1	Enrichment with classical music enhances affiliative behaviours in bottlenose dolphin
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- 21 Abstract
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23 Environmental enrichment is a crucial element for the promotion of welfare of animals kept under human 24 care. While a large variety of environmental enrichments has been proposed and studied for terrestrial animals, including a growing area represented by acoustical enrichment such as music, the same is not true 25 for marine mammals. The purpose of this study was to evaluate the ability of classical music to act as an 26 27 enrichment for bottlenose dolphins under human care (Tursiops truncatus); its effect on the dolphins' 28 behavior were compared with that of a less complex auditory stimulus (rain sound), another novel, but visual 29 enrichment (slideshow of photographs), and an already known form of enrichment (floating objects). The 30 study was conducted on two groups of dolphins (N = 6 and N = 2, respectively) housed in a dolphinarium in Riccione, Italy. Enrichments were provided for 20 minutes/day, on 7 days for each enrichment type, per 31 group of animals. Their effect was evaluated by observing changes in behaviours expressed during or shortly 32 33 after the provision of the enrichment. Some effects were unspecific, being shared by most, or all types of enrichment, including an increase of activity levels and synchronous swimming. However, only classical 34 35 music was able to increase several social affiliative behaviors both during its presentation and after its 36 removal. The results indicate that classical music has positive effects on behaviour, that qualify it as an 37 effective environmental enrichment for dolphins in this context. Some aspects remain to be elucidated, including the mechanisms by which music exerts its effects, and how specific to classical music the latter 38 39 are. Nonetheless, the specificity of effects on social behavior suggest that classical music could be 40 particularly useful when an enhancement in social behaviours is needed.

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42 Keywords: bottlenose dolphin, cetacean, classical music, enrichment, social behaviour, zoo

44 **1. Introduction**

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46 The bottlenose dolphin (Tursiops truncatus) is the most popular cetacean under human care, representing on its own 87% of cetaceans in closed facilities (Brando et al., 2018). In those conditions, due to their nature, 47 48 bottlenose dolphins are already subjected to a high stress level (Ugaz et al., 2013). Therefore, different 49 strategies are used to improve their living condition in ensuring a satisfactory quality of water (Wallis, 1973), 50 avoiding physical discomfort and providing appropriate nutritional requirements (Kastelein et al., 2002, 51 2003), preventive health care (Miller et al., 2015), and social management (Connor et al., 2000). 52 Furthermore, the quality of life of animals under human care is actively improved by increasing stimulation and/or control over the environment (Goldblatt, 1993), generally referred to as environmental enrichment. 53 Environmental enrichment is defined by Newberry (1995) as any technique designed to improve the 54 55 biological functioning of animals under human care, via changes in its environment. This includes, for example, providing "toys" or furniture, hiding food to increase forage behaviors, introducing new odorants, 56 modifying the social group and presenting the animals with puzzles, to provide structural, nutritional, 57 58 sensory, social and cognitive enrichment, respectively (Makecha and Highfill, 2018). 59 Not all new stimuli provisioned in animal's environment can be considered as an enrichment, as many 60 parameters can influence animals' response toward them. In fact, responses are depending on several aspects 61 such as, among others, the species, the sex, the social group and how the enrichment is provided. For 62 instance, the presentation of the exact same set of stimuli elicited more interest and bubble behavior in 63 rough-toothed dolphins than in bottlenose dolphins, and more aggressive behaviors were expressed by males 64 than females in both species (Winship and Eskelinen, 2018). Moreover, presenting the enrichment with 65 random schedules and session lengths thereby avoiding habituation (Kuczaj et al., 2002), and when the animals are at a high activation level, e.g., after a training session (Perez et al., 2018) helps keeping the 66 67 animals' interest toward the enrichment device. Preserving novelty characteristics of an enrichment by exchanging the items used or by randomizing the schedules of presentation is crucial to avoid habituation or 68 69 boredom (Tarou and Bashaw, 2007; Makecha and Highfill, 2018). Taken together, those studies highlight the 70 importance of considering various variables regarding the animals, their social and non-social environment, and also the modalities of the presentation of the stimuli. 71

72 Previous articles on the effect of environmental enrichment, and more specifically of sensory enrichment, in 73 cetaceans under human care are rare. Most of the existing studies examine the effect of floating objects as 74 forms of tactile and/or visual enrichment and dolphins seem to react positively to their provision on the whole (Kuczaj et al., 2002; Greene et al., 2011; Perez et al., 2018). Another type of sensory enrichment is 75 76 visual media broadcasted on a screen, in the form of videos. Hanna and co-authors (2017) analysed 77 behavioral responses of a killer whale to different videos presented with and without sound, and Winship and 78 Eskelinen (2018) observed behavioral responses of two groups of dolphins toward different video categories. 79 The behavioral response to this type of enrichment is less consistent than that towards the provision of 80 objects, as it elicits attraction (Hanna et al., 2017), but can also trigger aggressive behaviors towards the 81 enrichment (Winship and Eskelinen, 2018). To our knowledge, no studies have assessed the effect of 82 acoustical enrichment in cetaceans without any other sense at stake. 83 It is known that dolphins housed in dolphinaria are often exposed to acoustic stimulation during shows. In 84 this context, music represents a mean of entertaining humans, and it is not conceived as a potential 85 enrichment for dolphins. Moreover, uncontrollable exposure to chronic noise can impair the welfare of 86 experimental animals (Patterson-Kane and Farnworth, 2006). At the same time, hearing is a matter of

survival for dolphins. Through echolocation they create a representation of their close environment in order
to navigate and forage (Kremers et al., 2016). Moreover, acoustical stimuli also convey social (Herman and
Tavolga, 1980) and context-related information (Janik et al., 1994), and emotional states (Caldwell and
Caldwell, 1965; Kuczaj et al., 2013). Therefore, auditory enrichment could be particularly relevant for this
species, also considering that acoustical enrichment in the form of music has already proved its efficacy in

