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The amphipod *Talitrus saltator* as a bioindicator of human trampling on sandy beaches

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1	The amphipod Talitrus saltator as a bioindicator of human
2	trampling on sandy beaches
3	
4	Alberto Ugolini <sup>a,</sup> *, Giuseppe Ungherese <sup>a</sup> , Silvia Somigli <sup>a</sup> , Giuditta
5	Galanti <sup>a</sup> , Davide Baroni <sup>b</sup> , Francesca Borghini <sup>b</sup> , Nicola Cipriani <sup>c</sup> ,
6	Massimo Nebbiai <sup>c</sup> , Maurizio Passaponti <sup>d</sup> , Silvano Focardi <sup>b</sup>
7	
8	<sup>a</sup> Dipartimento di Biologia Animale e Genetica, Università degli Studi di Firenze, Via
9	Romana 17, 50125 Firenze, Italy
10	<sup>b</sup> Dipartimento di Scienze Ambientali, Università degli Studi di Siena, Via Mattioli 4,
11	53100 Siena, Italy
12	<sup>c</sup> Dipartimento di Scienze della Terra, Università degli Studi di Firenze, Via La Pira 4,
13	50121 Firenze, Italy
14	<sup>d</sup> Dipartimento di Chimica Organica, Università degli Studi di Firenze, Via della
15	Lastruccia 13, 50019 Sesto Fiorentino, Italy
16	
17	* Corresponding author: Tel.: +39 55 2288219, Fax: +39 55 222565
18	E-mail address: ugolini_alb@dbag.unifi.it
19	
20	Abstract
21	The present paper assesses the use of the supralittoral amphipod Talitrus saltator
22	as a bioindicator of the effects of human trampling on the supralittoral sandy band.
23	Samplings in delimited areas were carried out at sites subjected to different human
24	impact. The results showed a strong negative correlation between the number of
25	swimmers and the sandhopper population density, while there was no clear relationship
26	between sandhopper abundance and the other factors considered: granulometry,
27	compactness and organic carbon content of the sand, and trace metal contents in the
28	sand and sandhoppers. A field test of trampling conducted in a confined space showed
29	its direct negative effect on sandhopper survival. However, trace metal analysis
30	confirmed the ability of T. saltator to bioaccumulate some elements (Hg, Zn, Cu, Cd).

Our study demonstrates that *T. saltator* is a good bioindicator of human impact in the
supralittoral zone of sandy shores.

33

34 Keywords: Human trampling, Heavy metals, Mediterranean Sandy shores, Beaches,

- 35 Bioindicator, Amphipod, *Talitrus saltator*.
- 36

#### 37 **1. Introduction**

38

39 In the coastal zone, the input of materials and substances from terrestrial, fluvial 40 and marine sources is particularly important. In addition to elevated concentrations of 41 nutrients, there can be increased quantities of natural and anthropogenic contaminants. 42 Since the 1960s, European shores (particularly Italian ones) have seen a very strong 43 increase in the number of swimmers at beach resorts. Many studies on the submerged 44 and intertidal littoral environment of rocky and sandy shores have dealt with the use of 45 organisms as biomonitors of trace metal contamination. Similarly, many investigations 46 have attempted to evaluate the effects of human trampling and recreational uses on 47 natural populations and communities of the littoral zone of sandy shores (see Milazzo et 48 al., 2002 for a review). In contrast, the supralittoral band, an ecotonal environment of 49 great ecological importance in the economy of sandy shore ecosystems, has received 50 relatively little attention (see Brown and McLachlan, 1994; Schlacher et al. 2007). 51 Most research on the effects of trampling on natural communities concerns the 52 vegetation or macrofaunal components (especially birds) (Liddle, 1975; Brown and 53 McLachlan, 1994). Several studies have examined the effects of trampling on the 54 invertebrate fauna of rocky shores or of loose substrata of the intertidal zone (for 55 instance see Povey and Keough, 1991; Brosnan and Crumrine, 1994; Chandrasekara 56 and Frid, 1996; Casu et al., 2006; Dye, 2006; Johnson et al., 2007). Instead, there have 57 been very few investigations of the supralitoral fauna of sandy shores and the results of 58 these studies have been contradictory (Jaramillo et al., 1996; Moffet et al., 1998; 59 Weslawski et al., 2000b, c; Veloso et al., 2006). Recent studies have attempted to 60 establish the utility of supralittoral amphipods as indicators of the impact of tourist 61 activities on sandy shores (Fanini et al., 2005). However, because of their methods and 62 sampling sites, the investigations have not provided clear results.

