

Olfactory detection of dimethyl sulphide in a krill-eating Antarctic penguin

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Running title: DMS detection in an Antarctic penguin

1 ABSTRACT: In response to zooplankton grazing, phytoplankton release
2 dimethylsulfoniopropionate in the seawater that is catabolized to dimethyl sulphide
3 (DMS) that is emitted to the air. So, this molecule signals areas of high productivity in
4 the oceans, and it can be used by predators for locating foraging areas. Detection of this
5 compound has been described in several species of procelariiform seabirds and non
6 Antarctic fish-feeding penguins. However, there is no evidence of DMS detection by
7 krill-feeding penguins. The mechanisms of krill detection by its predators are especially
8 relevant in Antarctica, where trophic webs are mainly based on krill. We explore for the
9 first time whether a krill-feeding penguin species, the chinstrap penguin *Pygoscelis*
10 *antarctica*, is able to detect DMS. We examined whether chinstrap penguins could
11 detect DMS by locating DMS or control recipients in pathways that penguins used when
12 moving between the colony and the sea. We also analysed the attraction of nestling
13 penguins to DMS in a T-shaped enclosure. Our results showed that adult penguins are
14 attracted to DMS on land. Nestling penguins also tended to be attracted to DMS scent.
15 Further research is needed to examine whether chinstrap penguins use the natural DMS
16 concentration as a foraging cue at sea.

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18 KEY WORDS: Dimethyl sulphide · Avian olfaction · Antarctic penguin · *Pygoscelis*
19 *antarctica*

20

INTRODUCTION

21

22

23 The role of chemical signals in both intraspecific and interspecific relationships of birds
24 was first of all studied in birds with bigger-than-average olfactory bulb sizes such as
25 Procellariiformes (Bang & Cobb 1968). However, subsequent evidence suggests that
26 birds with smaller bulbs, such as Passeriformes, can also detect odours in different
27 contexts and with several functions. At the intra-specific level, olfaction based on
28 chemical compounds emitted by birds may play a key role in social behaviour (Caro &
29 Balthazart 2010, Hagelin 2007a, b). Birds have been shown to use chemical cues to
30 identify their own nest (e.g. Bonadonna et al. 2004, Caspers & Krause 2011).
31 Procellariiformes are able to discriminate the scent of their partners from the scent of
32 other conspecifics (Bonadonna & Nevitt 2004). Recently, it has been shown that birds
33 of diverse groups including Procellariiformes (Bonadonna & Sanz-Aguilar 2012),
34 Passeriformes (Krause et al. 2012) and Sphenisciformes (Coffin et al. 2011) use scent
35 for kin recognition. Psittaciformes (Zhang et al. 2010) and Passeriformes (Whittaker et
36 al. 2011, Amo et al. 2012) can discriminate the sex of conspecifics by using chemical
37 cues alone. It further appears that chemical cues affect how birds interact with other
38 species and their abiotic environment. For example, blue tits and starlings can use the
39 sense of smell to discriminate aromatic plants (Petit et al. 2002, Mennerat et al. 2005,
40 Gwinner & Berger 2008). Homing pigeons use their chemosensory abilities for
41 orientation and navigation (Wallraff 2004). Zebra finches (Kelly & Marples 2004) and
42 chickens (Marples & Roper 1996) can use the sense of olfaction to accept novel foods.
43 Passerines such as blue tits, great tits and house finches are also able to use chemical
44 cues released by predators to assess the level of predation risk (Amo et al. 2008, 2011,

45 Roth et al. 2008). These results show that birds have the ability to detect chemical cues
46 in both intra and interspecific interactions (Hagelin 2007a, Hagelin & Jones 2007).

