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ARTICLE

Movement patterns and rheoreaction of larvae of a fluvial specialist (nase, *Chondrostoma nasus*): the role of active versus passive components of behaviour in dispersal[1]

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Abstract: The dispersal of fish larvae in rivers might result from water movement but also from larval behaviour. Although potentially crucial for dispersion, knowledge of the role of behaviour is still fragmentary. This study intends to contribute to the question of how riverine fish larvae drift or move. All dispersal-relevant movement patterns of larvae of a characteristic rheophilic species were analyzed based on the parameters (*i*) swimming activity, (*ii*) direction of movement, and (*iii*) the orientation towards the current vector. Experiments were conducted in a novel flume mesocosm at three different flow scenarios covering the current velocity range of natural habitats. Mean current velocities in these scenarios were under, near, and over the "critical current velocity", above which fish larvae are not able to constantly hold their position in the water column. Three consecutive larval stages were tested to account for possible ontogenetic shifts in movement behaviour, both during the day and at night. Our results strongly suggest that the assumption of mainly passively drifting larvae has to be refused; in total, 92.6% of all observed movement events were characterized by swimming activity and directed orientation, whereas only 7.4% could be assigned to passive drift. During downstream movement, a significant portion of movement events (57.1%) was attributed to larvae that orientated in an upstream direction and performed active swimming movements.

Résumé : La dispersion des larves de poisson dans les rivières peut être le résultat du mouvement de l'eau, mais également du comportement des larves. Les connaissances sur le rôle du comportement demeurent fragmentaires, malgré l'importance potentiellement cruciale de ce dernier pour la dispersion. L'étude s'intéresse à savoir comment les larves de poissons de rivière dérivent ou se déplacent. Tous les motifs de déplacement pertinents pour la dispersion des larves d'une espèce rhéophile caractéristique ont été analysés en fonction des paramètres suivants : (*i*) l'activité natatoire, (*ii*) la direction des déplacements et (*iii*) l'orientation par rapport au vecteur de courant. Des expériences ont été menées dans un mésocosme en canal novateur pour trois scénarios d'écoulement différents couvrant la fourchette de vitesses du courant dans les habitats naturels. Les vitesses moyennes du courant dans ces scénarios étaient inférieures, semblables ou supérieures à la « vitesse critique du courant » au-delà de laquelle les larves de poisson ne peuvent maintenir constamment leur position dans la colonne d'eau. Trois étapes larvaires consécutives ont été étudiées pour tenir compte de possibles changements ontogéniques du comportement de déplacement, tant durant le jour que la nuit. Nos résultats donnent fortement à penser que l'hypothèse des larves dérivant principalement de manière passive doit être rejetée; au total, 92,6 % de tous les évènements de déplacement observés étaient caractérisés par une activité natatoire et une orientation dirigée, alors que seuls 7,4 % de ces évènements pouvaient être attribués à une dérive passive. Durant les déplacements vers l'aval, une portion significative des évènements de déplacement (57,1 %) était attribuable à des larves qui s'orientaient vers l'amont et effectuaient des mouvements de nage active. [Traduit par la Rédaction]

Introduction

Dispersal of fish larvae in lotic environments (streams, rivers, estuaries, and marine habitats) was often assumed to be a primary consequence of water movement (Wolter and Sukhodolov 2008). The suspected dominance of water flow led many modellers to make a "simplifying assumption" by treating larvae as passive particles. Nonetheless, a growing number of studies demonstrate that other factors beside the movement of water may be responsible for dispersal outcomes in fish larvae (Leis 2007). It has been shown that fish can detect fine-scale stimuli induced by the current and actively react to them from early larval stages onwards (Garner 1999; Stoll and Beeck 2012). In marine fish ecology, the importance of larval behaviour for dispersal has been recognized during the past decade (Fiksen et al. 2007; Gallego et al. 2007; Leis 2007), and the need to break "the behavioural black box" has been realized (Pineda et al. 2007). Moreover, basic behavioural features of fish larvae have already been successfully incorporated into elaborate 3D models of physical–biological interactions, which have increasingly become an integral tool for understanding larval fish dynamics in the sea (Gallego et al. 2007), while in rivers

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these methods are in their infancy (Schludermann et al. 2012; Lechner et al. 2016). It has been recognized that certain "long-term" behavioural changes may considerably contribute to larval dispersion — these factors involve environmental stimuli such as odours, sounds and light, time of day, water temperature, salinity, food availability, or ontogeny — and act on time scales of hours, days, and weeks (Pineda et al. 2007). Minimal knowledge, however, exists on behavioural adaptation operating on scales of seconds to minutes, which responds to temporary "short-term" physical and biological triggers (Pineda et al. 2007) or to factors that change rapidly in space (e.g., flow conditions, rheogradients) as fish larvae move.

