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12. ROLE OF DIAPAUSE IN DISPERSAL OF AQUATIC INVERTEBRATES

12.1   INTRODUCTION

Diapause can be a key determinant of dispersal ability in aquatic invertebrates. This is especially true for zooplankton that produce diapausing eggs. For decades, anecdotal evidence has suggested the overland movement of zooplankton dormant eggs by vectors such as wind, water, and vertebrates, and recent experimental and genetic evidence has supported this claim. During the last century, the role of human-mediated dispersal of zooplankton has increased, specifically with regard to shipping. Generally, the role of human-mediated vectors is most important for species dispersal across geographical barriers and into large aquatic ecosystems that experience considerable shipping traffic. Diapausing stages facilitate species survival during movement across geographical barriers under extreme conditions, such as in ballast tanks of ships. Once in their new environment, some cladocera show altered seasonal phenologies, switching to both early and prolonged gamogenetic reproduction, which facilitates invasion success and further dispersal into novel ecosystems by both natural and human-mediated vectors.

Most organisms live in habitats that vary in space or time. One way in which organisms respond to this variability is through dispersal, which can play a key role in altering relative fitness and influencing ecological and evolutionary dynamics (Gadgil 1971; Levin et al. 1984; McPeek & Holt 1992; Ricklefs & Schluter 1993). A second way in which many organisms respond to environmental variability is through diapause (Tauber et al. 1986; Brendonck et al. 1998), which can also be thought of as dispersal through time (Venable & Lawlor 1980; Hairston 1998). In aquatic invertebrates, the active stage often cannot survive for long periods out of water. Hence, the production of desiccation-resistant propagules such as eggs, cysts, gemmules, or statoblasts (embryonic diapause *sensu* Alekseev & Starobogatov 1996) has been frequently considered an adaptation for dispersal (Karlson 1992; Korovchinsky & Boikova 1996; Fell 1998; Hairston 1998; Brendonck & Riddoch 1999; Bilton et al. 2001).

Despite a long history examining the production of diapausing stages in aquatic invertebrates, most studies have focused on conditions that promote the occurrence of resting eggs, factors that affect their survival and hatching from sediments, the presence of egg banks in sediments, and the impact of hatchlings from resting eggs on plankton community structure (Marcus 1996; Brendonck et al. 1998). Moreover, because so many aquatic invertebrates produce resistant stages, dispersal and colonization ability has often been assumed to be "rapid and frequent" (Brooks & Dodson 1965; Pennak 1989; Lampert & Sommer 1997). This assumption has been supported

by decades of anecdotal evidence suggesting the overland dispersal of freshwater aquatic species (Darwin 1859; McAtee 1917; Lansbury 1955; Maguire 1959; Proctor 1964; Proctor & Malone 1965; Swanson 1984; Bohonak & Whiteman 1999). Only recently, however, have studies begun to focus explicitly on importance of resting stages for dispersal of aquatic invertebrates (Bilton et al. 2001; Cáceres & Soluk 2002; Panov et al. 2004; Vandekerkhove et al. 2005e).

In this chapter, we focus on impacts of diapause on dispersal of aquatic invertebrates, primarily on planktonic organisms. We review major mechanisms and vectors of dispersal, both natural and human-mediated, of diapausing invertebrates. Also, we discuss the importance of diapausing stages in anthropogenic introductions and invasion success of aquatic invertebrates.

12.2   MECHANISMS AND VECTORS OF DISPERSAL OF DIAPAUSING INVERTEBRATES

Diapausing aquatic invertebrates (including their resting stages) can be dispersed by natural (surface water connections, ocean currents, wind, and animals) and human-mediated vectors. Dispersal by human vectors are usually viewed as "introductions," either intentional or unintentional, and such vectors have been discussed by many authors (Carlton 1996; Bilton et al. 2001; Minchin & Gollasch 2002; Havel & Shurin 2004). Sections 12.2.1 and 12.2.2 briefly summarize natural and human-mediated vectors most important for dispersal of diapausing aquatic invertebrates.