47 Some of the most interesting interactions mediated by chemical cues are those
48 affecting several levels of a food web. In response to zooplankton grazing,
49 phytoplankton (e.g. *Phaeocystis* algae) release dimethylsulfoniopropionate (DMSP) to
50 the seawater (Pohnert et al. 2007). This compound attracts the predators of the
51 zooplankton, such as pelagic fishes (DeBose & Nevitt 2007, DeBose et al. 2008).
52 DMSP is catabolized to dimethyl sulphide (DMS) that is emitted to the air from the
53 water surface (Pohnert et al. 2007). DMS production is also higher when there is a high
54 productivity of algae (Nguyen et al. 1988). This fact is especially patent in polar areas
55 (Crocker et al. 1995). Hence, DMS signals areas of high productivity in the oceans
56 (Nevitt 2000, 2011). Recently, it has been demonstrated that several seabird species are
57 able to use DMS to locate these productive areas (Nevitt et al. 1995, Nevitt 2000, 2011).
58 Therefore, with this interesting mechanism, phytoplankton might attract the predators
59 (fishes and birds) of the zooplankton that is feeding on it (see Nevitt 2011 for a review).
60 Up to now detection of this compound has been described in several species of
61 procelariiform seabirds (Nevitt et al. 1995, Nevitt & Haberman 2003, Bonadonna et al.
62 2006, Nevitt 2008), and the African penguin *Spheniscus demersus* (Cunningham et al.
63 2008, Wright et al. 2011) that prey essentially on fish. It has also been suggested for
64 Humboldt penguins (Culik 2001). These penguin species forage on fish, and there is no
65 evidence of DMS detection in krill-feeding penguins. Therefore, to obtain
66 generalizations that allow a better understanding of how multitrophic interactions are
67 mediated by chemical cues in natural ecosystems, and of the response of birds to these
68 cues, it is essential to extend the knowledge about this mechanism to krill-feeding
69 species.

70 The mechanisms underlying prey detection in krill-feeding species are especially
71 relevant in the Antarctica, where trophic webs are mainly based on krill (*Euphausia*
72 sp.). Krill feeds on the phytoplankton that lives in the sea-ice interface. There is a clear
73 relationship between the temperature increase, consequence of climate change, and the
74 reduction in sea ice coverage, the decrease in phytoplankton and decrease in krill
75 density (Atkinson et al. 2004, Trivelpiece et al. 2011). As a consequence of such
76 decrease, populations of predators, especially krill-feeding penguins, have also been
77 affected (Fraser & Hoffman 2003). The strong correlations between penguin numbers
78 and krill abundance suggest that penguins may live under an increasingly krill-limited
79 system that negatively affects juvenile birds (Hinke et al. 2007). Within this context, it
80 is especially important to study the factors affecting the foraging success of Antarctic
81 penguins, and within these factors, the mechanisms underlying the detection of prey,
82 especially krill, that constitutes 90% of the diet in some species (Williams 1995),
83 reaching the 99 % of the diet in the chinstrap penguin (Polito et al. 2012).

84 We explore for the first time whether a penguin species that feeds mainly on
85 krill (Rombolá et al. 2006), the chinstrap penguin *Pygoscelis antarctica*, is able to
86 detect DMS. The chinstrap penguin is an Antarctic species that depends mainly on local
87 krill resources for the daily provisioning of chicks during the breeding period. This krill
88 dependence is so strong that chinstrap penguins increase the distance of their foraging
89 trips in order to obtain krill whereas other Pygoscelid species such as the Gentoo
90 penguin (*P. papua*) are more flexible and able to change their diet in response to the
91 availability of prey instead of increasing distance of their foraging trips (Kokubun et al.
92 2010, Miller et al. 2010). This species is one of the major consumers of Antarctic krill
93 in the Southern Ocean marine ecosystem (Croxall & Lishman 1987, Williams 1995,
94 CEMP 2004), and it has been included in the Ecosystem Monitoring Programme of the

95 Convention for the Conservation of Antarctic Marine Living Resources (CEMP,
96 CCAMLR) to monitor changes in krill populations (Rombolá et al. 2006). Therefore,
97 the use of DMS for locating krill may be especially important for optimizing foraging
98 during the austral summer. Despite Sphenisciform chinstrap penguins may have a
99 reduced olfactory bulb size compared to Procellariiformes (Bang & Cobb 1968), in this
100 species, the recognition of DMS could be under strong natural selection, as chinstrap
101 fledglings must find suitable food resources without prior foraging experience (Hinke et
102 al. 2007). The first few weeks of independence for fledgling penguins represent a
103 potential bottleneck to recruitment (Moreno et al. 1999, Hinke et al. 2007). Thus, the
104 use of DMS for finding krill may be relevant for recently independent fledglings. We
105 examined whether chinstrap penguins could detect DMS by locating DMS or control
106 recipients in pathways that penguins used to go from the colony to the sea and vice
107 versa (Cunningham et al. 2008). We also analysed the attraction to DMS of nestling
108 penguins in a T-shaped enclosure (Cunningham et al. 2008). We hypothesized that
109 naïve nestlings of chinstrap penguins may be able to detect DMS.