63	In summer, the supralittoral zone is intensely frequented by swimmers but it is					
64	also where the major consumers of stranded organic material, i.e. talitrid amphipods and					
65	isopod, find shelter during the day, buried in the wet sand. In fact, most of the energy					
66	flow deriving from the use of stranded material passes through supralittoral crustaceans					
67	(Branch and Branch, 1981; Griffiths et al., 1983; Brown and McLachlan, 1994).					
68	Sandhoppers occupy a trophic niche as detritivores and collector-gatherers. Hence they					
69	feed on plant and animal material from both marine and terrestrial sources (Palluault,					
70	1954; Wildish, 1988; Bergerard, 1989), integrating the two routes of contamination of					
71	the supralittoral.					
72	The aims of the present study were to examine the possibility of using the					
73	amphipod T. saltator as a bioindicator of the effects of human trampling on the					
74	supralittoral of sandy shores and to provide a simple low-cost monitoring tool. For this					
75	reason the relationship between sandhoppers abundance and swimmers has been					
76	considered, as well as the possible influence of some other factors such as granulometric					
77	composition and sand compactness, organic carbon content in the sand and trace metal					
78	contamination.					
79						
80	2. Materials and methods					
81						
82	2.1 Sandhopper sampling					
83						
84	Samplings to determine the density of sandhoppers were carried out at eight sites on					
85	the coast of Tuscany subjected to different tourist impact and either with or without					
86	beach cleaning procedures. In the five sites inside the Regional Park of Migliarino, San					
87	Rossore, Massaciuccoli (MSRM Park, Pisa), the sampling was performed once a month					
88	from June to September 2004, 2005 and 2006:					
89	O1) Oasi 1 (cleaned beach)					
90	SM) Serchio Mare (seashore, not cleaned)					
91	SF) Serchio Fiume (right bank of river mouth, not cleaned)					
92	BV) Batteria Vecchia (not cleaned)					
93	FM) Fiume Morto Vecchio (not cleaned)					

94	At the three sites along the Feniglia sandbar (southern Tuscany, Grosseto), the
95	sampling was performed in July and September 2005:
96	FA) bathhouse (Ansedonia side, cleaned beach)
97	FB1) beach ca. 1 km from FA (cleaned)
98	FB2) beach ca. 2 km from FA (cleaned)
99	At each sampling, all the sandhoppers were captured (also disturbing the surface
100	layer to sample those in the sediment) and counted inside four areas (75 cm <sup>2</sup> each)
101	delimited by plastic containers inserted a few cm into the sand. Each container was
102	inserted along the sea-land axis of the beach at a progressively increasing distance
103	starting from the water line until the beginning of dry sand. At each site, the sampling
104	was carried out twice, a few meters apart, since the distribution of sandhoppers in the
105	band of wet sand does not seem to be uniform (Weslawski et al., 2000a).
106	At each sampling site, the people to the right and left of the shoreline, along a
107	150 m transect in the central hours of the day on the days with the highest tourist
108	presence, were also counted (Saturday and Sunday).
109	At some locations, the following factors were also analyzed to exclude
110	differences between the sampling sites: 1) contents of trace metals in the sand and in the
111	sandhoppers (Hg, Cr, Cd, Cu, Zn, Fe, Al, Pb); 2) compactness of the sand; 3)
112	granulometric composition of the sand; 4) organic carbon content of the sand.
113	
114	2.2 Trace metals
115	
116	The trace metal analysis was conducted on sand samples collected from the zone
117	frequented by the sandhoppers during the day and on adult individuals of T. saltator
118	collected at all the sites. The animals were transported to the laboratory in plastic
119	containers with sand and detritus from the sampling site and then sacrificed by freezing.
120	At each site, a sample of sand was collected in the zone frequented by the sandhoppers
121	during the day. We dehydrated the samples at 40°C until they reached a constant weight
122	and then were finely ground. Around 150 mg of each sample were mineralized in
123	Teflon vessels containing 3 ml of HNO3 at 120°C for 8 h. After digestion, the solution
124	was brought to a volume of 10 ml by addition of deionized water. During each

125 mineralization, we performed several "blank trials", by putting only the reagents in the 126 Teflon containers, to verify the absence of contamination. 127 The analyses were performed by atomic absorption spectrometry. Cd, Cr, Cu 128 and Pb were determined by atomization with a graphite furnace and Zeeman 129 background corrector. The FIMS (Flow Injection Mercury System) was used for Hg, 130 while we determined Al, Fe and Zn by Inductively Coupled Plasma Atomic Emission 131 Spectrometry (ICP/EAS). Each analysis was carried out 3 times and the accuracy of the 132 analytical procedures was assessed by comparing the results of contemporary digestions 133 and analyses of Standard Reference Materials, SRMs): SRM 1566a "oyster tissue" from 134 the National Institute of Standards and Technology (NIST, Gaithersburg, USA). The 135 results of the determination were only accepted if the values measured for the SRMs 136 were within the certified concentration range; if not, we repeated the analyses. The 137 element concentrations (expressed as  $\mu g/g$ ) were calculated by the method of "internal 138 additions". The coefficients of variation for the various elements were between 5 and 139 10%. 140 141 2.3. Granulometric composition and sand compactness 142 143 For the granulometric analysis, the sand samples were split to obtain samples of 144 approximately 10 grams (the choice of the representative volume was based on the 145 median of the sediment, as estimated by visual examination). 146 A statistically representative portion of the sample was placed in an oven at 80°C to 147 eliminate the humidity, after which we sifted the material through a series of 148 progressively finer sieves (2500 µm, 1600 µm, 1000 µm, 630 µm, 400 µm, 250 µm, 160 149  $\mu$ m, 100  $\mu$ m, 63  $\mu$ m) for around 5 minutes with a Ro-Tap shaker. Since the samples 150 were prevalently sandy, it was not necessary to analyze the mud-clay fraction (<  $63 \mu m$ ) 151 of the sediment. 152 To assess the compactness of the sand in the zone frequented by sandhoppers 153 during the day, an aluminum pole was dropped from a height of 1 m and the distance 154 the pole penetrated into the sand was measured. The distance was transformed into 155 arbitrary units by assigning a value of 100% compactness to 0 cm and 50% 156 compactness to 10 cm.