Lechner et al. (2016) stated that the behavioural mode of drift or movement is rarely specified in field studies, and only very few studies sought to investigate movement behaviour in flume experiments. The behavioural mode and associated pattern of movement, however, is likely to ultimately determine the actual swimming trajectory, travel speed, and destination of dispersing fish larvae in rivers. Knowledge about these features is therefore indispensable when attempting to understand, model, or predict dispersal patterns of fish larvae in rivers and all other kinds of moving waters (Lechner et al. 2016). The reduction of characteristic and native riverine fish fauna mainly due to pervasive habitat alteration is a worldwide phenomenon that has been acknowledged for decades (Schiemer and Spindler 1989), but is still ongoing (Moyle and Mount 2007). A greater understanding of dispersal processes will contribute to our understanding of recruitment and will certainly aid future conservation measures targeting the growing number of endangered riverine fish populations (Cooke et al. 2012; Humphries et al. 2013; Lechner et al. 2016). Well-performing models of the migration of fish larvae from spawning to nursery areas could present an invaluable tool in designing effective river restoration measures in the 21st century. The scope of this study is to gain insights into behavioral aspects which we consider "basic input variables" for future modelling.

Our work is based on the fundamental concept of Pavlov et al. (2008), who proposed three different types of downstream dispersal of riverine fish larvae based on swimming activity and orientation of larvae: passive drift (P), active downstream (Ad) movement, and an intermediate type, active–passive (Ap) downstream movement. Beyond these, active upstream (Au) and lateral movements are also assessed in the present study, and the effects on displacement distances are evaluated.

It is our objective to provide a quantitative estimation of all dispersal-relevant patterns of larval fish movement and displacement distances, reflected as a consequence of behaviour (i.e., swimming activity, direction, and orientation). The frequencies of distinct movement patterns are presented for different flow conditions, larval stages, and for day and night based on the study of single individuals. Current velocity is a major factor for the distribution of fish larvae in rivers. Highest abundances of young fish typically occur in low-flowing inshore areas that offer average flow velocities lower than their maximal sustainable swimming performance (Schiemer et al. 2002). In this study, we cover flow conditions under, near, and over this critical flow velocity (v_{crit}). Furthermore, we describe a novel but relatively simple experimental approach for observing, assessing, and analyzing movement patterns and displacement in riverine fish larvae.

Materials and methods

Model organism

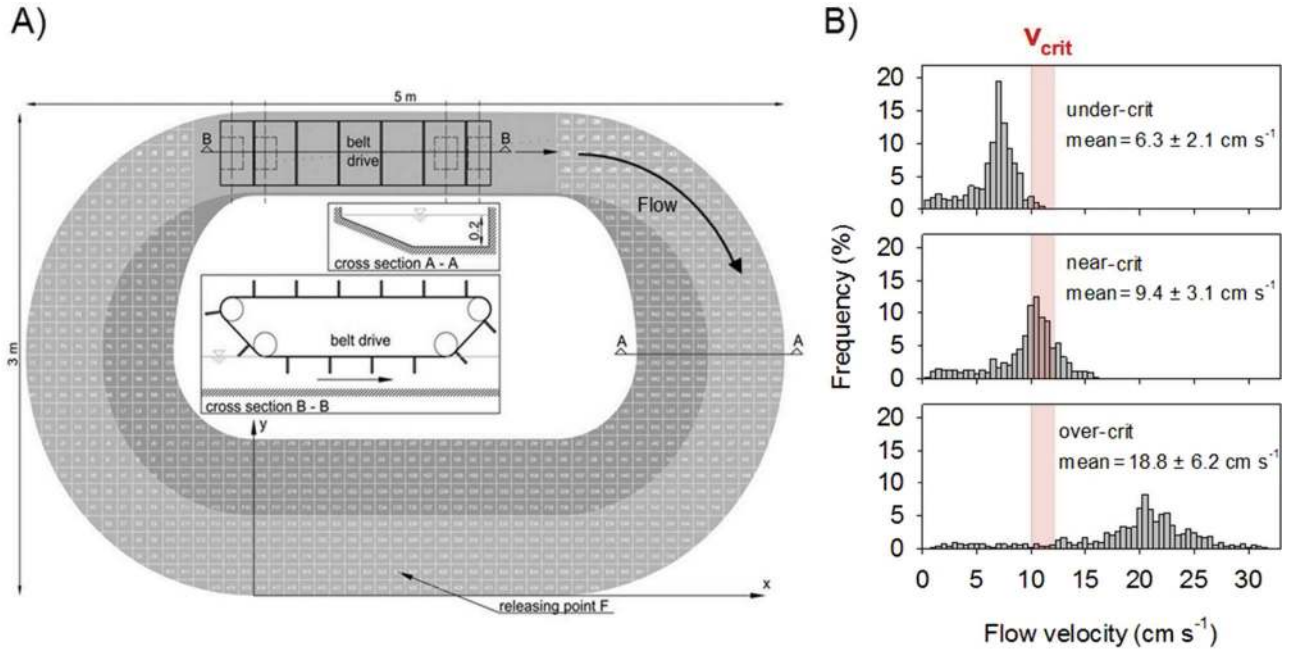
In this study we used lab-reared larvae of nase (*Chondrostoma nasus*, Cyprinidae), ranging from 23 to 43 days posthatch (dph) and from 12.1 to 17.4 mm in total length (TL). The nase is a widespread but increasingly endangered riverine cyprinid inhabiting moderate to fast-flowing rivers of Central and Eastern Europe (Szabó et al. 2002; Kottelat and Freyhof 2007). Adults undertake spawning migrations into smaller tributaries or to gravel bars with swift currents for up to several dozen kilometres (Keckeis et al. 1996; Peňáz 1996). After the period of yolk-sac depletion during which larvae exhibit a benthic life style (usually 4 to 7 dph; Keckeis et al. 1996; Kamler et al. 1998), larvae and early juveniles commonly disperse downstream from spawning sites to low-flowing inshore areas where they feed on small invertebrates (Kamler and Keckeis 2000; Reichard et al. 2001).