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MATERIAL AND METHODS

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Study site

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116 We performed an experiment in natural conditions at a breeding rookery (12,000
117 breeding pairs, Barbosa et al., unpublished data) of chinstrap penguins *Pygoscelis*
118 *antarctica* in the Vapour Col rookery on Deception Island, South Shetlands (63°00'S,

119 60° 40'W) during the austral summer (January/February) of 2011. Experiments were
120 performed from 11:00 to 17:00 hours.

121

122 **Response of adult penguins to DMS in natural conditions**

123 We located 13 separate observation points in different pathways that penguins used to
124 go to forage from the colony to the sea and vice versa. These points were in different
125 parts of the colony to minimize the proportion of individuals resampled. The colony
126 was situated in several hills and penguins need to go down from the colony to the sea.
127 The slope of the pathways differed between observation points. In each observation
128 point we placed a Petri dish and we marked the point with 2 metal rods situated 1 meter
129 from the Petri dish in each direction along the path. We deployed DMS or control
130 solution in the Petri dish for a period of 30 minutes. After that, we changed the Petri
131 dish for a clean new one and added the other treatment in order to do repeated
132 measurements in each point. The order of treatments was randomised across sampling
133 points. We added 7,85 ml of a DMS solution ($0,002 \text{ molml}^{-1}$) to 17,15 ml of water in a
134 petri dish to obtain a volume of 25 ml. The control solution was prepared with 7,85 ml
135 of vegetable oil and 17,15 ml of water. The vegetable oil has a detectable scent to
136 humans, suggesting that birds had to discriminate between two scented compounds
137 rather than the presence or absence of odour. An observer, with knowledge of the
138 treatment, was situated approximately 20 meters from the observation point and
139 recorded the time that randomly selected penguins ($n = 1084$) spent within the 2 m
140 sector. In order to have independent data and as many penguins were continuously
141 walking close to the points, we recorded the behaviour of only one penguin at a time,
142 and when it passed we recorded the behaviour of the next penguin that entered within
143 the 2 m sector. We distinguished between penguins going to forage at sea or returning

144 to their nest after a foraging trip. We measured wind speed (mean 20 km/h), and
145 temperature (mean 3 °C) with a Kestrel Weather K3000 Wind Meter. We noted the
146 wind direction (from the sea to the land or from the land to the sea).

147 We used a General Linear Mixed Model to analyse differences between
148 treatments (DMS vs. control) in the time spent close to the stimuli. We included the
149 direction of birds (from the colony to the sea vs. from the sea to the colony), the
150 direction of the wind (from the sea to the land vs. from the land to the sea), and the
151 order of treatment presentation in the model as a fixed factors, and sampling location as
152 a random factor. We also included the interactions between treatment and the direction
153 of penguins, between treatment and direction of wind and between treatment, direction
154 of penguins and direction of wind in the model. Data were log-transformed to ensure
155 normality.

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157 **Response of nestling penguins to DMS in a T-shaped enclosure**

158 We performed an experiment to examine the attraction of nestling penguins to DMS.
159 The experiment was performed in a T-shaped mesh enclosure, built with 50 cm high
160 chicken wire (mesh size 1.3 cm) and located in front and 50 m far from the closest
161 breeding sub-colony, and also in front and 300 m far from the sea coast (Fig. 1). In that
162 way, both experimental arms of the enclosure were equally distant from the colony and
163 to sea, and we avoided any confounding effect due to the possible attraction of nestlings
164 to such places. We used a mesh enclosure since chinstraps are not burrow nesters. Both
165 the “vertical” and “horizontal” segments of the T were 150 x 50 cm corridors (Fig. 1).
166 Just outside these arms, in the farthest side from the central arm, we placed two Petri
167 dishes, one with DMS and the other one with a control solution. Therefore, nestling
168 where offered a simultaneous choice between two stimuli: DMS and a control

169 (vegetable oil). Both DMS and control solution were prepared following the same
170 methodology as in the previous experiment. The location of treatments in the enclosure
171 was balanced between both sides of the enclosure between trials. After each trial, the
172 enclosure was clean with ethanol. We performed the experiment on days with no or low
173 wind, that always blew from the sea to the land. The location of the maze, that was
174 situated cross wind ensures that both petri dishes were equally exposed to the wind (Fig.
175 1).

176 We used a long-handle net to capture 35 nestlings during the crèche phase. To
177 minimise time in captivity, after a brief habituation of 3 minutes period at the base of
178 the central arm (habituation area, Fig. 1) nestlings were released into the choice area,
179 from which they could enter the left and right arms (experimental areas, Fig. 1) of the T-
180 maze.