157		
158	2.4.	Organic carbon
159		
160	,	The sand samples were collected at each site in July 2006. The organic carbon in
161	the sand	d samples was determined as the difference between the total carbon and the
162	inorgan	ic carbon. The sand was crushed to a fine powder so as to homogenize the
163	sample.	Total carbon was determined with a CHNS/O ANALYZER (mod. 2400 ser. II,
164	Perkin	Elmer), while inorganic carbon was measured with a De Astis calcimeter.
165		
166	2.5.	Trampling test
167		
168	,	The trampling test was carried out at Fiume Morto Vecchio (MSRM Park). Two
169	plastic o	cylinders (110 cm diameter) were fixed in the sand in the wet band frequented by
170	sandhoj	oppers during the day. All the individuals inside each of the delimited areas were
171	capture	d, counted and then freed within the same area. After around 15 minutes
172	(sufficie	ent time for the sandhoppers to return to their refuges in the wet sand), two
173	people	performed 400 footsteps in 15 minutes inside one of the two cylinders. The
174	number	of footsteps was chosen on the basis of the footsteps recorded during a survey
175	carried	out from 11:00 to 12:00 on a Saturday in July 2006 on the Marina di Pietrasanta
176	beach (a	a popular beach on the Tuscan coast). During this survey, a mean transit of 866
177	swimm	ers per hour at a selected point on the shoreline was recorded. The sandhoppers
178	within t	the other cylinder were left undisturbed. At the end of the trial, the sandhoppers
179	still aliv	ve in both cylinders were captured, counted and transported to the laboratory in
180	basins v	with wet sand. The numbers of individuals still alive 24 hours after the treatment
181	was also	o recorded.
182		

- 183 2
  - 2.6. Statistical analysis
- 184

The granulometric data were analyzed with specific software (Dept. of Earth
Sciences, Univ. of Florence) that provided parametric statistics indicative of the sorting
of the samples, as well as the percentages of the granulometric classes for the purposes
of textural classification (Shepard, 1954).

189 The statistical analysis of sandhopper density was performed independently of 190 an analysis of natural population fluctuations (see Anastácio et al., 2003; Margues et al., 191 2003) and/or a comparison of the population densities at different sites. Such analyses 192 were beyond the scope of our study and would not have provided information useful for 193 our aims. It is possible that sandhopper populations living in the same environment and 194 practically at the same latitude (ca. 200 km apart) present small differences in the 195 natural fluctuation of population density. However, since we performed relative 196 comparisons of the number of captured individuals among several localities of the same 197 zone, any differences in population density between different zones in the same period 198 of the year would not affect the interpretation of the results. The tests used are among 199 the most common ones in non-parametric statistics (Zar, 1984; Siegel and Castellan, 200 1989).

The possible influence of trace metal pollution on sandhopper density was assessed with the Spearman rank-order correlation coefficient test (Siegel and Castellan, 1989). In the figure illustrating trace metal pollution, we arranged the sampling sites in decreasing order of sandhopper density: the site with the highest sandhopper abundance in July 2006 (FM, captured sandhoppers = 1013) was given the arbitrary value of 100. The values for the other sites were expressed as a percentage of FM.

207

#### 208 **3. Results**

209

- 210 3.1 Sandhopper density
- 211

212 The frequency of T. saltator individuals collected at the MSRM Park sites was 213 negatively correlated with the frequency of swimmers (Fig. 1). Despite the small sample 214 sizes, statistical significance was reached for each of the four months (June-September) 215 (June and July,  $r_s = -1$ , p = 0.01; August and September,  $r_s = -0.925$ , n = 5, p < 0.05, n = -0.925, 216 5, Spearman rank-order correlation coefficient test, Fig. 1). This is also confirmed 217 considering the mean number of sandhoppers and swimmers (Table 1). In fact, the 218 Fiume Morto Vecchio site (FM) consistently had the lowest tourist impact and the 219 highest sandhopper abundance (Table 1). In contrast, no sandhoppers were found at the 220 site with the highest tourist impact, Oasi 1 (Table 1).