12.2.1   Natural Vectors of Dispersal

Natural mechanisms and vectors of dispersal of aquatic invertebrates have been discussed in detail in recent reviews by Bilton et al. (2001), Bohonak and Jenkins (2003), Havel and Shurin (2004), and Panov et al. (2004). Diapausing eggs or cysts of aquatic invertebrates have been often considered as potential agents of dispersal by natural vectors, and even depicted in the literature as adaptations for dispersal (Maguire 1963; Korovchinsky & Boikova 1996). Several authors have suggested that diapausing eggs may be transported by wind and rain (McAtee 1917; Maguire 1963; Cáceres & Soluk 2002). However, the scale at which this mode of dispersal is most effective remains unresolved. Wind dispersal of anostracan eggs may result only in their short-distance transport (Brendonck & Riddoch 1999). In general, long-distance dispersal of diapausing stages as aerial plankton is unlikely (Jenkins & Underwood 1998; Bilton et al. 2001; Bohonak & Jenkins 2003).

In contrast, currents in marine ecosystems and rivers may be important vectors for long-distance dispersal of active and diapausing invertebrates (Minchin & Gollasch 2002; Havel & Shurin 2004). Dispersal of gemmules of sponges by currents (as well as by fish and waterfowl) is discussed by Fell (1998). A midsummer shift toward sexual reproduction was observed by Makrushin (1984) in populations of the marine cladocerans *Podon leuckarti* and *Evadne nordmanni* in the Northern Atlantic. He hypothesized that the ability to produce resting eggs throughout most of the summer season was an adaptation to the large-scale dispersal of *Podon* and *Evadne* by oceanic currents, which are an important natural dispersal vector for marine

organisms. Shanks et al. (2003) compiled available information on the dispersal distance by currents of the propagules of benthic marine organisms, and found a significant positive correlation between the duration of propagules in plankton and the dispersal distance, which ranged from minutes to months and meters to thousands of kilometers, respectively.

In freshwater, zooplankton may be transported among systems during periods of overflow (Michels et al. 2001). Short-distance transport of resistant diapausing eggs in fish stomachs is also potentially possible (Jarnagin et al. 2000), as well as by terrestrial animals (Maguire 1963), but the significance of these vectors in nature is likely limited. However, fish guts as vector of dispersal of diapausing invertebrates can be important if coupled with human-mediated introductions of fish. For instance, ephippia of *Daphnia lumholtzi* might have been introduced first to one of the lakes of the southern USA in 1983 with the intentionally introduced Nile perch (*Lates niloticus*) from Lake Victoria, Kenya (Havel and Hebert 1993).

Vertebrates have also long been considered to be a primary vector of dispersal for aquatic invertebrates (Darwin 1859). Transfer of resting eggs by waterfowl can be considered a more effective vector of dispersal of invertebrates in inland waters (Bohonak & Jenkins 2003); however, direct evidence of its importance is also limited (but see Figuerola et al. 2005). Eggs may be dispersed by birds with ingested food (Charalambidou et al. 2003), by sticking to their legs and bills, within the plumage. A recent review by Figuerola and Green (2002) showed that bird-mediated transport of propagules of aquatic invertebrates is a frequent process, but limited to local spatial scales. Low resistance to desiccation of some resting stages (Fell 1998) may limit distance of dispersal of diapausing aquatic invertebrates by waterbirds (Figuerola & Green 2002). However, studies of genetic distributions of some zooplankton species along with analysis of major waterfowl flyways suggests a potentially significant role of birds in long-distance intracontinental dispersal of some cladocerans and bryozoans (Taylor et al. 1998; Freeland et al. 2000a, b).

Assumptions of the significant role of dispersal of resting stages of freshwater invertebrates by natural factors were recently criticized in the review by Bohonak and Jenkins (2003), which suggested that genetic and direct experimental studies failed to demonstrate evidence of effective passive dispersal, specifically by wind (Jenkins 1995; Jenkins & Underwood 1998). Clearly, zooplankton are not uniformly "good" dispersers. Rather, species both differ in their dispersal ability and the primary vectors by which they are dispersed (Jenkins 1995; Jenkins & Buikema 1998).