181 In order to determine whether the number of nestlings that preferred the DMS-
182 scented arm differed from the number of nestlings that preferred the control arm of the
183 enclosure, an observer situated 20 m from the enclosure and blind to treatments
184 recorded the time that nestlings spent in each sector of the enclosure during 5 minutes.
185 After trials nestlings were marked with an indelible pen in one foot to avoid recapture
186 and immediately released in the exact place where they had been captured. Birds were
187 kept in captivity a maximum of 15 minutes. All the birds showed a normal behaviour
188 after released, i.e. nestlings joined some other nestlings forming a crèche in the breeding
189 sub-colony as before capture.

190 We calculated the number of birds that spent more time in the DMS than in the
191 control sector during the experiment. We built Generalized Linear Models with
192 binomial errors and a logit link function (GLM) to analyse whether the number of birds
193 that spent more time in the DMS sector was significantly different from the number of

194 birds that spent more time in the control sector. We included the side of the enclosure
195 (left vs. right) where the DMS was located as a fixed factor. Statistical analyses were
196 performed with STATISTICA 8.0.

197 Although we performed the same experiment with adult penguins, we will not
198 report the results of the study because most adult penguins spent a fair amount of time
199 trying to escape from the enclosure. Comparison with previous studies suggests two
200 methodological issues that may have exacerbated the problem of stress and should be
201 avoided in future experiments. First, the use of adult, recently caught birds –
202 Cunningham et al. (2008), for instance, used penguins from a rehabilitation centre.
203 Second, the use of a chicken-wire enclosure that allowed penguins to see their colony
204 could increase their motivation to escape. In contrast, in the previous study with adult
205 penguins (Cunningham et al. 2008), as well as other studies with procellariiforms (e.g.
206 Nevitt & Bonadonna 2005, Bonadonna et al. 2006), the experiments were performed in
207 opaque close enclosures, where subjects could not see their environment, possibly
208 decreasing their stress response (Cockren et al. 2008).

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RESULTS

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Response of adult penguins to DMS in natural conditions

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Adult penguins spent more time within one metre of the Petri dish when it contained
DMS solution than when it contained a control solution (GLMM, $F_{1,1037} = 12.85$, $p =$
0.0004, $n = 1084$). However, the time spent close to the DMS was affected by the
interaction between wind direction and penguin direction (interaction between
treatment, wind direction and penguin direction: $F_{2,1037} = 17.27$, $p < 0.0001$). When

219 penguins faced the wind they spent more time close to the DMS solution than to the
220 control solution, but the time spent within one metre of the Petri dish did not differ
221 between treatments when penguins moved with the wind (Fig. 2). The order of
222 treatment presentation did not influence the time spent close to the stimuli ($F_{1,1037} =$
223 0.36 , $p = 0.55$). There were differences among observations points (random factor;
224 $F_{12,1037} = 12.95$, $p < 0.0001$). These differences between points were due to the
225 differences in the slope of the pathways where the observation points were located, so
226 penguins walked more slowly in some points than in others. However, such differences
227 between points did not influence the effect of treatment because the interaction between
228 treatment and point was not significant ($F_{9,1028} = 0.81$, $p = 0.60$) when we considered it
229 in a previous model.

230

231 **Response of nestling penguins to DMS in a T-shaped enclosure**

232 Most nestlings (66 %, 23 of 35) spent more time on the DMS than on the control sector,
233 although differences only approached significance levels (Wald Stat = 3.41, d.f. = 1, $p =$
234 0.06 , $n = 35$; Fig. 3). The arm of the enclosure where the DMS dish was located did not
235 influence the preference of nestlings (Wald Stat = 0.35, d.f. = 1, $p = 0.56$).

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238 **DISCUSSION**

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240 Our results show for first time that a krill-eating penguin is able to detect DMS. Adult
241 chinstrap penguins that walked into the wind spent more time close to the DMS
242 deployments than close to the control deployments. These penguins were probably able
243 to detect the DMS before reaching the 2 meter sector and they might be following the

244 DMS-scented trace upon arrival to this sector. However, there were no differences in
245 the time penguins spent close to both olfactory stimuli when they were walking with the
246 wind, probably because these penguins would not be able to detect the DMS scent until
247 they passed the odor source. We performed a repeated measures analysis, applying both
248 treatments at each observation point, so we may attribute differences in the response of
249 penguins to the DMS to the direction of the wind relative to the direction they were
250 travelling. Our results also agree with those of Cunningham et al. (2008) and Wright et
251 al. (2011) that showed that African penguins responded to DMS deployments on land.
252 We used vegetable oil as a control scent, so our results clearly show that the attraction
253 of penguins to DMS seems to be specific to DMS rather than a general response to any
254 novel scent.

