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Differences in behavioral responses of *Eurytemora affinis* (Copepoda, Calanoida) adults to salinity variations

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2 **Differences in behavioral responses of *Eurytemora affinis* (Copepoda, Calanoida)**
3 **reproductive stages to salinity variations**
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42 **Keywords:** *Eurytemora affinis*, estuarine copepod, salinity, swimming behavior, symbolic analysis
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ABSTRACT

Estuarine copepods experience strong and frequent salinity variations caused by alternating tides. Salinity is known to be a major parameter in the ecology of copepods from brackish waters but its effects on small scale behavior are relatively unclear. In this study we used 2-dimensional laboratory filming techniques to record swimming paths of the three reproductive stages (i.e. male, non-ovigerous female and ovigerous female) of *E. affinis* from the Seine estuary, under different salinity conditions. We show that the probability density of both the instantaneous swimming speed and the duration of the two most frequent swimming states (i.e. break and slow swimming) followed a power-law trend regardless of the salinity. Increase in salinity (i.e. 5, 15, 25 and 30) steadily decreased the instantaneous speed of *E. affinis* reproductive stages. At the extreme salinity of 0.5, the mean speed decreased for non-ovigerous females and ovigerous females but increased for males, when compared to higher salinities. Maximum speed was observed at salinity 0.5 for males and at salinity 5 for non-ovigerous females and ovigerous females. Low swimming speeds were associated with break events of high frequency and long duration. High swimming speeds were exhibited with cruising states of high frequency and break events of short duration. This supports laboratory and field studies indicating a preference of *E. affinis* for low to medium salinities.

INTRODUCTION

Maintenance and fitness optimization of copepod populations in macrotidal estuaries is a complex process because of the highly dynamic environment, leading to frequent variations in environmental conditions which can be considered as real selection pressures. Alternating tides reconfigure the local estuarine hydrodynamics and are responsible for variations in freshwater and seawater inputs, leading to important and rapid changes in salinity which local populations of copepods have to face. Salinity is a major factor affecting the ecology of estuarine copepods, driving the spatial distribution of species (Collins and Williams, 198; Roddie *et al.*, 1984; Soetaert and Van Rijswijk, 1993; Laprise and Dodson, 1994; Mouny and Dauvin, 2002; David *et al.*, 2005) and affecting life history traits (Holste and Peck, 2005; Calliari *et al.*, 2006; Chen *et al.*, 2006; Devreker *et al.*, 2009; Beyrend-Dur *et al.* 2009).

The calanoid copepod *Eurytemora affinis* (Pope, 1880) is one of the commonest species in most European and North American estuaries (Winkler and Grieve, 2004) and often dominates the zooplankton community in the low to medium salinity zone (Lawrence *et al.*, 2004). In the Seine estuary, *E. affinis* is the dominant species in the oligohaline and mesohaline zones throughout the

1
2 36 year (Mouny and Dauvin, 2002; Devreker *et al.*, 2008). Both physiological and behavioral
3
4 37 adaptations to short-term salinity variations have been seen in *E. affinis* and include a high capacity
5
6 38 of osmoregulation (Roddie *et al.*, 1984; Kimmel and Bradley, 2001) and active vertical tidal
7
8 39 migrations. These are directed toward the surface during low tide and flow and toward the seabed
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10 40 during high and ebb tide, avoiding advection by seaward and landward flows toward areas with less
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12 41 optimal salinities (Hough and Naylor, 1991 and 1992; Morgan *et al.*, 1997; Kimmerer *et al.*, 1998).
13
14 42 In the Seine estuary, Devreker *et al.* (2008) showed considerable complexity and variability in
15
16 43 population structure during a tidal cycle, with a differential distribution of developmental stages as
17
18 44 a function of salinity and depending on their swimming abilities. Nauplii were distributed as passive
19
20 45 particles in the low salinity zone, early copepodids were mainly found at low-medium salinities and
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22 46 late developmental stages were distributed over almost the entire salinity range, with an increasing
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24 47 density from salinity 0.5 to 5, a sharp decrease above 7.5, and a very few individuals above 20. The
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26 48 results of these studies indicate that adults benefit from better swimming capabilities to stay within
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28 49 a favorable salinity zone, supporting a preference of *E. affinis* for a certain range of salinity and
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30 50 demonstrating the influence of salinity on individual distribution.

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32 51 Nevertheless, it is still unclear whether salinity has a direct influence on small scale swimming
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34 52 behavior, where key behaviors such as feeding, mating or predator avoidance occur. This study has
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36 53 been designed to determine the effects of salinity on the swimming behavior of *E. affinis*
37
38 54 reproductive stages.