221	FIG. 1
222	
223	TABLE 1
224	The July vs. September comparison for the various sampling sites was
225	statistically significant for the localities in the MSRM Park (G = 114.5, df = 4, p< $0.001$ ,
226	fig. 1) but not for the sites on the Feniglia sandbar ( $G = 1.9$ , $df = 2$ , p>0.1, G test, Fig 3).
227	There was a significant monthly difference in sandhopper abundance even at a
228	single locality with very low tourist impact (Fiume Morto Vecchio, Table 1, Fig. 2, G =
229	664, df = 1, p< $0.001$ , G test) due to the natural population fluctuation. However, these
230	differences did not affect the swimmers-sandhoppers relationship.
231	FIG. 2
232	
233	It was not possible to make statistical comparisons for the Feniglia sandbar (Fig.
234	3) because of the small number of sampling sites. Nevertheless, the number of
235	sandhoppers captured at the Feniglia sites clearly followed the same trend in relation to
236	tourist impact as the one recorded at the MSRM Park sites, i.e. localities with higher
237	tourist impact had lower T. saltator densities.
238	FIG. 3
239	
240	Regarding the possible influence of beach cleaning on sandhopper survival,
241	there was a significant difference between the only cleaned site $(O1, n = 0)$ and the total
242	of the other MSRM sites $(SM + SF + BV + FM, n = 7551; G = 10434, df = 1,$
243	P<0.0001, G test). However, there were also significant differences among the not
244	cleaned sites (SM, n = 28; SF, n = 164; BV, n = 2160; FM n = 5 199;G = 10 077, df = 3,
245	P<0.0001, G test). A similar result was obtained for the cleaned beaches of the Feniglia
246	sandbar (FA, n = 131; FB1, n = 41; FB2, n = 3; G = 164, df = 2, P<0.01, G test).
247	
248	3.2 Trace metals
249	
250	At the sites with sandhoppers, the Cu, Cd, Hg and Zn concentrations (Fig. 4)
251	were constantly higher in the sandhoppers than in the sand. In contrast, the Fe, Pb and
252	Al contents (Fig. 4) were higher in the sand than in the sandhoppers. For Cr, there was a

253	higher concentration in the sandhoppers at two sites and in the sand at the other two
254	sites.
255	There was no significant correlation between sandhopper abundance (in Fig. 4
256	abscissa, the sites are in decreasing order of abundance) and trace metal contents in the
257	individuals or in the sand ( $P = NS$ in each case, $n = 6$ , Spearman rank-order correlation
258	test).
259	FIG. 4
260	
261	3.3. Granulometric composition and sand compactness
262	
263	The granulometric composition of the sand was fairly uniform among sites: all
264	samples were well classified and in the medium- to coarse-grained range (Table 2).
265	TABLE 2
266	
267	For the MSRM Park sites, there was no correlation between mean sand grain
268	diameter and sandhopper density (Spearman rank-order correlation coefficient test, $r_s$ =
269	- 0.1, $n = 5$ , $p = NS$ , Fig. 5A). A qualitative evaluation also suggested a lack of
270	correlation between sandhopper abundance and mean sand grain diameter at the
271	Feniglia localities (Fig. 5B).
272	Sand compactness at the MSRM Park sites (Fig. 5C) was not significantly
273	correlated with the number of sandhoppers ( $r_s = -0.175$ ; $n = 5$ , p=NS, Spearman rank-
274	order correlation coefficient test). Indeed the highest values occurred at site SF where
275	sandhoppers were present (Fig. 5C). For the Feniglia sites, a qualitative evaluation
276	indicated a negative correlation between sand compactness and sandhopper abundance
277	(Fig. 5D).
278	
279	3.4. Organic carbon
280	
281	For the MSRM Park sites, the organic carbon content of the sand was not
282	significantly correlated with sandhopper density ( $r_s = 0.7$ , $n = 5$ , $p = NS$ ; Spearman
283	rank-order correlation coefficient test, Fig. 5E). However, the localities along the
284	Feniglia sandbar showed a negative correlation between the two parameters (fig. 5F).

