157 selected waterbody using five baited traps set every two weeks from spring to autumn.  
158 Thermal localities were additionally monitored during the winter. The traps were exposed and  
159 checked for two consecutive days. The morphological identification of captured crayfish  
160 followed Holthuis (1949) and Souty-Grosset et al. (2006).

161

#### 162 *Genetic analysis*

163 Due to confusion in the taxonomy of some *Cherax* species (see Austin 1996, Bláha et al.  
164 2016), we included DNA analysis in our study. The importance of DNA identification has  
165 been previously highlighted for several non-native crayfish species (Filipová et al. 2011).

166 The morphological identification of each captured specimen was confirmed by a polymerase  
167 chain reaction (PCR) of a selected gene (mitochondrial cytochrome oxidase subunit I – COI),  
168 utilizing universal primer pair LCO1490 (5'-GGTCAACAAATCATAAAGATATTGG-3')  
169 and HCO2198 (5'-TAAACTTCAGGGTGACCAAAAAATCA-3') (Folmer et al. 1994). The

