39 40 41 55 42 56 **METHOD**

43 57 44 58 **Experimental animals**

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46 60 Individual *E. affinis* were collected from surface tows with a WP2 plankton net (50 cm mouth
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48 61 diameter, 2 m length, 200 μ m mesh aperture) during the first moments of the ebb tide, on February
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50 62 2008, from the Seine estuary near the "pont de Tancarville" (49°28'23"N 0°27'51"E). Live
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52 63 copepods were brought to the laboratory within a few hours of capture in isotherm containers filled
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54 64 with aerated water from the sampling point. Once in the laboratory, individuals were placed in a
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56 65 tank and maintained at salinity 15 and a temperature of 8°C, within a 12L/12D light cycle. These
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58 66 temperature and salinity conditions provide the best survival rate with minimal reproduction, thus
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60 67 retaining sample integrity (Devreker *et al.*, 2004; 2007; 2009). Copepods were fed on a mono-algal
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62 68 diet of *Rhodomonas marina* from laboratory cultures, at the same time every day and 8 hours before
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64 69 experiments, but not during the recording.

Filming conditions

Individuals were moved from the cold room acclimated to an intermediate temperature (13-15°C) and subsequently to the temperature of the darkroom (20°C) at least 8 hours before the experiments. Preliminary experiments showed that *E. affinis* from the Seine estuary can withstand large but gradual salinity variations within a few hours (Souissi, unpublished data). Individuals were acclimated to salinity over a few hours (i.e. 4 to 8 hours) by a progressive increase or decrease of salinity until the experimental salinity was reached, thus roughly matching a tide time scale. Adult *E. affinis* were picked up using a pipette under a dissecting microscope for the experiments, and damaged individuals were discarded. The swimming behavior of *E. affinis* in response to salinity was examined at 5 salinity conditions (0.5, 5, 15, 25 and 30). Ten individuals were placed in a small transparent plastic vessel (5 x 5 x 6 cm) filled with 0.125L of water made from filtered sea water (from the English Channel) adjusted to salinity with deionized water. Only mature individuals were used for experiments, with mean prosome lengths of 0.95 and 0.85 mm for females and males, respectively. The vessel was considered big enough to obtain trajectories independent of the side wall effect while allowing an accurate distinction of the moving particles. The density of individuals fell within the range of the observed abundance for this species in the Seine estuary (Mouny and Dauvin, 2002; Devreker *et al.*, 2008). The tank was finally placed in the darkroom to avoid any phototropism and lit from the bottom by a 4X7 IR diode array (12V, 3.75A, emitting at 880nm), 50 cm away from a Sony DCR camera (25 frames/sec) facing the tank. After 15 min of acclimation, the recording began and finished after 30 min of filming. Four replicate observations were made for each reproductive stage (male, non ovigerous female, ovigerous female) at each salinity condition, to compensate for inter-individual variability (Seuront *et al.*, 2004). During acclimation and after experiments, the temperature of the experimental vessel water was carefully monitored and was almost constant (18-19°C).

Video techniques

Video recordings were transferred to computer and cut into sequences of 5 min each (Adobe® Premiere® Pro 2.0), allowing better running of the software used to extract the trajectories (Labtrack v.2.1 ©Bioras). Only swimming paths in which the individuals were at least three body lengths away from the walls parallel to the camera axis and from the surface and bottom were considered. Trajectories with a duration shorter than 10 sec were similarly rejected. The coordinates of the trajectories were extracted and combined into a 2D picture. Errors and artifacts resulting from the extraction process were cleaned up. For each sex and at each salinity condition, a total number

of trajectories ranging from 60 to 143 were considered for the data analysis (Table I). Such an important difference in the trajectory numbers came from the different overall activities observed between the replicates, with concordance neither with salinity nor sex. Total data number (i.e. number of values of instantaneous speed between two successive positions) ranged from 90433 to 302478 (Table I).

Characterization of the swimming paths

Swimming speed

The instantaneous speed was estimated as the distance between two successive positions of a copepod divided by the time step of the camera. Its value v_i was thus computed as:

$$v_i = \frac{[(x_t - x_{t+1})^2 + (y_t - y_{t+1})^2]^{\frac{1}{2}}}{t_r} \quad (1)$$

where t_r is the temporal resolution of the camera (i.e. 0.04 sec) and $(x_t - x_{t+1})$ and $(y_t - y_{t+1})$ the copepod coordinates at time t and $t+1$, respectively. Mean instantaneous speeds were computed as follows: for each experimental condition, mean speeds were extracted for every trajectory available, and then averaged. Probability density functions (PDF hereafter) of instantaneous speeds were computed using Matlab® (v.7.5, ©The Mathworks, Inc.).