285	FIG. 5
286	
287	3.5. Trampling test
288	
289	The effect of trampling on T. saltator survival (Fig. 6) was evident immediately
290	after the treatment: the difference between the number of live individuals before and
291	after treatment was highly significant (G = 86, df = 1, p< $0.001$ , G test). After 24 hours,
292	the percentage of surviving sandhoppers dropped almost to zero ( $G = 428$ , $df = 1$ ,
293	p<0.001, G test) while the survival of the control sandhoppers remained the same.
294	FIG. 6
295	O
296	4. Discussion
297	
298	Of all the factors that might have negatively influenced the population density of
299	T. saltator, only the number of swimmers showed a significant correlation with the
300	number of captured sandhoppers. Although the effect of trampling in a confined test
301	space is probably not the same as in natural conditions (in which case the animals could
302	escape), our test results are the first direct proof that human trampling affects
303	sandhopper survival. This is likely an important factor at Mediterranean latitudes and
304	for species that burrow only slightly into the sand (ca. 3-5 cm depth).
305	This does not mean that other factors are never influential, only that they are not
306	particularly important in the conditions encountered in this study. Sand compactness
307	probably has a negative effect on sandhopper abundance beyond a certain threshold: a
308	particularly compact substrate could hinder burrowing, thus reducing the probability of
309	survival. However, sites like BV and SF, where sandhoppers were present, had more
310	compact sand than O1, where sandhoppers were never found. The negative correlation
311	at the Feniglia sites probably depended on the fact that the increasing order of sand
312	compactness at the sites corresponded to an increasing order of tourist impact.
313	Furthermore, the sand compactness at O1, where sandhoppers were absent, was lower
314	than at FB2 and FB1 where sandhoppers were present. Therefore, sand compactness
315	does not seem to be a primary determinant of T. saltator population density.

316 There did not appear to be any relationship between sandhopper abundance and 317 sand grain size at the various sites. In fact, T. saltator lives on beaches with sand that 318 ranges from medium- to coarse-grained (i.e. from around 0.5 to 2 mm, Ugolini et al., 319 1995). All the locations considered in this study fell within this range. 320 The results for organic carbon suggested a negative correlation with sandhopper 321 abundance at the Feniglia sandbar sites. However, as for sand compactness, the 322 increasing order of organic carbon content at the sites corresponded to an increasing 323 order of tourist impact. In addition, the FM site (MSRM Park) had a high sandhopper 324 abundance but a higher organic carbon content than at FA where there were very few 325 sandhoppers (but many swimmers). 326 The trace metal analysis confirmed T. saltator's good ability to bioaccumulate 327 Hg, Cu, Cd and Zn (Rainbow and Moore, 1986; Moore and Rainbow, 1987; Rainbow, 1998; Rainbow et al., 1998a, b; Marsden and Rainbow, 2004; Ugolini et al., 2004, 328 329 2005). Although there were some differences in trace metal contamination among sites, 330 there was no clear relationship with the abundance of sandhoppers. Hg accumulation 331 seemed to be negatively correlated with sandhopper abundance at the various sites (Fig. 332 4). Nevertheless, the Hg concentration in the sand was lowest at O1 where there were 333 no sandhoppers. Thus, the absence of sandhoppers at O1 and the low density at FA 334 cannot be attributed to Hg contamination. Furthermore, the trace metal concentrations in 335 both the sandhoppers and sand samples were much lower than those recorded 336 previously at heavily polluted localities where sandhoppers or beachflies were present 337 (Rainbow et al., 1998a, b; Fialkowski et al., 2000; Ugolini et al., 2004). 338 Since we did not evaluate the effects of beach cleaning separately from the 339 effects of trampling, we cannot exclude a possible influence of beach cleaning on 340 sandhopper density. Nevertheless, at the sampling sites not subjected to mechanical 341 beach cleaning, sandhopper abundance still varied in relation to the different tourist 342 impact. In fact, the sandhopper distributions along the Feniglia sandbar (where the 343 beach is regularly cleaned) and at Fiume Morto Vecchio and Batteria Vecchia (where 344 beach cleaning is not carried out) all presented the same pattern in relation to the 345 numbers of swimmers. Therefore, contrary to the hypothesis of Fanini et al. (2005), 346 trampling rather than beach cleaning has a negative effect on the survival of 347 sandhoppers. We can also add a qualitative datum: at site O1, the one with the highest

348 tourist impact in the MSRM Park, we never captured any sandhoppers during the 349 present research, whereas hundreds of sandhoppers were regularly collected around 25 350 years ago when the zone had no beach resorts and swimming was forbidden. 351 We studied populations living within 200 Km of each other and thus subject to 352 very similar general climatic and environmental conditions. Therefore, the natural 353 fluctuations of population density are probably synchronous. In any case, since the 354 proposed method is based on the comparison of densities recorded at the various sites in the same time period, it is independent of natural population fluctuations and does not 355 356 require biodiversity estimates for the considered environment. Hence, the method is 357 simple and low-cost. Other recently proposed methods using sandhoppers to assess 358 human disturbance are based on quantification of the variability of spatial-temporal 359 habitat use (ElGtari et al., 2002; Nardi et al., 2003; Fanini et al., 2005; Scapini et al., 360 2005) or on determination of the degree of genetic variability (Ketmaier et al., 2003). However, T. saltator's high behavioral plasticity and remarkable ability to learn new 361 362 directions of the sea-land axis of the beach (see Ugolini and Macchi, 1988; Ugolini and 363 Scapini, 1988; Ugolini et al., 1988) prevent a reliable and rapid determination of the 364 reasons for variability in orientation behavior. The determination of genetic variability is interesting but not easily performed. Therefore, since these methods require specific 365 366 expertise in various fields, sophisticated analytical techniques and complex statistical analyses, their practical application appears to be problematic. 367 368 The effect of human trampling and heavy tourist use of beaches on sandhopper

populations raises the question of their conservation. *T. saltator* is a good osmoregulator
(Morritt, 1988; Calosi et al., 2007) and metapopulations of this species are also found
on the shores of retrodunal pools and on riverbanks near the mouth (Ugolini and
Macchi, 1988; Ugolini and Scapini, 1988; Ugolini et al., 1988). These places are usually
shunned by swimmers and if protected could be valid reserves for natural (spontaneous