Ripe nase were obtained from a traditional spawning site in the Schwechat River located ~9 km upstream of the confluence with the Danube River east of Vienna, Austria. Eggs and sperm were stripped and fertilized artificially. Incubation and rearing took place in a rectangular 150 L flow-through tank with constant supply of filtered and well-aerated tap water at a temperature (mean ± standard deviation, SD) of 11.7 ± 0.7 °C. Larvae were fed with live nauplii of *Artemia salina* (Great Salt Lake Artemia Cysts, Sanders) and powdered dry food (Vipagran Baby, Sera) ad libitum. We tested three consecutive developmental larval stages (according to Peňáz 1974) to investigate dispersal-relevant ontogenetic shifts related to swimming performance or behavioural attributes: (*i*) second stage (L2) with a mean (±SD) TL of 12.9 ± 0.4 mm (age: 23 to 33 dph), (*ii*) third stage (L3) with TL of 14.7 ± 0.6 mm (27 to 40 dph), and (*iii*) fourth stage (L4) with TL of 16.4 ± 0.5 mm (36 to 43 dph). Treatment of parental fish and larvae was carried out in full accordance with the guidelines provided by the Canadian Council on Animal Care (CCAC 2005) and the Austrian law of animal care (BGBl. II Nr. 486/2004).

Mesocosm, experimental setup, and procedures

We used an oval-shaped racetrack flume (Fig. 1A) in which a paddled belt drive actuated a fully adjustable clockwise water flow. Our mesocosm featured basic properties of a natural river's morphology; the inner bank was characterized by a slope of 22°, resembling a point bar in a river, whereas the outer bank was designed vertically to correspond to the natural situation of a cut bank. This design induced the formation of gradients in water current velocity (ranging from nearly zero to 32 cm·s^{-1} depending on the flow scenario; Fig. 1B) and available depths (ranging from 0 to 20 cm). Experiments were conducted during daylight (and additional artificial light in the lab) between 1000 and 1900 h. We used three different flow scenarios that were chosen based on the v_{crit} of the larvae. v_{crit} refers to the size-dependent "maximum sustainable water velocity" according to Flore et al. (2001) above which fish larvae are not able to hold their position in the water column for longer than 2 min. Flow scenarios in our experiments were (*i*) "under-critical" with mean (±SD) flow velocity (*U*) of 6.3 ± 2.1 cm·s^{-1}, (*ii*) "near-critical" with *U* of 9.4 ± 3.1 cm·s^{-1}, and (*iii*) "over-critical" with *U* of 18.8 ± 6.2 cm s^{-1}. Mean values (±SD) of v_{crit} for L2, L3, and L4 were 10.3 ± 0.2, 11.1 ± 0.3 and 11.9 ± 0.2 cm·s^{-1}, respectively. Additionally, experiments were conducted in darkness from 2030 to 2345 h at the under-critical flow scenario. For each experimental flow scenario, a fine-scale 3D current vector field was calculated based on a network of 1996 measurements using acoustic Doppler velocimetry. As a result, highly accurate information on flow velocity and (3D) direction of current vectors could be obtained for any point within the flume. To determine the position of the studied fish larva, we used an observation grid with squares of 10 cm length and an alphanumeric code that was drawn on the bottom of the flume. Larvae were released at two different release points: one was situated in a shallow (depth = 11 cm at the centroid of the grid square) low-flow zone close to the inner bank with *U* of 2.8, 4.2, and 8.4 cm·s^{-1}, depending on the flow scenario. The other release point was situated in deep water (depth = 20 cm) with higher *U* of 9.0, 13.1, and 26.3 cm·s^{-1}, respectively. The flume was filled with fresh, well-oxygenized (saturation 95% ± 4%) tap water at 13.9 ± 1.8 °C at the beginning of each day or night of experimental work. Water temperature and oxygen saturation was measured to the nearest 0.1° C and 1% using

Fig. 1. (A) Schematic diagram of the flume mesocosm illustrating the dimensions, shape, and functional principle. The inner bank was designed with a slope (dark gray area), while the outer bank was arranged vertically. A clockwise flow was induced by a paddled belt drive. Maximum depth was 20 cm (light gray area). (B) Frequency distributions of available flow velocities in the three experimental flow scenarios. The red-shaded area indicates the size-dependent critical current velocity (v_{crit}) of fish larvae used in this study. Mean flow conditions in these flow scenarios were under-critical, near-critical, and over-critical, respectively. Note that in all scenarios, low-flowing zones were available. [Colour online.]



an oximeter (Oxi 330, WTW) at the beginning and end of each set of experiments.

Before release, each larva was adapted in a transparent acrylic glass cup with a 400 μm mesh bottom for a few minutes in the flume. Larvae were released individually and filmed with a hand-held video camera (Sony, HDR-CX700VE) over a period of 5 min. The observer was always positioned in the center of the flume to keep an adequate distance to the studied fish larva and a perpendicular angle of view to the outer border of the flume. During daylight, the automatic focus function was used, whereas in darkness we used the infrared detectable night-shot function together with an additional infrared light source (Sony, HVL-HIRL) and focused manually. After the filming period, the studied fish larva was euthanized using a highly overdosed (~0.2%) solution of Tricaine (MS-222, Fulka Analytical), individually labeled, and preserved in 4% formalin for later size measurement and stage identification.