255 From our results it can also be proposed the idea that nestlings can detect and
256 tended to exhibit a preference for DMS before they experience it in a foraging context at
257 sea. When offered the choice between a DMS and a control source in an enclosure, most
258 nestlings preferred to stay in the DMS-scented part of the enclosure, although results
259 only approached significance levels ($p = 0.06$). Stress due to recent captivity (Cockren
260 et al. 2008) may explain why we did not observe a greater attraction to DMS in
261 enclosures. Thus, a previous study that has shown a positive response of adult African
262 penguins to DMS in an enclosure (Cunningham et al. 2008), was run with penguins
263 coming from a rehabilitation Centre. These captive penguins were possibly habituated
264 to humans, not under the stress of just being caught as in our study.

265 Our results are in accordance with those of Bonadonna et al. (2006), who clearly
266 showed that blue petrel, *Halobaena caerulea*, chicks were even able to detect the
267 natural DMS concentration ($<10 \cdot \text{pmol} \cdot \text{l}^{-1}$). Whether the ability to discriminate DMS
268 from other scents is innate or learned during the nestling period cannot be disentangled

269 in our study because, although nestlings were not previously exposed to DMS in a
270 foraging context at sea, they may have learnt to recognize the scent from krill fed by
271 their parents (Bonadonna et al. 2006). Regardless of the mechanism, an early ability to
272 detect DMS may be especially important for fledged penguins because once they reach
273 the independence age (53-57 days, Viñuela et al. 1996) they are left unattended by their
274 parents. In that moment, young penguins must go to the sea and be able to find suitable
275 food resources without prior foraging experience. Therefore, the use of DMS for finding
276 krill may be relevant for recently independent fledglings.

277 We performed the experiment on the colony, on land, an environment where
278 normally penguins do not find DMS. Furthermore, we used a concentration of DMS
279 much higher than birds may encounter at the sea (see Nevitt 2000, Nevitt & Bonadonna
280 2005), and even higher than previously used in other studies with procellariiform
281 (Cunningham et al. 2003, Bonadonna et al. 2006) and penguin species (Cunningham et
282 al. 2008, Wright et al. 2011). DMS is an irritant chemical compound, so under a high
283 concentration, we might have observed an aversive response in penguins, as it has been
284 observed with other irritant compounds such as ammonia in other bird species
285 (Kristensen et al. 2000). However, despite of that, we found that penguins spent more
286 time close to the DMS than to the control stimuli, and therefore our results gives first
287 evidence that this species is able to detect this chemical compound. Further research is
288 needed to determine whether chinstrap penguins can detect the natural concentration of
289 DMS and use it as a foraging cue at the sea.

290 DMS signals areas of high concentration of krill in the oceans (Nevitt 2000,
291 2011). Although penguin colonies may be located where local oceanic circulation or
292 bathymetry concentrates food and promotes access to foraging areas (Fraser &
293 Trivelpiece 1996, Trivelpiece & Fraser 1996, Hinke et al. 2007), the use of DMS

294 gradients may help chinstrap penguins to maximize their foraging efficiency, especially
295 because this species feed almost exclusively (99 %) on krill, and previous studies about
296 its diet and habitat use have shown that, when krill is not available close to the colony,
297 chinstrap penguins travel longer distances to find krill instead of capturing other prey as
298 other Pygoscelid penguins do (Kokubun et al. 2010, Miller et al. 2010). This may be
299 especially important during breeding, when penguins need not only find food for
300 themselves but must also comply with the daily provisioning of chicks. When travelling
301 to their feeding areas penguins emerge to the water surface to breath, and in that
302 moment they may obtain information about DMS gradients – information that they can
303 use to modify their travelling direction.