12.2.2 *Human-mediated Dispersal*

Human-mediated dispersal vectors (introductions) are broadly classified into two main categories: deliberate or intentional introductions and unintentional introduction. Regarding the first type of vector, Alekseev (1986) suggested that intentional transfers of crustaceans in latitudinal directions are hindered by genetically fixed differences in timing of diapause, and successful acclimatization is more likely for crustaceans transferred after completion of diapause. In some cases, intentional long-distance transfer of target aquatic organisms for stocking purposes can be coupled

with unintentional introductions of other organisms possessing diapause (see case study of *Daphnia lumholtzi* in section 12.2.1). Recreational and commercial boating has been shown to be an important vector for both short- and long-distance dispersals of aquatic invertebrates and their diapausing stages for inland waters of North America (Buchan & Padilla 1999; Havel & Stelzleni-Schwent 2000; Johnson et al. 2001). Some intercontinental transfers of diapausing eggs of aquatic invertebrates and their subsequent invasions of inland waters have been linked to the introduction of commercially useful plants (McKenzie & Moroni 1986), importation of industrial equipment (see review by Havel & Shurin 2004), and even transportation of military amphibian vehicles as has been suggested by Flössner and Kraus (1976) in the case of *Daphnia parvula* Fordyce (Cladocera: Daphnidae) accidental introduction in European inland waters from North America.

However, at present the majority of biological invasions into coastal and even inland waters worldwide can be linked to unintentional introductions via different shipping-related vectors: construction of canals, ship's hull fouling, and ballast water release. Ballast water of ships is a principal vector of global long-distance transfer of aquatic invertebrates and their resistant resting stages, which readily breaches geographic barriers to dispersal and gene flow (Carlton & Geller 1993). Results of several ballast water studies indicated high biological diversity of aquatic communities within ballast water; these communities were often dominated by crustacean taxa (Carlton & Geller 1993; Gollasch et al. 2000; Gollasch et al. 2002). It is likely that harsh conditions in ship's ballast tanks (darkness and rapid changes in water temperature) may result in induction of embryonic, larval, or even adult diapause in some crustaceans, and thus facilitate their survival during ship journey (Panov et al. 2004).

Resting eggs and cysts of algae and aquatic invertebrates have been frequently reported in ballast tank sediments. These sediments vary from a few centimeters to more than 30 cm depth (Hamer 2002), resembling the upper layers of lake, estuarine or sea-bottom sediments. Bailey et al. (2003) recorded the presence of invertebrate diapausing eggs in residual sediments from transoceanic vessels and experimentally studied viability of the eggs collected from ballast tanks on vessels operating on the North American Great Lakes. In this study, 17 cladoceran, copepod, and rotifer taxa hatched from these sediments have been identified, demonstrating that diapausing eggs in sediments may survive treatment of ballast tanks with oceanic water and could potentially hatch in dark ballast tanks if freshwater were added.

It is also likely that the observed trend of increasing role of crustacean invaders from marine to freshwater ecosystems can be a result of high susceptibility of the latter to invasions of cladocerans and copepods, e.g. taxa possessing strong diapause (Panov et al. 2004). This phenomenon may favor the hypothesis that the evolutionary appearance of embryonic diapause in crustaceans may facilitate the penetration of marine crustaceans into inland waters (Hairston & Cáceres 1996; Hairston & Bohonak 1998).

In an assessment model of the risk of future introductions of aquatic species with ballast waters into the Great Lakes, considering species' invasions histories, shipping traffic patterns, and physicochemical factors that constrain species survivorship during ballast-mediated transport, Grigorovich et al. (2003) identified 26 high-risk

species. Among them 24 species belong to crustaceans, with more than half the species possessing embryonal, larval or adult diapause (seven species of Cladocera and six species of Copepoda). The authors suggested that ability to possess diapause, and/or parthenogenetic reproduction and short generation time in these taxa, already introduced into the Great Lakes with ballast water, have fostered their survival during ballast-mediated transfer and ensured rapid population growth in the recipient ecosystems.

The results of the genetic analysis of European and North American lineages of freshwater cladocerans indicate that human-mediated vectors of dispersal may affect extraordinarily rates of intercontinental species dispersal: the current rates of species invasions are nearly 50,000 times higher than historical levels (Hebert & Cristescu 2002). It is important that most of these recent human-mediated intercontinental invasions originated, most likely, from transport of diapausing resting stages in ballast tanks of ships as in the recent case studies for cercopagid cladocerans *Bythotrephes longimanus* (Cladocera: Cercopagidae) and *Cercopagis pengoi* (Cladocera: Cercopagidae), both invasive species possessing prolonged embryonic diapause as adaptation to dispersal and invasion success (Panov et al. 2004).