Determined variables, movement patterns, and displacement

Videos were processed manually using the software package Adobe Premiere Pro 5.0/CS5. The position was obtained from the observation grid and noted in a continuous timetable (intervals of 0.1 s) every time the larva has passed over a grid line with the full length of its body. The travelled distance between two consecutively passed-through grid squares was calculated from centroid to centroid of these squares. Displacement was calculated as

$$\text{Disp} = \text{DMU} - \text{DMD} \quad (1)$$

where Disp is the displacement, DMU is the distance (m) moved upstream, and DMD is the distance (m) moved downstream during the observation period of 5 min.

Consequently, the displacement is shown as positive for larvae that demonstrated net upstream movement and negative for larvae showing net downstream movement over the 5 min observation period.

To determine the actual speed of the fish towards stationary landmarks (V_s) between two points of observation, the travelled distance was divided by the residence time in the respective square. For each event of downstream movement, the proper movement rate (relative to water flow; V_f) was calculated according to Pavlov et al. (2008) as

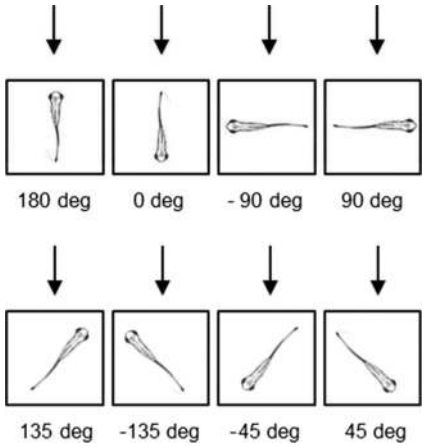
$$V_f = V_s - U \quad (2)$$

where U is the flow velocity within the respective grid square.

V_f can attain any value: positive (if the fish moves faster than the surrounding water), negative (fish moves slower than surrounding water), or zero (fish speed equals flow velocity). Hence, V_f can be regarded as an expression for a fish's swimming activity. Both negative and positive values indicate that propulsive movements are carried out, the intensity of propulsion being reflected in the magnitude of V_f. The orientation of the larva was visually assigned to one of eight categories depending on the approximated angle enclosed by the fish's body axis and the longitudinal gridline. The following categories of orientation were defined: –45°, –90°, –135°, 180°, 135°, 90°, 45°, and 0° in either clockwise (negative values) or counterclockwise (positive values) rotation. The angle of the fish's body axis enclosed with the 2D current vector was then calculated for any point of observation and assigned to the same categories of orientation (Fig. 2). When the orientation of the fish larva could not be assigned to one of the eight categories with certainty, this observation was omitted from later analysis of movement patterns.

Differentiation of movement patterns (mP) was achieved using an appropriate IF-function in MS-Excel based on the following factors: (*i*) direction of movement relative to the flow direction in terms of upstream, lateral, and downstream, (*ii*) the orientation

Fig. 2. Scheme of the eight different categories of orientation towards the current vector (black arrows). The angle enclosed between the virtual body axis and the current vector was visually assigned to 180°, 135°, –135°, 90°, –90°, 45°, –45°, or 0°.



of larvae against the current vector in terms of the categories described above, and (*iii*) the value of V_f. The explicit criteria are provided in Table 1.

Data analysis

For each individual larva, the proportions of each mP out of the total number of observed movement events were calculated. One movement event was defined as the passage of a grid square of defined size, so the calculated proportions of mP correspond to the proportions of travelled distances. The proportion data were arc-sine-square-root-transformed according to McCune and Grace (2002) for any statistics applied. General linear models (GLM) were used to determine if the factors larval stage, flow scenario, release point, and light level had a significant effect on the proportions of distinct mP. In case of significance, these factors were treated as separate categories for further statistical analysis. Otherwise, data were pooled for factors for which GLM did not reveal significant effects. To test for differences among proportions of different mP within distinct larval stages, flow scenarios, and light levels, bootstrap ANOVA (1000 resamples) and Bonferroni post-hoc tests were used. Significance was accepted at p values equal to or lower than 0.05 for any statistical test applied. A nonlinear regressions of the form $y = a \times e^{bx}$ was fitted to describe the relationship between displacement and U_{mean} of fish larvae. Statistical analyses were conducted using PASW Statistics 20.0 (SPSS Inc., Chicago). Plots and regressions were generated with SigmaPlot 12.5 (Systat Software, San Jose).

Results

Experimental factors influencing the proportions of distinct mP

GLMs showed evidence that the factors larval stage, flow scenario, and light (day–night) were significant predictors for the proportions of distinct mP (all F and p values are provided in Table 2). The flow scenario was a significant factor for the proportions of most mP, significance was detected for active upstream (Au), active–passive downstream (Ap), passive downstream (P), traversing (T), and the cumulative proportion of all downstream movements (dmP). Stage and light were significant factors for Ap and T. The factor release, however, was not found to significantly affect the proportions of any mP, and data from both release points were pooled for further analysis. Comparisons among the proportions of distinct mP were thus made within each category of stage, flow scenario, and light (day–night).