Swimming state frequency and duration

To create the swimming state categories, instantaneous swimming speeds were separated into 4 different speed groups (Cowles and Strickler, 1983), as follows: “break” for a swimming speed below 1mm/sec, when the copepod is not swimming but has minimal activity to avoid sedimentation; “sinking” for a swimming speed between 1 and 8 mm/sec and a direction straight towards the bottom, when the copepod is not swimming but sinks slowly due to the influence of gravity; “cruising” at a speed between 1 and 20 mm/sec when the copepod is actively swimming; and “fast swimming” for speed values over 20 mm/sec, when the copepod shows short swift movements. Durations of events were computed as the product of the elementary time step number and the temporal resolution of the camera (i.e. 0.04 sec). The minimal duration was thus one frame duration.

Statistical analysis

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2 140
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4 141 As the swimming speed values were not normally distributed (Kolmogorov-Smirnov test), non-
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6 142 parametric statistical analyses were carried out. Comparisons between males, non-ovigerous
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8 143 females and ovigerous females were performed using the Kruskal-Wallis test (KW test hereafter).
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11 145 **RESULTS**

12 13 146 14 15 147 **Effect of salinity on instantaneous speed**

16 148 17 18 149 *Mean instantaneous speed*

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22 151 Significant differences were observed in mean instantaneous swimming speed between the five
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24 152 salinity conditions for each reproductive stages (KW test, $p < 0.001$ for males and ovigerous females;
25 153 $p = 0.002$ for non-ovigerous females) (Figure 1). The swimming speed of non-ovigerous females,
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27 154 ovigerous females and males decreased when salinity increased between 5 and 30. At the extreme
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29 155 condition of salinity 0.5, average speed dropped off for non-ovigerous females and ovigerous
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31 156 females but not for males. Maximum speed was observed at salinity 0.5 for males and at salinity 5
32 157 for non-ovigerous females and ovigerous females.
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34 158 35 36 159 *Probability density functions*

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38 160
39 161 Figure 2A shows, as an example, the PDF of instantaneous speeds of *E. affinis* non-ovigerous
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41 162 females at salinity 5 and the PDFs of three common theoretical distributions (i.e. Gaussian, Log-
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43 163 normal and Gamma). Values are random numbers from respective theoretical distributions
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45 164 following the same parameters as the experimental distribution. We can see that the probability of
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47 165 high speed was much larger than that expected in the case of Gaussian distribution. The
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49 166 experimental PDF does not belong to a Gaussian distribution and deviates from the Log-normal and
50 167 Gamma PDF, corresponding rather to a heavy-tailed distribution. The probability density functions
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52 168 all exhibited a clear power-law trend and supported significant power-law fits (Table II).

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54 169 Figure 2B shows the PDFs of instantaneous speeds of *E. affinis* males at the five salinity conditions
55 170 tested, for speed values ranging from 6 to 40 mm.sec⁻¹. Correlations between means and PDF of
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57 171 instantaneous speeds can be seen, with the highest proportion of slow movement when the average
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59 172 speed is low, and more important proportion of fast movement when the average speed is large.
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173 174 **Swimming state parameters**

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Swimming state frequencies

The swimming state frequencies were significantly different for each reproductive stage between salinity conditions (Pearson χ^2 test, $p < 0.001$). The swimming state frequencies of non-ovigerous females and males exhibited a clear concordance with their respective swimming speed (figure 3), i.e. high cruising and break frequencies came with high and low instantaneous speeds, respectively. Break and cruising were the two main states involved in the swimming activity, ranging respectively from 50% to 80% and from 15% to 40% of the total amount of data. Sinking and fast swimming accounted for less than 10% and less than 1%, respectively. Break and cruising states showed respectively their lowest and highest frequencies at salinity 5 for non-ovigerous females. Highest frequencies of break events were exhibited by males at salinity 30, with lowest frequencies of cruising. Ovigerous females showed an increasing frequency of break events from salinity 5 to 30, with the lowest cruising behavior frequency at the maximum salinity. Under very low (0.5) and high (25 and 30) salinities, *E. affinis* non-ovigerous and ovigerous females spent more time in a break state, and males spent more time cruising at 0.5, which was the salinity at which they exhibited maximum speed.

Durations of events

Among the different swimming states considered here only break, cruise and sinking provided sufficient quantities of data. We have previously seen that break and cruising seemed to be mainly responsible for the observed differences in activity between salinity conditions. Only these two swimming states were therefore considered here. The mean duration of break events was significantly different between salinities for each reproductive stage (KW test, $p < 0.001$ for non-ovigerous females, ovigerous females and males), generally showing a good correlation with the instantaneous speeds and the swimming state frequencies (figure 4). Non-ovigerous females exhibited lower (0.14 sec) and higher (0.24 sec) durations at salinity 5 and 30, respectively, corresponding to higher and lower instantaneous speeds. Males showed an increasing duration (0.13 to 0.4 sec) from minimal to maximal salinity, inversely correlated with their swimming speed. Ovigerous females did not show a minimal duration at salinity 5 but at 0.5 (0.13 sec). However, their maximal duration (0.23 sec) was exhibited at salinity 30 which was also the minimal speed salinity. These short durations indicated a high overall activity of the species considered here. The mean duration of cruising events was significantly different between salinities for each reproductive stage (KW test, $p < 0.001$ for females, ovigerous females and males). Mean duration of cruising

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2 210 events did not follow the trend as that observed for mean speed and swimming state frequencies.
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4 211 However, maximum durations occurred at salinity 5 for non-ovigerous females (0.07 sec) and
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6 212 ovigerous females (0.09 sec). Minimum durations were observed at the two highest salinities for
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8 213 non-ovigerous females with 0.07 sec at salinities 25 and 30. Males also showed a minimal duration
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10 214 of 0.07 sec in the cruising state both at salinities 25 and 30.