374 migrations) or artificial sandhopper repopulation of the adjacent beaches.

In conclusion, 1) our method is reliable and low-cost, 2) *T. saltator* is a good biomonitor of trace metal pollution (Cu, Hg, Zn, Cd), and 3) this species is a valid bioindicator of the effects of human trampling and could be used for ecocompatible management of the tourist impact on the supralittoral zone.

379

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506	Table captions
507	
508	Table 1. Captured sandhoppers and number of swimmers (mean $\pm$ SD; n = 3) for each
509	month of sampling
510	
511	Table 2. Results of the granulometric analysis for each sampling site. For further
512	explanations, see Figs. 1, 3 and text.
513	
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517	Figure captions
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519	Fig. 1. Frequency (% of total numbers) of captured sandhoppers (black bars) and
520	swimmers (white bars) for each month of sampling. The order of the sampling sites is
521	based on the number of swimmers observed at each locality: FM = Fiume Morto; BV =
522	Batteria Vecchia; SF = Serchio Fiume; SM = Serchio Mare; O1 = Oasi 1. The total
523	numbers of sandhoppers and swimmers at all five sites are also given.
524	
525	Fig. 2. Monthly variation of sandhopper numbers (% of total sampled) at Fiume Morto
526	(FM). Total number of sandhoppers collected = $5199$ .
527	
528	Fig. 3. Frequency (% of total numbers) of captured sandhoppers (black bars) and
529	swimmers (white bars) along the Feniglia sandbar in July and September. The order of
530	the sampling sites is based on the number of swimmers observed at each locality: FA =
531	Feniglia bathhouse, $FB1 = 1$ km from bathhouse, $FB2 = 2$ km from bathhouse. The total
532	numbers of sandhoppers and swimmers at all three sites are also given.
533	
534	Fig. 4. Trace metal concentrations ( $\mu g g^{-1}$ dry wt) in the sand (white bars) and in the
535	sandhoppers (black bars). For each element, the sampling sites are ordered according to
536	decreasing frequency of sandhoppers recorded in July 2006. For further explanations,
537	see Figs. 1, 3 and text.

538

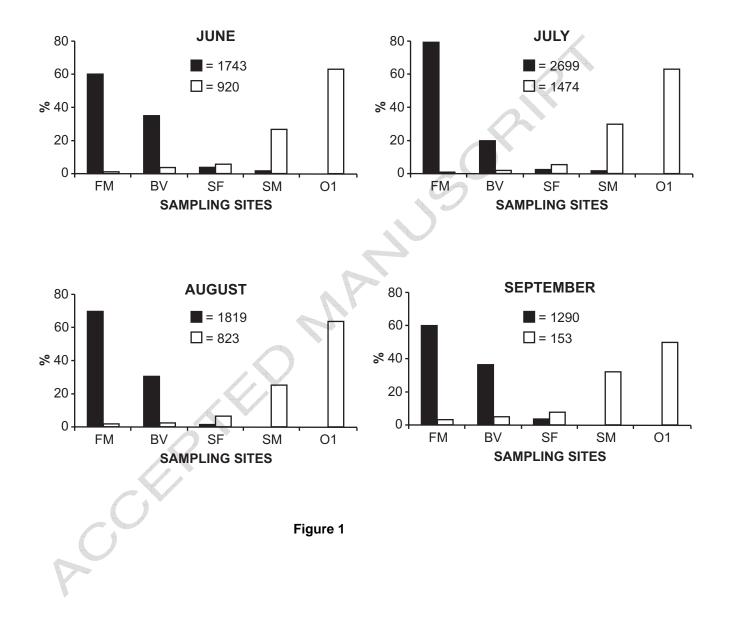
539 Fig. 5. A-B: mean sand grain diameter (white bars) and frequency of captured

- 540 sandhoppers (black bars; % of total numbers). The order of the sampling sites is based
- 541 on mean diameter. C-D: sand compactness (white bars) expressed in arbitrary units (a.
- 542 u.) and frequency of captured sandhoppers (black bars). The order of the sampling sites
- 543 is based on sand compactness. E-F: percentage of organic carbon in the sand (white
- 544 bars) and frequency of captured sandhoppers (black bars). The order of the sampling
- 545 sites is based on percentage of organic carbon. For further explanations, see Figs. 1 and
- 546