Upstream, lateral, and downstream movement

Larvae of all stages were mainly moving upstream (Au) at under-critical flow conditions during day; frequencies of Au were higher than those of any other mP, but these differences were significant only in L4 larvae ($p < 0.05$; Fig. 3). At night (at under-critical flow conditions), frequencies of Au were on average higher than those of any other mP in L2 and L4 larvae; however, significance could be detected only in L4 ($p < 0.05$). Mean frequencies of lateral movement (L) ranged between 8.5% ± 7.8% (L2–near-crit–day) and 31.9% ± 15.1% (L3–near-crit–day). Less surprisingly, larvae of all stages moved mainly downstream at the over-critical flow scenario. This dominance was significant in L2 ($p < 0.001$) and L4 ($p < 0.01$). At the over-critical flow scenario, means of the cumulative proportions of all downstream movement patterns together (dmP) accounted for 71.0% ± 17.0% in L2, 49.2% ± 27.6% in L3, and 62.0% ± 22.5% in L4 larvae.

Downstream movement patterns

In general, the use of downstream movement patterns that incorporate swimming activity and directed orientation were clearly dominant over totally passive movement (P; Fig. 4.). Larvae of all stages were predominantly using Ap at all tested flow scenarios both during day and night. The differences between Ap and all other dmP were highly significant ($p < 0.001$) in all stages at the over-critical flow scenario. At the near-critical flow scenario, these differences were highly significant for L2 and L4 larvae ($p < 0.001$), while at the under-critical flow scenario, frequencies of Ap were significantly higher than all other mP in L3 and L4 larvae ($p < 0.05$). Mean proportions of Ap ranged between 45.2% ± 26.8% (L2–under-crit–night) and 74.7% ± 20.0% (L3–over-crit–day). Among the proportions of A, T, and P, no significant differences could be observed (all $p > 0.05$).

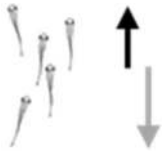
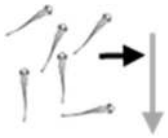
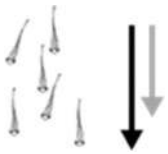
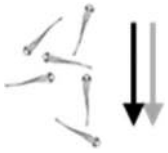
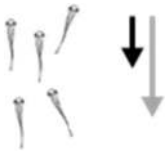
Displacement

The consequences of high proportions of Au movement and Ap downstream movement were clearly reflected in the calculated displacement distances of different larval stages at different prevailing mean flow velocity conditions (Fig. 5). At under-critical mean flow velocities ($U_{mean} < 10$ cm·s^{-1}), the displacement distance within 5 min ranged between 8.3 m upstream and –10.5 m downstream from the release point for all larval stages. At clearly over-critical mean flow velocities ($U_{mean} > 14$ cm·s^{-1}), displacement within 5 min was exclusively directed downstream and ranged from –5.0 to –54.3 m from the release point. It was apparent, however, that observed larval downstream displacement at over-critical values of U_{mean} was markedly lower than hypothetical P displacement by the current alone (U_{mean} extrapolated to 5 min). On average, L2 larvae would have reduced downstream displacement by approximately 58%, L3 larvae by 82%, and L4 larvae by 77% when compared with the simplifying assumption of totally passive displacement at U_{mean} values greater than the maximum of v_{crit} (12.3 cm·s^{-1}).

Discussion

To our best knowledge, this study presents the first that quantifies downstream, upstream, and lateral movement patterns as well as the resulting displacement under different flow conditions. We think that the observed behavioural patterns might also apply to other riverine species that show similar habitat requirements, life histories, and threats. The results support the notion of Lechner et al. (2016), who argue that a strict separation of active versus passive drift appears inappropriate. Instead, these authors propose a continuum drift mode and term it "actipassive" drift. However, the particularities of larval movement behaviour as such and its implications have not yet been described. In general, the observed variability in the use of different downstream movement patterns, even at the level of individuals, suits well in this concept. Most recently, Glas et al. (2017) developed a novel, rheo-

Table 1. Movement patterns (mP) of fish larvae assessed in this study.

Movement pattern	Scheme	Direction of movement	Orientation towards current vector	V_f
Active upstream (Au)		Upstream	±135°, 180°	NA
Lateral (L)		Lateral	±90°, ±135°, 180°	NA
Active (A)		Downstream	±45°, 0°	Positive (>1 cm·s^{-1})
Passive (P)		Downstream	Any orientation (at random)	Zero (≈0 cm·s^{-1})
Active–passive (Ap)		Downstream	±135°, 180°	Negative (< −1 cm·s^{-1})
Traversing (T)		Downstream (with lateral component)	±90°	Positive (>1 cm·s^{-1})

Note: In the schemes, grey arrows indicate the current vector, while black arrows show the resulting direction of larval fish movement; the lengths of the arrows reflect the actual speed of the fish larvae compared with the flow velocity. The criteria for the differentiation of distinct mP were as follows: direction of movement relative to the flow, the orientation towards the current vector (cf. Fig. 2), and V_f (deviation of fish speed from flow velocity) during downstream movement. Abbreviations used in the Results section are provided in parentheses. The patterns active, passive, and active–passive downstream movement refer to those described in Pavlov et al. (2008). V_f was not assessed (NA) in case of upstream and lateral movements.