11 215 Figure 5A shows, as an exemple, the duration of successive break events for *E. affinis* males at
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13 216 salinity 15. It can be seen that long durations are frequent. The corresponding PDF is shown in
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15 217 Figure 5B. The heavy tail indicates that long duration values are more frequent than with a
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17 218 Gaussian PDF. For each reproductive stage, the PDF of both break and cruising states followed a
18 219 clear power-law trend, the parameters of which are given in Table III for break and cruising states at
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20 220 each salinity condition.

21 22 221 23 222 **DISCUSSION**

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27 224 Due to alternating tides, estuarine copepods undergo strong salinity variations over short temporal
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29 225 scales. The effects of salinity on small-scale swimming behavior are still unclear. In this study we
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31 226 tested for the existence and extent of the effects of salinity on the swimming behavior of *E. affinis*
32 227 by comparing the swimming paths of the three reproductive stages under different salinity
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34 228 conditions.

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36 229 We found a marked effect of salinity on the swimming behavior of the three reproductive stages.
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38 230 Maximum speed and activity were exhibited at low salinities then decreased at higher salinities. *E.*
39 231 *affinis* seems well adapted to live in fluctuating salinities, having great osmoregulatory capabilities
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41 232 over a wide range of salinity (Roddie *et al.*, 1984; Gonzales and Bradley, 1994; Kimmel and
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43 233 Bradley, 2001). Nevertheless, field samplings and laboratory studies suggest the existence of a
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45 234 salinity preferendum for this species. *E. affinis* is unlikely to stay within high salinity zones of an
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47 235 estuary and is often the dominant species in the oligohaline and mesohaline zones in the upper
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49 236 estuary (Castel and Veiga, 1990; Mouny and Dauvin, 2002; Devreker *et al.*, 2008). Laboratory
50 237 studies have indicated a higher survival, reproductive activity and a shorter development time at
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52 238 low to medium salinities (Roddie *et al.*, 1984; Devreker *et al.*, 2004, 2007, 2009). Using *E. affinis*
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54 239 as an experimental model to assess contaminant impacts on copepods, Cailleaud *et al.* (2007)
55 240 investigated the effect of salinity on two biomarker enzymatic activities (i.e. acetylcholinesterase
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57 241 and glutathione S-transferase) and found maximal expression related to an optimal salinity range
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59 242 (i.e. at salinity 5 and 10), showing a physiological preference of *E. affinis*. The authors assumed an
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243 increasing energy devoted to osmotic regulation and a decrease in energetic cost allocated to other
244 biological processes due to stressful salinity conditions. Modifications in metabolism when salinity

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2 245 conditions diverge from the normal habitat of the species have been studied in the copepod *Acartia*
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4 246 *clausi* by Calliari *et al.* (2006) who observed detrimental effects on total metabolism at salinities
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6 247 beyond the optimal range. Similar results were obtained elsewhere. Gaudy *et al.* (2000) found an
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8 248 increasing respiration rate in *A. tonsa* and *A. clausi* when salinity conditions diverged from the
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10 249 optimal salinity zone of these species, indicating a need of supplementary energy for
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12 250 osmoregulation. This was similarly observed earlier in *Eurytemora hirundoides*, a strain of *E.*
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14 251 *affinis* (Gyllenberg and Lundqvist, 1979). According to Goolish and Burton (1989), the daily
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16 252 energy required to adjust to osmotic stress accounts for 11.6% of the total energy. Deviation from
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18 253 the optimal salinity range can be stressful, and increased energy demand required for
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20 254 osmoregulation may decrease energy allocated to other processes. For instance, the exposure of
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22 255 *Calanus helgolandicus* to reduced salinity water resulted in major changes in its feeding rate
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24 256 (Lance, 1964). In our study, the observed decrease in swimming speed and activity with increasing
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26 257 salinity may be related to the decrease in energy allocated to swimming.