304 Detection of DMS has also been observed in other species, mainly
305 procellariiform seabirds such as blue petrels *Halobaena caerulea* (Bonadonna et al.
306 2006) and Antarctic prions *Pachyptila desolata* (Nevitt & Bonadonna 2005). African
307 penguins are able to detect the DMS (Cunningham et al. 2008, Wright et al. 2011). This
308 penguin species feeds predominantly on anchovies (*Engraulis* sp.) and sardines
309 (*Sardinops sagax*) (Crawford & Dyer 1995, Wilson et al. 1995). This species, as many
310 procellariiform species that feed on fishes and squids, may use DMS concentration to
311 locate the fishes that feed on krill, whereas chinstrap penguins may use DMS to directly
312 locate the krill concentrations they feed on. From an evolutionary point of view, the
313 phytoplankton that release DMSP – the precursor of DMS, could benefit from attracting
314 krill predators, as they decrease grazing pressure. Therefore, for phytoplankton, the
315 attraction of krill-eating species such as fishes or chinstrap penguins could be better
316 than the attraction of superpredators that feed on fishes that feed on krill, such as
317 procellariiform species or the African penguins. These species remove the predators of
318 krill and, therefore, they may impose a cost to the phytoplankton DMS signalling. A

319 balance between predatory species may occur to make these complex systems
320 evolutionary stable, as has been observed in terrestrial systems (Sabelis & Dejong 1988,
321 Godfray 1995). In terrestrial ecosystems, the role of induced indirect defences mediated
322 by chemical compounds has been largely studied in systems composed by plants,
323 herbivorous insects and predatory insects (Schoonhoven et al. 2005, Dicke & Baldwin
324 2010). When plants are wounded by herbivorous insects, they release volatile
325 compounds to attract the predators (or parasitoids) of these insects (Schoonhoven et al.
326 2005). While insectivorous birds can also use the defense of attacked plants to locate
327 their prey (Mäntylä et al. 2004, 2008a,b, 2011), in the terrestrial systems studied so far,
328 volatiles emitted by attacked plants do not seem to be used by superpredators or
329 hiperparasitoids (Buitenhuis et al. 2005, Poelman et al. 2008). Further research is
330 needed to understand the extent to which superpredators eavesdropping on the signals
331 released by plants or algae to attract predators can destabilize the evolution of induced
332 indirect defences.

333 On conclusion, although penguins were traditionally thought to be visual hunters
334 (Wilson et al. 1993, Wilson & Wilson 1995, Ryan et al. 2007), our results show that the
335 chinstrap penguin is able to detect DMS. This olfactory capacity seems to be expressed
336 even in nestlings without prior foraging experience. Further research is needed to
337 examine whether chinstrap penguins are also able to detect DMS at naturally occurring
338 concentrations, as Procellariiforms (Nevitt & Bonadonna 2005, Bonadonna et al. 2006)
339 and in natural conditions, at sea (see Nevitt et al. 1995, Nevitt 2000, Wright et al. 2011).

340

341

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530

531 Table 1. Results of the analysis of time spent close to the stimuli (DMS or control) of
 532 adult nestlings in several observation points in the pathways when they were going to
 533 sea to forage or returning to the colony from the sea (penguin direction) and when they
 534 walked with the wind or into the wind (wind direction).
 535

Factor	Effect	<i>F</i>	<i>P</i>
Treatment (DMS vs Control)	Fixed	$F_{1,1037}=12,8460$	$P=0,0004$
Order of treatment presentation	Fixed	$F_{1,1037}=0,3646$	$P=0,55$
Observational Point	Random	$F_{12,1037}=12,9528$	$P<0,0001$
Penguin direction (from vs to the sea)	Fixed	$F_{1,1037}=13,1241$	$P=0,0003$
Wind direction (from vs to the sea)	Fixed	$F_{1,1037}=2,5252$	$P=0,11$
Treatment * Penguin direction	Fixed	$F_{1,1037}=4,9286$	$P=0,03$
Treatment * Wind direction	Fixed	$F_{1,1037}=5,9322$	$P=0,02$
Treatment * Penguin direction * Wind direction	Fixed	$F_{2,1037}=17,2655$	$P<0,0001$

536

537

538

539 **Figure legend**

540 Fig. 1. The enclosure where the experiment was carried out, with sizes indicated in
541 meters. Black points represent the Petri dishes where the correspondent treatment was
542 added (DMS vs. control). The enclosure was perpendicularly located at 50 m from the
543 nearest sub-colony and 300 m from the sea coast.

544

545 Fig. 2. Mean (\pm SE) time spent (sec) by Chinstrap penguins within two metres of a Petri
546 dish with 25 ml of DMS (close squares) or control (open circles) solution. Data are
547 presented separately for penguins that were going from the colony to sea and penguins
548 that were returning from the sea to the colony, and when the wind was blowing from the
549 sea or towards the sea.