Table 2. Results from general linear models (GLMs) calculated for all mP assessed.

	Constant term		Stage		Scenario		Release		Light	
mP	*F*	*p*	*F*	*p*	*F*	*p*	*F*	*p*	*F*	*p*
Au	330.248	**<0.001**	2.171	0.127	7.142	**0.002**	1.169	0.286	0.726	0.399
L	176.576	**<0.001**	2.250	0.118	0.115	0.892	2.883	0.097	0.219	0.643
All dmP	401.600	**<0.001**	2.839	0.070	8.263	**0.001**	0.008	0.928	1.254	0.269
A	57.069	**<0.001**	1.050	0.358	0.699	0.502	0.180	0.673	2.047	0.159
Ap	581.083	**<0.001**	3.531	**0.037**	5.685	**0.006**	0.793	0.378	4.816	**0.033**
P	76.086	**<0.001**	2.996	0.060	3.604	**0.035**	0.321	0.574	2.437	0.125
T	65.493	**<0.001**	5.634	**0.006**	4.404	**0.018**	1.078	0.305	5.589	**0.022**

Note: Au = active upstream, L = lateral, all dmP = cumulative proportion of all downstream movement patterns, A = active downstream, Ap = active–passive downstream, P = passive downstream, T = traversing. Significant *p* values ($p < 0.05$) are highlighted in bold.

reaction based model approach for the analysis of mP. Although, in this approach frequencies of distinct mP were calculated as proportions of the residence time in the specific mP, the results largely support the findings of the present study concerning (*i*) the dominances of specific mP (i.e., of Ap downstream movement and of Au movement at low flow conditions) and (*ii*) the significance of the experimental factors stage and flow velocity as predictors of frequencies of distinct mP.

Active upstream (Au) movement

Au movement accounted for more than 50% of total longitudinal movement in ~70% of tested larvae at under-critical flow conditions.

Fig. 3. Frequencies of active upstream (Au), lateral (L), and all downstream movement patterns together (dmP) in percent for all tested larval stages (L2, L3, and L4), flow scenarios (under-, near-, and over-critical), and light conditions (day and night). Asterisks indicate significant differences of the frequency of the respective mP compared with all other mP and refer to Bonferroni post-hoc tests, which were conducted in case of detected significance within each category of stage–scenario–light following a bootstrap ANOVA (1000 resamples).

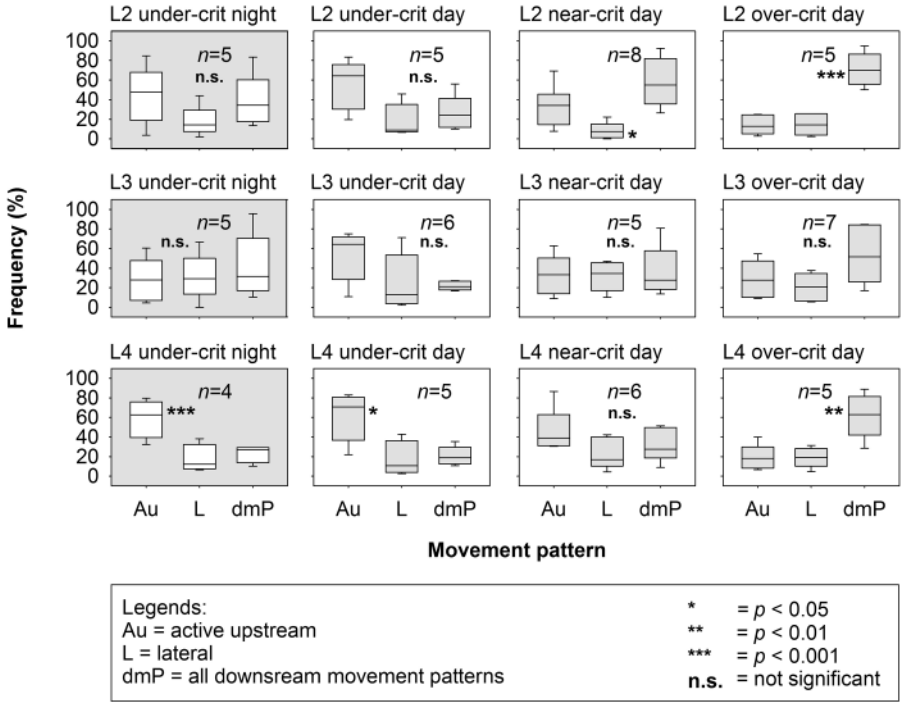


Fig. 4. Frequencies of distinct downstream movement patterns (dmP) in percent for all tested larval stages (L2, L3, and L4), flow scenarios (under-, near-, and over-critical), and light conditions (day and night). Asterisks indicate significant differences of the frequency of active–passive (Ap, grey shaded boxes) compared with all other mP and refer to Bonferroni post-hoc tests, which were conducted in case of detected significance within each category of stage–scenario–light following a bootstrap ANOVA (1000 resamples).