25 258 We also found a behavioral difference between females and males at extremely low salinity. Unlike
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27 259 males, non-ovigerous and ovigerous females displayed maximal activity at medium-low salinity
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29 260 (i.e. 5) but not at very low salinity. This is consistent with previous studies from the Seine estuary
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31 261 survey program, which reported the presence of *E. affinis* males far upstream in the Seine estuary in
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33 262 nearly full freshwater, during the exceptional heatwave in August 2003, when the freshwater input
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35 263 was low. Using the Asian sub-tropical estuarine copepod *Pseudodiaptomus annandalei* (Sewell,
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37 264 1919), Chen *et al.* (2006) found a difference in tolerance to salinity shock between females and
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39 265 males, indicating a possible difference in osmoregulation capacity between sexes. Males of *E.*
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41 266 *affinis* might have a different osmoregulatory capacity to that of females, which could explain the
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43 267 observed maximum speed at very low salinity.

43 268 Copepods display intermittent moves with different speeds (e.g. Schmitt *et al.*, 2006; Alcaraz *et al.*,
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45 269 2007) whose respective frequencies can be seen through their probability densities, in addition to
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47 270 common metrics such as mean speed and standard deviation. Here, the probability density functions
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49 271 of both the instantaneous swimming speeds and the durations of events were best fitted by a power-
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51 272 law distribution. They also exhibited long tails, showing the relative importance of large amplitude
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53 273 events. In a more theoretical framework, these results can be used to calibrate numerical simulations
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55 274 of copepod swimming paths (Schmitt *et al.*, 2006).

55 275 A previous study (Seuront, 2006) investigated the effect of increasing salinity on the swimming
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57 276 behavior of *E. affinis* from a laboratory culture and found an increase in overall swimming activity,
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59 277 speed and path complexity for males and non-ovigerous females but not for ovigerous females. The
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278 present observations showed different results, highlighting the importance of the experimental
279 conditions employed for observations. In the present study, we used an experimental procedure and

1
2 280 an analysis process which greatly differed from the one applied in Seuront (2006), with differences
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4 281 in the rearing period, number of individuals per recording, feeding during experiments, salinity and
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6 282 temperature acclimation time, and trajectory numbers. Acclimation to salinity and temperature is
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8 283 known to be a critical element in osmoregulation capabilities and tolerance to salinity (Lance, 1963;
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10 284 Roddie *et al.*, 1984), and differences in the acclimation process may have led to different responses
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12 285 to salinity variation. It is also difficult to compare behavioral and physiological responses of
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14 286 copepods originating from a continuous culture (Seuront, 2006) and from field (this study).
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16 287 Beyrend-Dur *et al.* (2009) discussed the effect of the number of generations (i.e. age of the copepod
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18 288 culture) on the life cycle traits of *E. affinis* maintained at different salinities and indicated a possible
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20 289 selection pressure in laboratory continuous cultures. Since copepods are known to behave
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22 290 differently in the presence of food (Tiselius, 1992; van Duren and Videler, 1995; Seuront and
23
24 291 Vincent, 2008) and in order to avoid any possible synergistic effects of food and salinity, copepods
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26 292 were not fed during our experiments. We assume that the behavioral response observed in this study
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28 293 reflects the effect of salinity. Finally, because of the high inter-individual variability in copepod
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30 294 behavior and also due to its sensitivity to experimental conditions, a high number of trajectories
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32 295 should be considered.

30 296 In conclusion, the proposed metrics used in this study can be routinely applied to study the effects of
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32 297 different external factors on copepod behavior. Further investigations are needed to compare *E.*
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34 298 *affinis* responses with those exhibited by other species, contributing to a better understanding of the
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36 299 ecology of copepods within estuaries. Such studies offer the hope of elucidating some of the key
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38 300 factors driving the amazing inter and intra-specific behavior diversity displayed under different
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40 301 environmental conditions.