These two predatory cladocerans have a complicated invasion history both in European and North American inland waters, mediated by the multiple long- and short-distance dispersal vectors (see Panov et al. 2004 for review). Most probably, *Bythotrephes* and *Cercopagis* were transferred from Europe to America with the ballast water of cargo vessels, presumably as diapausing resting eggs (Lehman 1987; MacIsaac et al. 1999). Genetic studies revealed that in both cases the eastern Baltic Sea basin (Lake Ladoga and Neva River estuary) served as a source area of the initial invasion to North American Great Lakes, indicating presence of the invasion corridor between eastern Baltic and Lawrentian Great Lakes (Cristescu et al. 2001; Berg et al. 2002).

In North America *Bythotrephes* is mainly spread by boaters and anglers attached to equipment such as fouled fishing lines, boat anchor lines, downrigger cables, via infected bilge water and live well water, and live minnow bait, which contain females bearing resting eggs (Jarnagin et al. 2000; MacIsaac et al. 2004). MacIsaac et al. (2004) revealed that species spread occurred via a combination of dominant, local diffusion (median distance 12.5 km) and rare, long-distance (>100 km) dispersal. For example, one invaded lake (Muskoka Lake, Ontario) apparently served as an invasion "hub," resulting in up to 18 additional direct and 17 indirect invasions (MacIsaac et al. 2004).

Rapid short-distance dispersal of invasive cladocerans is likely facilitated by changes in their reproductive strategy (Fig. 12.1). As most freshwater Cladocera, in native habitats *Bythotrephes* usually switches from parthenogenetic to gamogenetic reproduction at the end of a season and even then the densities of males and females with resting eggs are relatively low (Straile & Hälbich 2000). However, in new invaded habitats this species showed prolonged embryonic diapause. For instance, during some years in the eastern basin of Lake Erie, Ontario, Canada, males and ephippial females of *Bythotrephes* appeared in midsummer (Garton et al. 1993). The most remarkable changes in sexual reproduction of this species were noted in
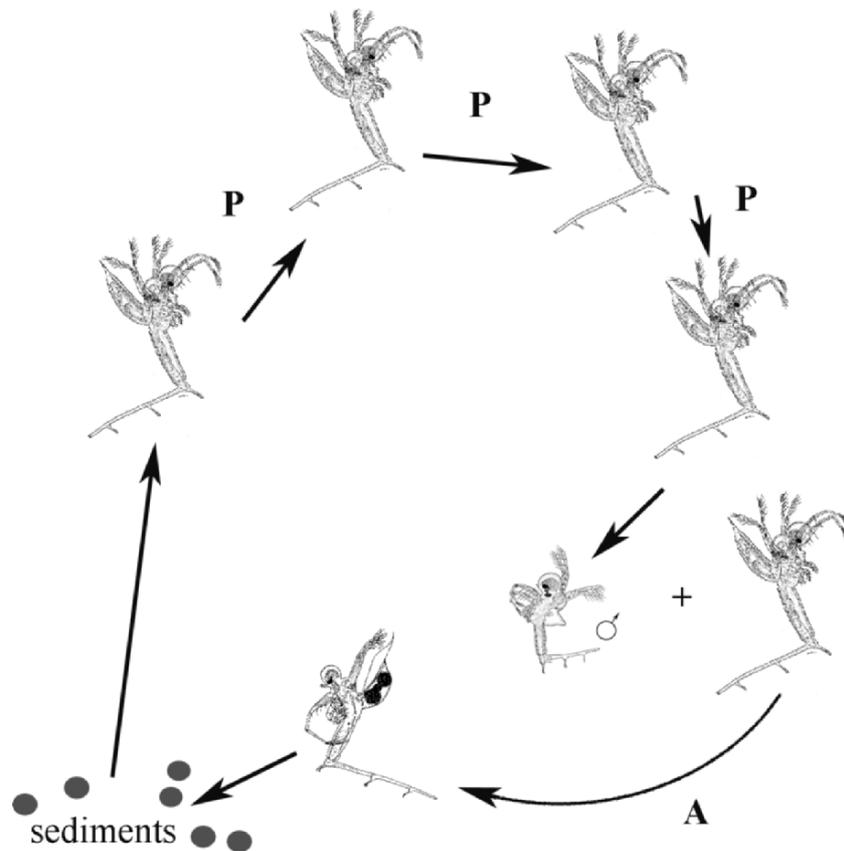
Figure 12.1. Heterogonic reproductive cycle in Cladocera: parthenogenetic vs amphigonic reproduction.