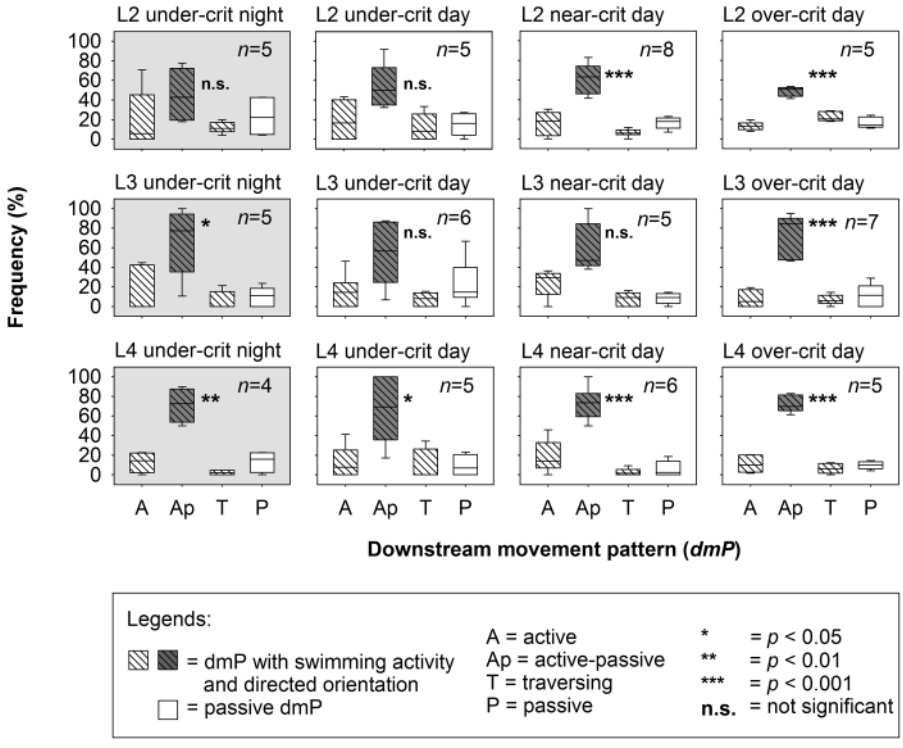
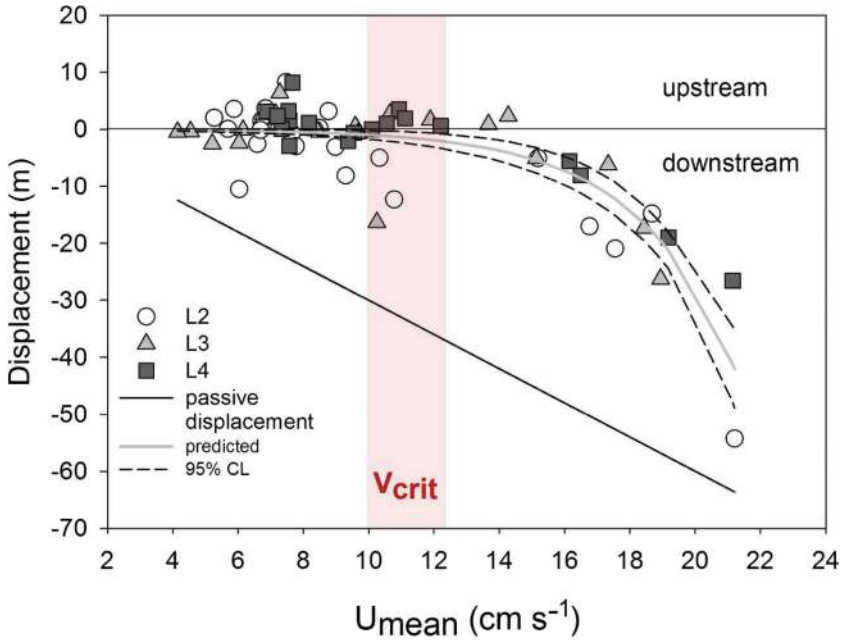


Fig. 5. Displacement distances of different larval stages (L2, L3, and L4) in relation to mean current velocity of swimming trajectories (U_{mean}) within the observation period of 5 min. Data points above the horizontal line indicate upstream displacement, while data points below this line indicate downstream displacement. Each data point represents the result of one experiment. The solid black line shows hypothetical passive downstream displacement if larvae were transported downstream exactly as fast as U_{mean}. The red-shaded area indicates the size-dependent critical current velocity (v_{crit}) of fish larvae used in this study. A significant nonlinear regression (solid grey line) was fitted to describe the relationship between displacement and U_{mean} for all tested fish larvae: $y = -0.0175 \cdot \exp(0.3788 \cdot x)$, $p < 0.001$ (y = displacement and $x = U_{mean}$ of the trajectory). [Colour online.]



This finding was unexpected because very few studies were able to detect significant Au movement of fish larvae in rivers so far. Schludermann et al. (2012) documented upstream movement of nase larvae in a mark–recapture study along a natural shoreline rearing habitat of the Danube River. A very small proportion of released larvae remained in the study reach — these were classified as "retained". Out of those, however, the vast majority (22 out of 24 larvae) were found upstream of the release point. They were significantly larger than larvae of the same release event that were captured in drift nets. Lechner et al. (2014) released marked L2 and L4 larvae simultaneously in the Danube River. On the fifth day after release, only L4 larvae were detected up to 150 m upstream of the release point, while L2 larvae were exclusively recaptured downstream. These findings are partly supported by our results; Au was highest in L4 larvae, which were significantly larger than L2 and L3 larvae ($p < 0.001$, Kruskal–Wallis test). However, Au occurred also frequently in L2 and L3 larvae, and the factor stage was not a significant predictor for the proportions of Au ($F = 2.171$, $p = 0.127$, GLM). Our results suggest that riverine fish larvae potentially use zero- and low-flow zones along the shoreline as corridors for upstream migration to a hitherto unknown large extent. A considerable number of larval riverine fish thereby possibly compensate for accidental drift and reach promising rearing habitats upstream — already during early developmental stages. From this perspective, the continuity of corridors with under-critical flow conditions along river shorelines, which allow upstream movement, might be of major importance for natural dispersal dynamics and would strongly promote recruitment.