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55
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57
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2 408 **Table and Figure legends**
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6 410 **Table I.** Number of trajectories - data for *Eurytemora affinis* non-ovigerous females, ovigerous
7 411 females and males, for the five salinity conditions tested.
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11 413 **Table II.** Parameter values of the power-law fits adjusted to instantaneous speed probability density
12 414 functions for non-ovigerous females, ovigerous females and males of *Eurytemora affinis*, for the
13 415 five salinity conditions tested, of the type: $\ln(y) = -a \times \ln(x) + b$.
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18 417 **Table III.** Parameter values of the power-law fits adjusted to the probability density functions of
19 418 event durations for non-ovigerous females, ovigerous females and males of *Eurytemora affinis*, for
20 419 the five salinity conditions tested, of the type: $\ln(y) = -a \times \ln(x) - b$.
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26 421 **Fig. 1.** Mean instantaneous speed (mm/sec) of *Eurytemora affinis* non-ovigerous females (black
27 422 line, triangular symbol), ovigerous females (grey line, circular symbol) and males (light grey line,
28 423 cross symbol), for the five salinity conditions tested.
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33 425 **Fig. 2.** (A.) Probability density functions of a Gaussian distribution (solid line, black), a Log-normal
34 426 distribution (solid line, dark grey), a Gamma distribution (solid line, light grey), and experimental
35 427 PDF of instantaneous speeds (dashed line, black) of *Eurytemora affinis* non-ovigerous females at
36 428 salinity 5. Theoretical PDFs were computed from distributions following the same parameters as the
37 429 experimental distribution. (B.) Probability density functions of instantaneous speeds, ranging from
38 430 6 to 40 mm.s⁻¹, of *Eurytemora affinis* males for the five salinity conditions tested (salinity 0.5: solid
39 431 line, black; salinity 5: dashed line, black; salinity 15: solid line, dark grey; salinity 25: dashed line,
40 432 dark grey; salinity 30: solid line, light grey).
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49 434 **Fig. 3.** Swimming state frequencies of *Eurytemora affinis* non-ovigerous females (A.), ovigerous
50 435 female (B.) and male (C.), for the five salinity conditions tested. Break state: black; cruising state:
51 436 grey.
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56 438 **Fig. 4.** Mean duration of break state (A.) and cruising state (B.) for *Eurytemora affinis* non-
57 439 ovigerous females (black, cross symbol), ovigerous females (grey, triangular symbol) and males
58 440 (light grey, circular symbol), for the five salinity conditions tested.
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2 442 **Fig. 5.** (A.) Plot of the time duration of 10000 successive break events for *Eurytemora affinis* males
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4 443 at salinity 15. Durations above 10s are not shown. (B.) Corresponding PDF (gray curve). The black
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6 444 discontinuous line represents the power-law fit.
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Table 1.

Salinity	0.5	5	15	25	30
Non- Ovigerous Females	60 - 96217	117 - 131802	80 - 151353	143 - 302478	84 - 132478
Ovigerous Females	65 - 90433	90 - 118574	99 - 127342	72 - 155664	118 - 260441
Males	141 - 11885	123 - 146715	125 - 178812	108 - 243986	89 - 213564

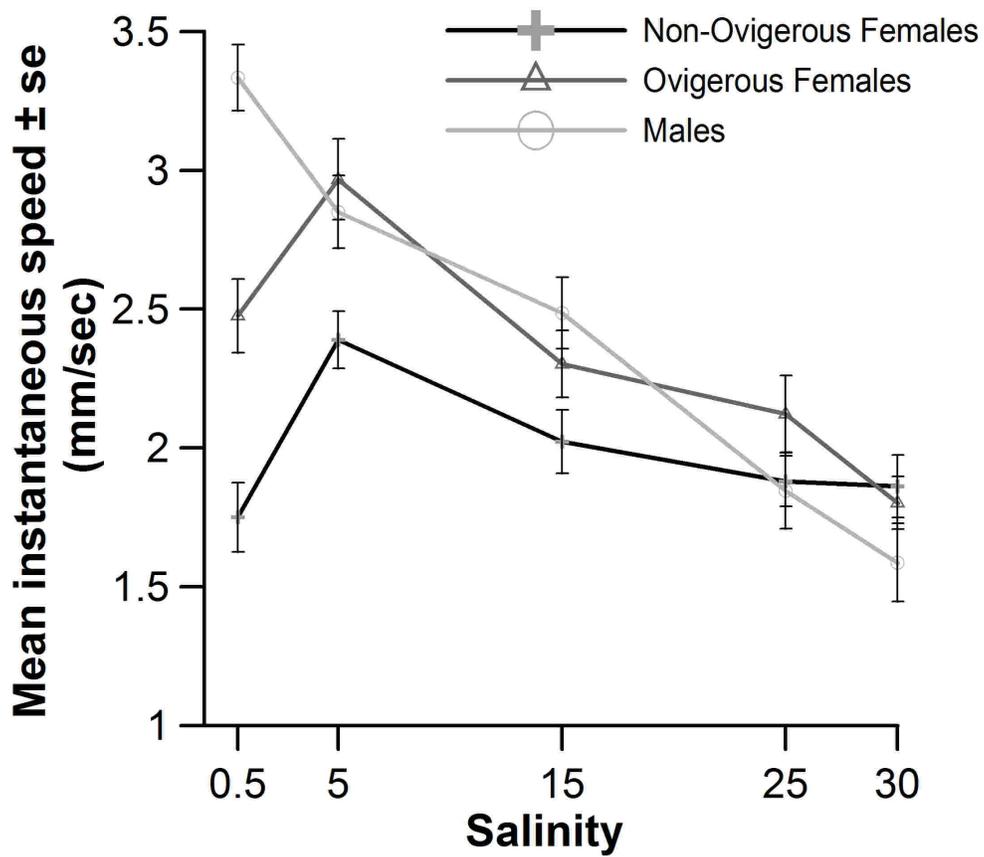
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Table II.