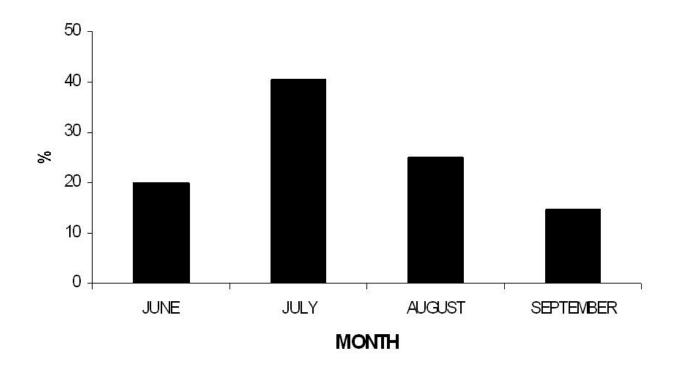
3.

- 547
- 548 Fig. 6. Frequency of surviving sandhoppers subjected to the trampling test (black bars)
- and controls (white bars). Before = before the test, After = after the test, 24h = 24 hours
- after the beginning of the test. See text for further details.

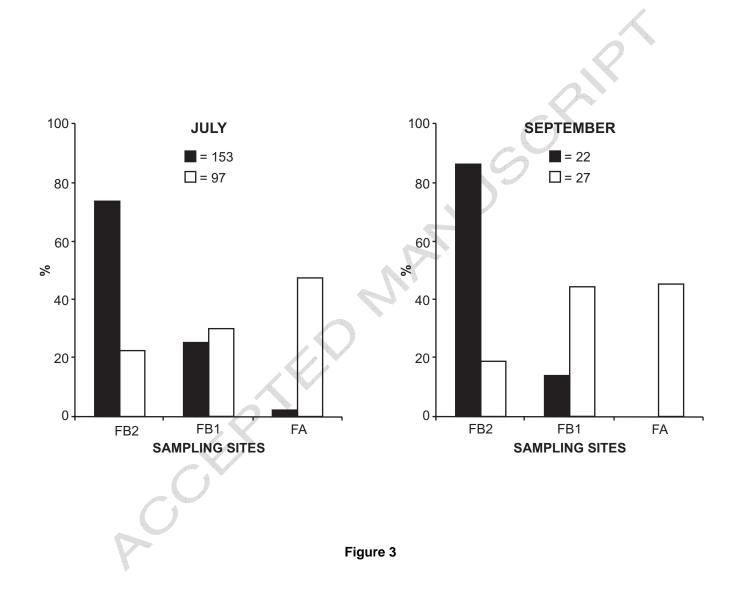
COF.F



 $\boldsymbol{\mathcal{A}}$ 







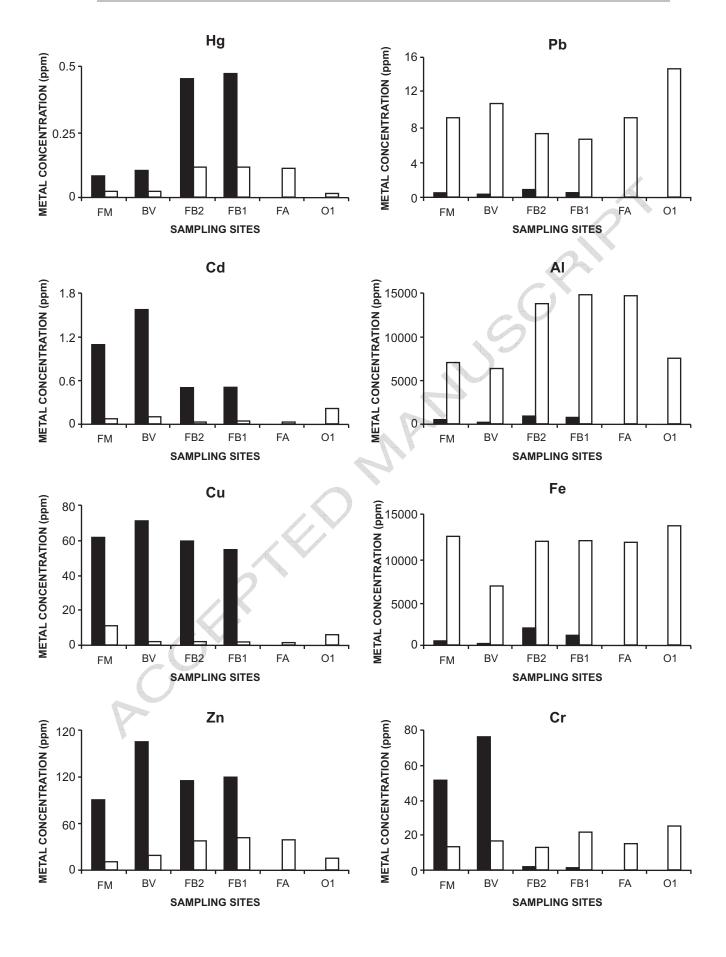


Figure 4

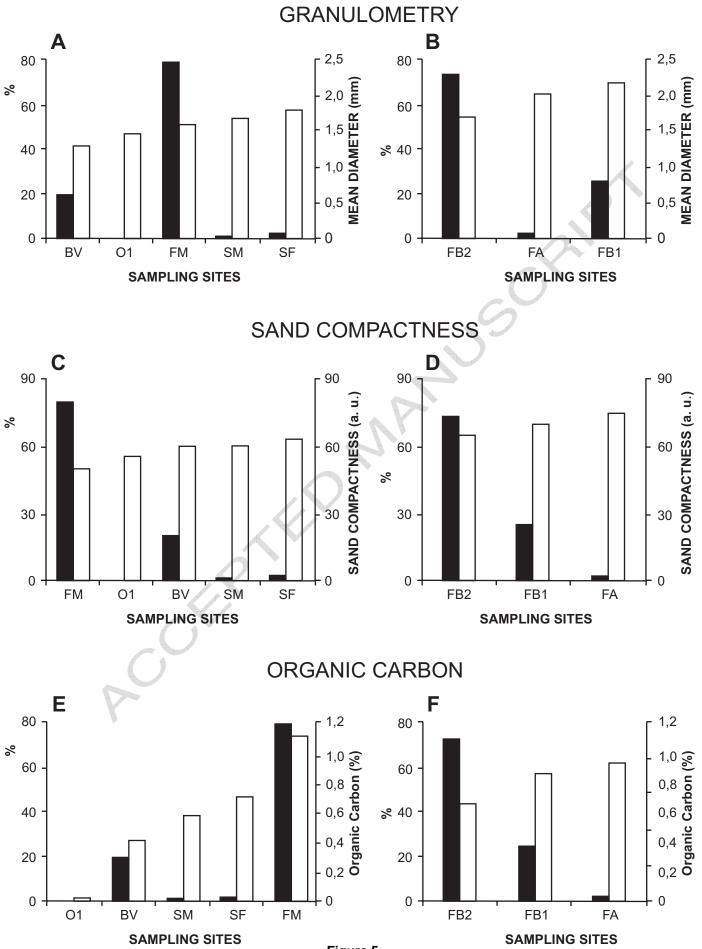
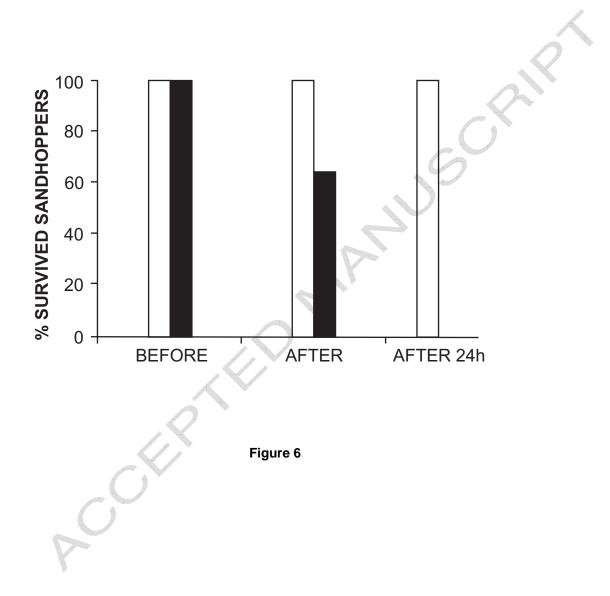


Figure 5



Sampling sites		June July Augu		August	gust September	
FM	sandhoppers	348.3±59	709±332	438±466	256±325	
	swimmers	1.3±1.5	4±2	3.6±4.4	1.66±1.5	
BV	sandhoppers	204±85	174±40	184.6±104	158±192	
	swimmers	10±13	6±2.6	6.6±9	1.3±1.5	
SF	sandhoppers	21±31	15±15	2.3±4	16.3±18	
	swimmers	18±19	25.6±15	18.6±12	4.3±5	
SM	sandhoppers	8±12	1.33±1.5	0	0	
	swimmers	82±70	146±137	70±21.2	16.6±13.5	
01	sandhoppers	0	0	0	0	
	swimmers	196±190	310±134	175±76	28.6±17.8	

Table

Sampling sites	Mean sand grain diameter (mm)	Standard deviation	Skewness	Kurtosis				
FM	1.605	0.471	-0.012	0.503				
BV	1.324	0.507	0.247	0.494				
SF	1.751	0.506	0.180	0.598				
SM	1.669	0.469	0.051	0.582	$ \land $			
01	1.484	0.486	0.170	0.543	Q			
FB2	1.738	0.436	0.234	0.631				
FB1	2.166	0.435	-0.043	0.511				
FA	2.032	0.489	0.065	0.552				
Table								