Downstream movement patterns

The Ap pattern was the predominantly used type of downstream movement during all tested flow scenarios and larval stages. Previously, it was assumed that this pattern reflected inhibition of swimming abilities due to starvation or low water temperature (Pavlov et al. 2008). In our experiments, flume water temperatures equaled those in the rearing tank and were near the optimal temperature of the species' physiology (Kamler and Keckeis 2000). Additionally, larvae were adequately fed throughout the study period. We therefore argue that starvation and (or) a decrease in water temperature does not underlie the observed dominance of the Ap pattern. Based on the reslts of lab experiments, Pavlov et al. (2011) stated that Ap movement might be a manifestation of negative rheoreaction and not merely reflect inhibited locomotive activity. The authors analyzed patterns of downstream movement in juvenile carp (*Cyprinus carpio*, standard length (SL) = 28–43 mm), bream (*Abramis brama*, SL = 38–57 mm), and roach (*Rutilus rutilus*, SL = 34–55 mm) and found that 50.0% of studied specimens of bream, 36.8% of roach, and 18.6% of carp were using the Ap pattern at a mean flow velocity of 20.85 cm·s^{-1}. Evidence for the occurrence of Ap at over-critical flow conditions has also been reported in larvae of a marine gobiid, *Gobiosoma bosc* (Breitburg 1994).

The ecological reasons of using Ap, however, remain unclear and the following assumptions are largely speculative; possibly, the benefits associated with the use of Ap result from greater skills in sensing, orientating, and maneuvering (i.e., controlling their movement), thereby favoring successful settlement in appropriate habitats, prey capture, and predator avoidance. For example, Pavlov et al. (2011) discovered that juveniles had a greater chance to enter micro-eddy zones when they were moving downstream with their heads in upstream direction. Moreover, reducing downstream displacement in over-critical currents (e.g., during accidental drift in case of floods) seems to provide a reasonable strategy and would reflect evolutionary adaption to life in fast-running waters. Pavlov et al. (2008) suggested the P form of downstream movement to be typical for early larvae and to be in general more common at night and (or) in highly turbid conditions. In the present study, frequencies of P were only slightly higher in L2 and L3 compared with L4 larvae and stage was not a significant factor for proportions of this mP. The newly described

movement pattern, traversing (T), occurred in almost all larval stages and flow scenarios, but in a low percentage of all observed movement patterns. The only exception were L2 larvae at the over-critical flow scenario, where T accounted for 22.6% (±5.0%) of all downstream movements. The movement pattern that we term T here was indicated by strong propulsive activity (clearly positive values of V_f) and orientation perpendicular to the current. Based on these characteristics, we argue that T could be an effect of spontaneous burst swimming and (or) represents a brief attempt to scan the surroundings in lateral direction (e.g., to approach favorable zones or escape unfavorable zones). In the lab experiments from Pavlov et al. (2011), actively moving juveniles of carp, bream, and roach were sometimes orientated such that "the axis of the fish body was perpendicular to the vector of current and the head was always directed to the same stationary landmark". Proportions of this type of orientation accounted for 13.6%, 0.0%, and 15.8% of individuals of these species, respectively. The manifestation of the Ad movement was low to moderate in our study; means ranged from 7.4% ± 8.0% (L3–over-crit–day) to 24.4% ± 14.3% (L3–near-crit–day). These findings suggest possible differences in movement behaviour between larvae and juveniles. Pavlov et al. (2011) discovered relatively high proportions (47.4% to 64.4%) of juvenile cyprinids that moved downstream actively and assumed that the manifestation of Ad movement depends on the motivational state of the individuals. The dissimilarity in the findings of the two studies implies that Ad movement is more typical for juveniles than for larvae or simply reflects significant species-specific differences.

Our results strongly suggest that dispersion in larvae of the studied species (and probably of others) is a primary consequence of active behaviour rather than being a process of passive dislocation. This was indicated by (*i*) the high proportion of active upstream movement at conditions lower than the critical flow velocity, (*ii*) the significant dominance of downstream movement patterns, which include active swimming and directed orientation, in particular the high proportion of the active–passive downstream movement pattern and, (*iii*) the relatively low frequency of the passive movement pattern. Consequently, in the sense of the framework of actipassive drift proposed by Lechner et al. (2016), we argue that the drift mode of nase larvae should be referred to as mainly active–passive. Our results demonstrate that movement behaviour definitely has to be taken into account when attempts are made to understand or model dispersal patterns of fish larvae in rivers, while the simplifying assumption of largely passive drift has to be clearly rejected. Furthermore, our study highlights that larval fish dispersion in rivers might not almost exclusively act in the downstream direction, but also upstream, to a hitherto unknown large extent.

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