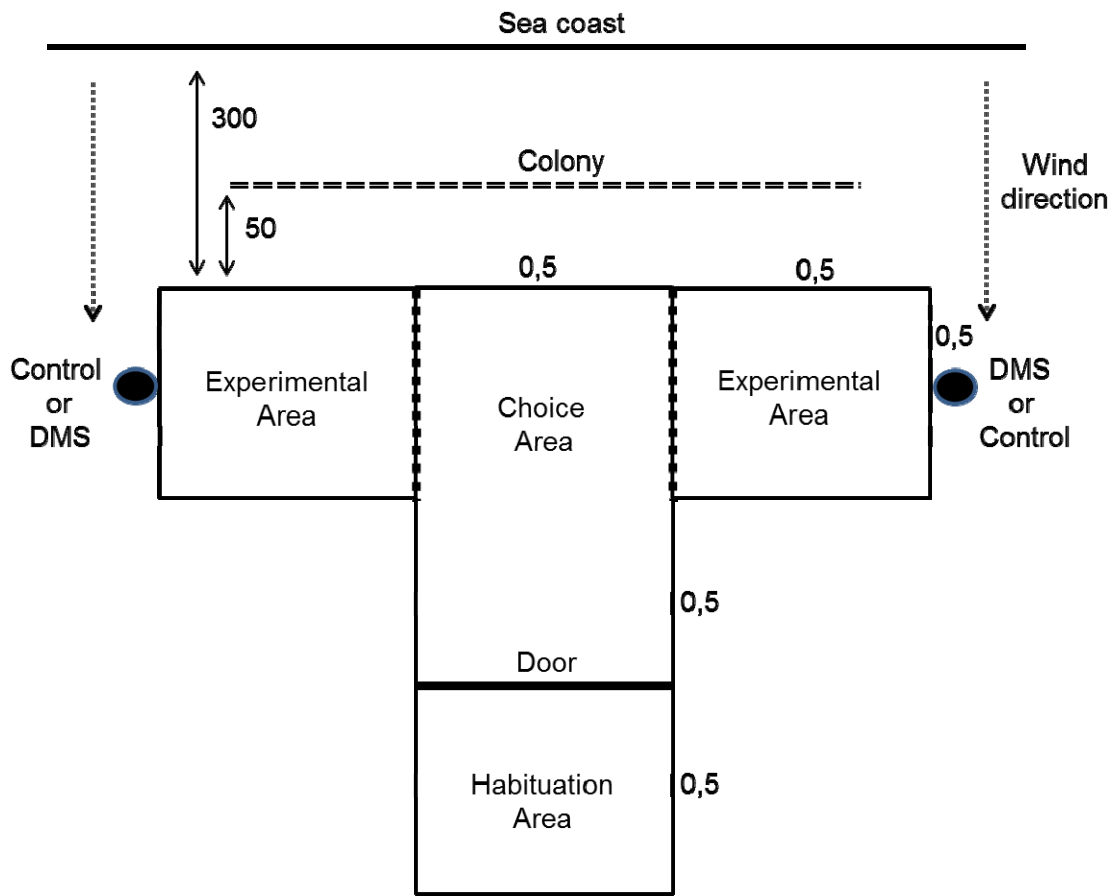
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551 Fig. 3. Number of nestling chinstrap penguins that spent most of the time in the DMS or
552 the control side of the enclosure.