	Salinity	a	b	R ²
Non-Ovigerous Females	0.5	2.7	1.4	0.95
	5	2.9	2.2	0.96
	15	3.1	2.4	0.95
	25	3	2.3	0.91
	30	2.8	1.6	0.97
Ovigerous Females	0.5	3.2	3.3	0.95
	5	2.7	2	0.97
	15	3.2	3.2	0.95
	25	3.2	3.2	0.94
	30	3.3	2.8	0.96
Males	0.5	3.5	4.3	0.93
	5	2.9	2.8	0.96
	15	2.7	1.3	0.75
	25	2.9	2.1	0.92
	30	3.4	2.6	0.96

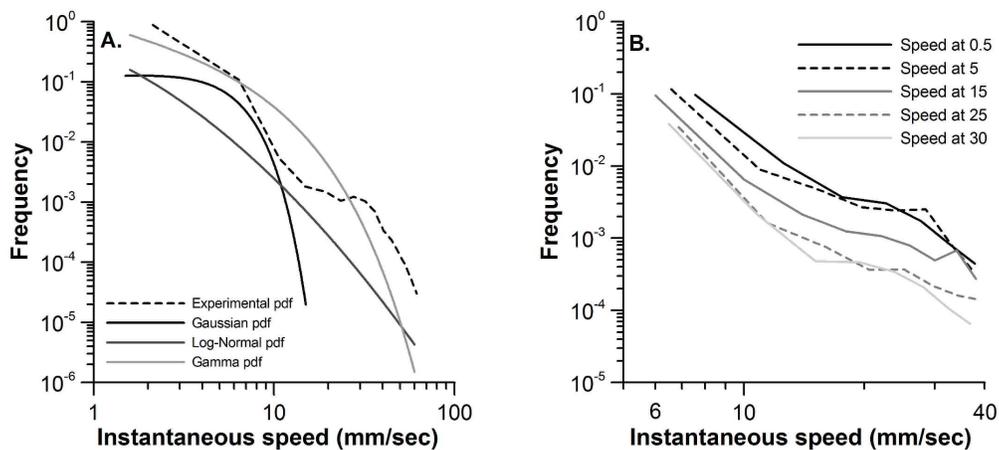
Table III.

	Salinity	Break			Cruising		
		a	b	R ²	a	b	R ²
Non-Ovigerous Females	0.5	2.1	6.2	0.99	3.2	8.3	0.99
	5	2.3	6.5	0.99	3.1	8.0	0.99
	15	2.3	6.3	0.99	3.2	8.3	0.99
	25	2.2	6.2	0.99	4.3	10.5	0.94
	30	2.2	5.9	0.99	3.4	8.6	0.99
Ovigerous Females	0.5	2.3	6.4	0.99	3.4	8.6	0.98
	5	2.2	6.3	0.99	2.9	7.5	0.98
	15	2.1	6.1	0.99	2.8	7.4	0.99
	25	2.2	5.9	0.99	3.8	9.5	0.99
	30	2.2	5.9	0.99	3.0	8.0	0.99
Males	0.5	2.3	6.5	0.99	2.8	7.2	0.99
	5	2.1	6.3	0.99	2.9	7.5	0.99
	15	2.1	6.1	0.99	2.6	6.9	0.98
	25	2.1	6.0	0.99	3.2	8.4	0.99
	30	2.0	5.8	0.99	2.8	7.6	0.99