Harp Lake, Ontario, Canada, where *Bythotrephes* appeared in the early 1990s. In 1994 and 1995, females with resting eggs were noted from July, and by the end of August 50–80% of females carried resting eggs (Yan and Pawson 1998; Yan et al. 1992; N. D. Yan and T. W. Pawson, personal communication). Later, in 1998, the Harp Lake population returned to a more typical parthenogenetic mode of reproduction during summer: first females with resting eggs were recorded in late August (Yan et al. 2001).

Like invading *Bythotrephes* in Harp Lake in the first 2 years after its first record, a *C. pengoi* population newly established in the easternmost Baltic Sea (Neva estuary) also showed this remarkable reproductive strategy, producing a large number of resting eggs during summer months during the first years after the invasion (Krylov & Panov 1998). As for *Bythotrephes* in Harp Lake, the mean seasonal percentage of both males and gamogenetic females in *Cercopagis* population in the Neva estuary gradually declined during following years after invasion (Panov et al. 2004).

It has been suggested that the large pool of resting eggs in the Neva estuary *Cercopagis* population has enabled this species *C. pengoi* to achieve fast population growth in new environments, and an increasing risk of *C. pengoi* being dispersed by ships' ballast water (Panov et al. 1996; Panov et al. 1999). In summer 1998, soon after its establishment in the eastern Baltic, *C. pengoi* was first found in the North American Great Lakes (MacIsaac et al. 1999; see also above). Populations of *C. pengoi* in Lake Ontario during first years after invasion also possessed midsummer sexual reproduction (Grigorovich et al. 2000; Makarewicz et al. 2001), characteristic for the source population in the Baltic Sea (Neva estuary), and for *Bythotrephes* in Harp Lake (see above). Despite existing potential of waterfowl to transfer resting eggs of *Cercopagis*, boaters and ballast water of ships are considered as primary vectors of *C. pengoi* dispersal in the Great Lakes area (Makarewicz et al. 2001). Potential for dispersal with fishing equipment for *Cercopagis* is even higher, than for *Bythotrephes*, because of the specific morphological feature of its caudal appendage, which is longer and possesses a terminal loop (this feature is reflected in North American common name of *C. pengoi*: "fishhook waterflea").

It is important to note that the invasion of *C. pengoi* to the Laurentian Great Lakes has been taken place after implementation of ballast water management options for the ships entering Great Lakes, namely exchange of ballast water in open ocean, which is considered to be an effective measure to decrease risk of transfer of fresh-water organisms. However, as has been shown by Bailey et al. (2003), resting eggs of freshwater invertebrates may hatch from the ballast water sediments, even those that have previously been exposed to salt water. Thus, the example of *C. pengoi* invasion to North America demonstrates the limited effectiveness of ballast water exchange programs in preventing introductions of aquatic invertebrates producing resting eggs, which may accumulate in sediments of ballast tanks (MacIsaac et al. 1999).

Enhanced gamogenetic reproduction (prolonged embryonic diapause) during first years after invasion was also recorded in *D. parvula* (Riccardi et al. 2004), the North American species most likely introduced in European inland waters as ephippia by such specific long-distance human-mediated dispersal vectors as military amphibian vehicles (Flössner & Kraus 1976). According to Flössner (2000), both natural (waterbirds and surface water connections) and human-mediated vectors (e.g. transport and introduction of fish fingerlings; water transport related to the excavation of artificial basins and canals) are responsible for the quick dispersal of this species from southern Germany to other European locations.

12.3 CONCLUSIONS: GENERALIZED MODEL OF DISPERSAL OF AQUATIC INVERTEBRATES WITH PROLONGED DIAPAUSE

The case studies discussed earlier about successful invaders into inland waters of Europe and North America suggest the possible importance of prolonged (embryonic) diapause for effective short- and long-distance (both inter- and intracontinental) dispersals of aquatic invertebrates. These invasive cladocerans were transferred across geographic barriers (Atlantic Ocean) by different human-mediated vectors possibly by means of their diapausing eggs, exposed to adverse conditions during