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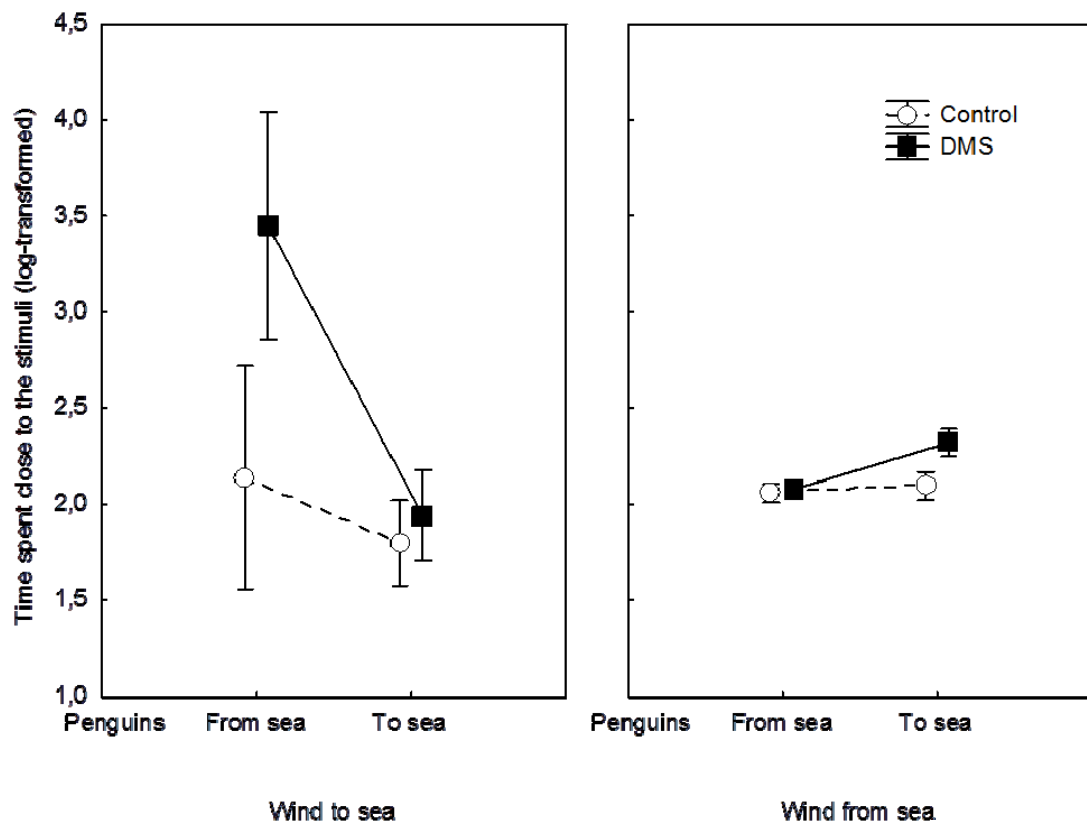
555 Fig. 1



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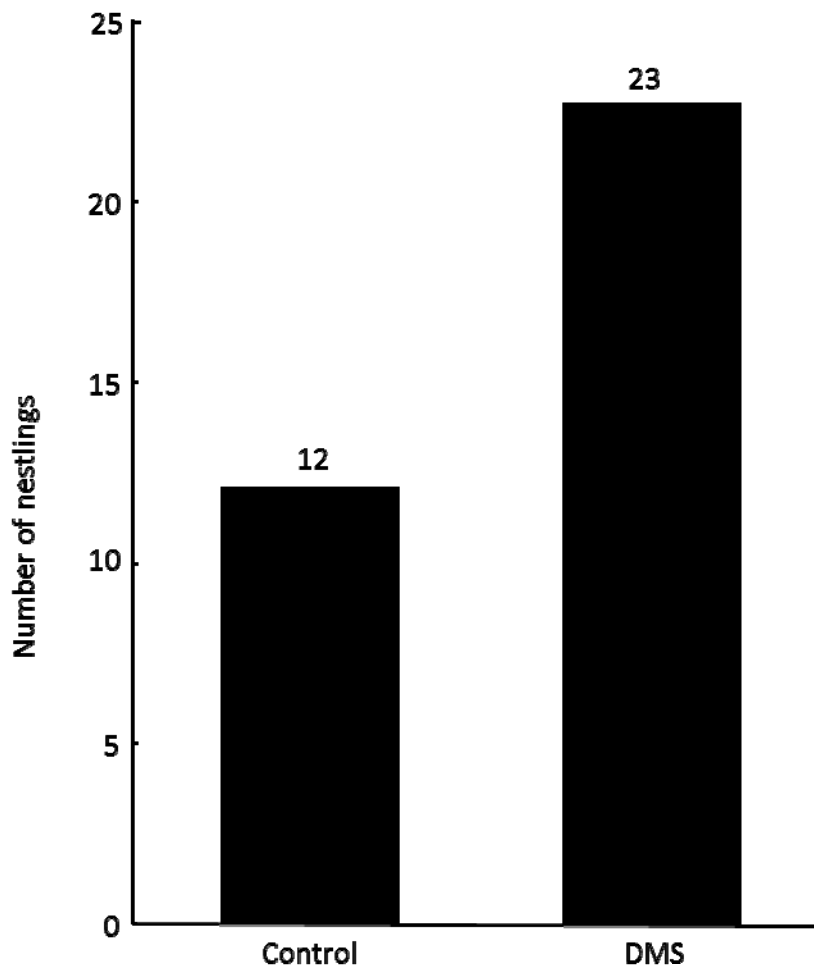
558 Fig. 2



559

560

561 **Fig. 3**



562