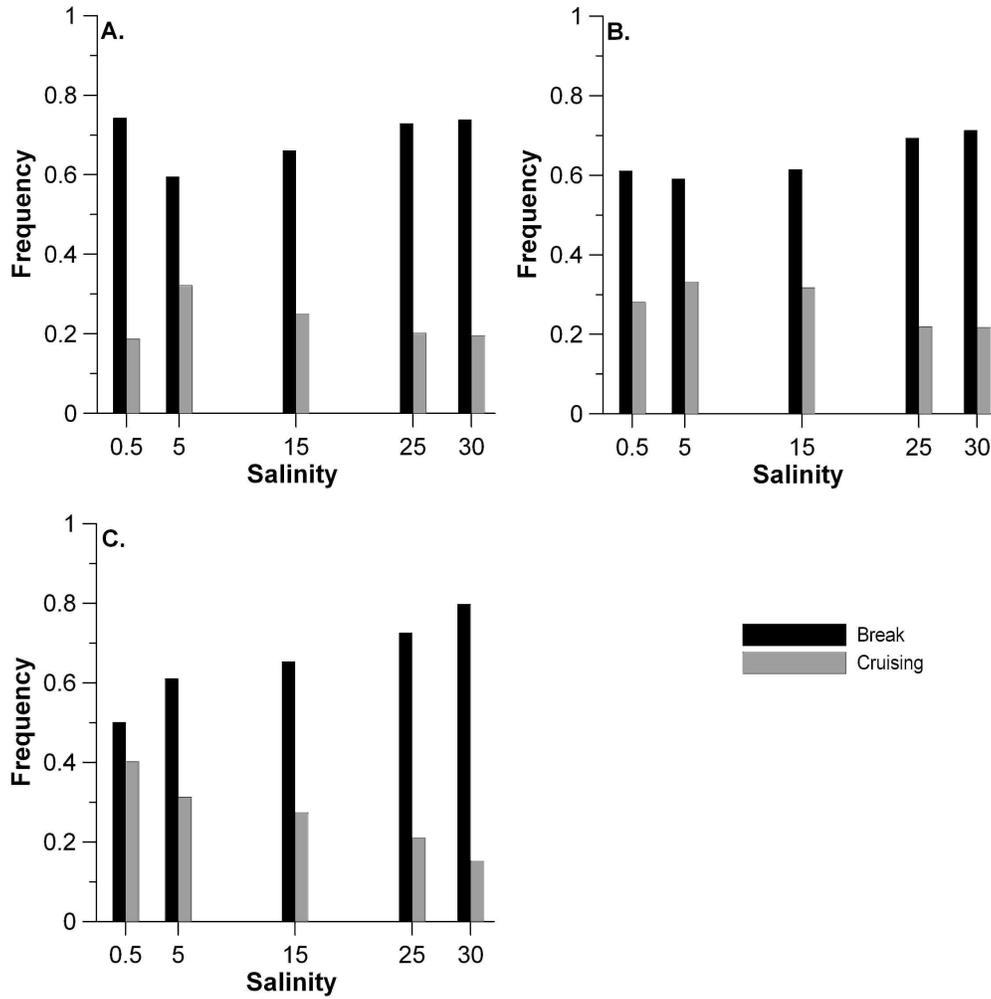
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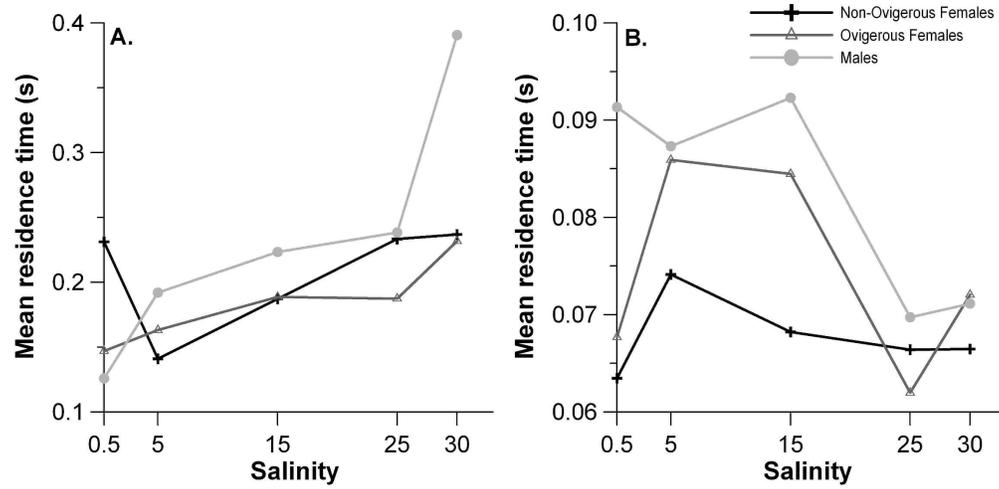
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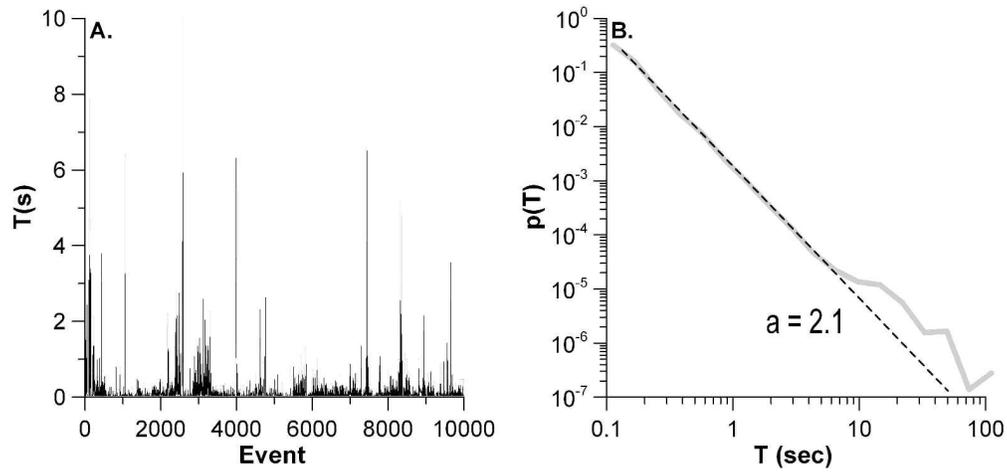
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