transfer, and survived even possible ballast water management options (in case of *C. pengoi*) and quarantine treatment (in case of *D. lumholtzi*). Their following rapid short-distance transfer and, less frequent, long-distance transfer by mainly multiple human-mediated vectors might be attributed to their life-cycle patterns in "infected" ecosystems: rapid development of a large pool of diapausing eggs in populations, and, in some cases, even switching from "normal" pattern with short period of gamogenetic reproduction after a prolonged period of parthenogenetic reproduction, to the early and prolonged gamogenetic reproduction. A generalized conceptual model of dispersal patterns in aquatic invertebrates with such a reproduction strategy, involving a high level of development of resistant diapausing resting eggs, is represented in Fig. 12.2. Our model can be considered a variant of the human-vectored invasion model, initially suggested by MacIsaac et al. (2001) (dispersal is determined by the probability of propagule movement by humans from the source to the recipient site), with incorporation of adaptive reproduction strategies in invasive species, which increases the probability of dispersal and successful establishment in the novel ecosystem.

   Rapid development of large pools of resting eggs in bottom sediments might facilitate invasion success of these species, and their rapid integration into local plankton communities. Diapausing eggs allow an escape from competition (Cáceres 1997), which is likely severe for most invading species. This refuge from competition in the sediment egg bank likely facilitates the rapid establishment of invasive species (Panov et al. 2004). There is also evidence for evolution of life-history traits of invasive microcrustaceans with respect to diapause: while the initial colonizing population appears to possess early prolonged production of diapausing eggs, this characteristic erodes over time. Although in some cases a midsummer switch toward sexual reproduction in Cladocera can be considered to facilitate dispersal by natural factors in marine ecosystems (see above), early prolonged production of resting eggs recorded in *B. longimanus*, *C. pengoi*, and *D. parvula* during first years after their
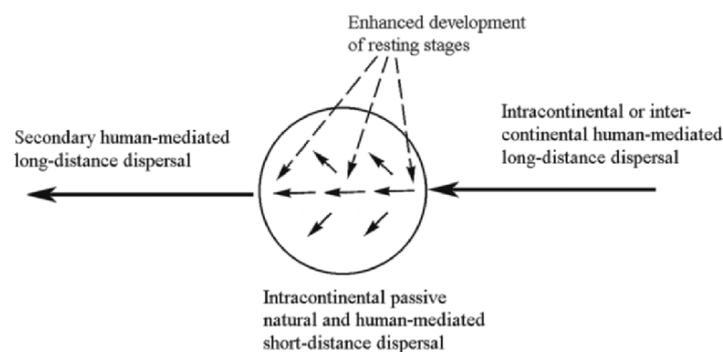

Figure 12.2. Generalized model of dispersal of aquatic invertebrates with prolonged diapause. (After Panov et al. 2004.)

invasion into some lake and/or estuarine ecosystems and following fast "erosion" of this phenomenon in *B. longimanus* and *C. pengoi* can be attributed, most likely, to rapid microevolution processes characteristic for these polymorphic species in highly variable environments.

As we suggested earlier for *C. pengoi* in the Neva estuary (Krylov & Panov 1998), switch to prolonged period of sexual reproduction in the introduced population can be attributed to some kind of the effect of "founder population." The probability of clones with an extended gamogenetic phase being pumped into ships' ballast tank is much higher than for the strains possessing the more "normal" life cycle with a comparatively short period of sexual reproduction. Fast erosion of this phenomenon in *Bythotrephes* in Harp Lake and *Cercopagis* in Neva estuary most likely related to fast evolution of the life cycle of invading species under strong selection. In some cases, this was most likely from invader-selective fish predation: by lake herring *Coregonus artedii* in Harp Lake (Coulas et al. 1998) and by the Baltic herring *Clupea harengus membras* in the Gulf of Finland (Antsulevich & Välipakka 2000).

Fast dispersal and successful establishment of several exotic zooplankton species in inland waters of Europe and North America demonstrate the high potential for dispersal in aquatic invertebrates that develop resistant propagules. Colonization ability may also be facilitated by the ability of invertebrates with prolonged diapause to "travel in time" for persisting through adverse environmental conditions and to use the "storage effect" of egg banks to avoid competition with native species or clones. Our review suggests that diapause in some taxa of aquatic invertebrates might play a crucial role in their dispersal and colonization success in recipient ecosystems, with human-mediated vectors of dispersal acting as a powerful selective force. Human-related selection factors may facilitate dispersal of species with a high level of gamogenetic reproduction, while natural selection in novel habitats may result in fast erosion of this "founder population" effect toward "normal" life cycle with a prolonged period of parthenogenetic reproduction.