93 One growing area of research focuses on the potential effect of classical music on a wide range of animals,

94 highlighting its physiological and behavioral effects (for review, see Alworth and Buerkle, 2013 and

terrestrial and other marine animals (Alworth and Buerkle, 2013).

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95 Dhungana et al., 2018). Classical music increases affiliative behavior in laboratory housed chimpanzees both

96 during and after exposure (Videan et al., 2007), it decreases aggressive and abnormal behaviors in gorillas

97 (Wells et al., 2006) and decreases stereotypies in captive Asian elephants (Wells and Irwin, 2008). Most of

98 the studies done on rats proved that classical music exerts positive effects on stress-related behaviors and

99 physiological parameters (for a review, see Kühlmann et al., 2018). Lindig and collaborators (2020)

reviewed nine reports on "therapeutic" effects of music in stressed dogs proving that, in general, classical
music has a calming effect compared to no sound or other genres of music. Classical music also decreases
behavioral and physiological anxiety-related marker in laboratory zebrafish (Barcellos et al., 2018) and
modulates common carp physiological and metabolic states inducing a larger growth and a better food
consumption (Papoutsoglou et al., 2007, 2010). All this considered, classical music is the most studied
acoustical stimulus provided as an enrichment in a large variety of animal species, in which, overall, it seems
to have a positive effect.

107 The purpose of the current study is to assess whether classical music can serve as an enrichment for 108 bottlenose dolphins. Given the lack of scientific research about acoustical enrichment programs in dolphin 109 under human care, we decided to start using a compilation of classical pieces. Other types of stimuli were 110 provided to the same subjects to compare the effect of classical music with a sound having different 111 acoustical properties (complex vs repetitive sound), with a stimulus perceived by a different sensory channel 112 (auditory vs visual), and with an enrichment already in use (novel vs non-novel).

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114 2. Materials and Methods

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116 2.1 Subjects and housing

Eight bottlenose dolphins (Tursiops truncatus), five females (of which two wild-born) and three males (of 117 118 which one wild-born), aged between 5 and 49 years, were involved in the study (Table s1). All of the dolphins were housed at the 'Oltremare' aquatic-park, Riccione (Italy), in a dedicated area equipped with by 119 five interconnected pools (main pool: 1.173 m²; reproduction pool: 243 m²; holding pool: 160 m²; medical 120 pool: 37 m²; connecting channel: 83 m²), for a total area of 1.696 m² and 9 million L of water (Fig. S1). The 121 122 structure is realized in agreement with criteria set by the Italian legislation concerning zoological gardens 123 (Legislative Decree 73/05), and dolphinaria (Ministerial Decree 469/01). The dolphins were split in two separate groups, one comprising two adult males, the other one consisting of 124

the five females and the sub-adult male. The two groups were regularly turned between the different sectors,

so all subjects were highly familiar with the environment used as experimental site (the reproduction pool,

see details below). This grouping was maintained throughout the study, in order to not to alter the normalmanagement of the animals.

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130 2.2 Experimental setting

Experimental observations were conducted in the reproduction pool (rectangular plan for a total area of 243 131 m^2 and a depth of 3.5 m) as experimental site. This pool is separate from the exhibition area, it is out of the 132 133 view of the public and it is accessible only by the staff, ensuring an undisturbed and easily controlled 134 environment. The pool featured two round portholes (1.5 m of diameter) and a rectangular window (5 x 2.5 m). Water temperature and salinity were monitored for the entire study period and were $26.5 \pm 1.2 \text{ C}^{\circ}$ (mean 135 \pm SD) and 34,1 \pm 1,0 ‰ (mean \pm SD), respectively. An underwater speaker (Akuasound, Mausound, Modena, 136 Italy) was used to emit acoustic stimuli inside the experimental pool. The speaker was placed under the 137 pool's raisable floor, out of the dolphin's reach. The signal came through the output of a smartphone 138 connected to a power amplifier (Champion 20, Fender, Fullerton, CA, USA). A 36" LCD TV monitor 139 (Philips, Amsterdam, The Netherlands) with 1920 x 1080 resolution, was positioned behind one of the two 140 141 portholes and was used to display visual stimuli.

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143 *2.3 Enrichments*

Four types of enrichment condition were used in the study, of which one (classical music) representing the focus of the study, whereas the other three were included to assess the effects of the novelty, complexity and nature of the enrichment. The characteristics of the four conditions are described hereafter.

a) Classical music (CM): a twenty-minute audio file constituted by six pieces of classical music ("Prelude I,

148 BWV 846" by Johann Sebastian Bach; "The morning de Peer Gynt" by Edvard Hagerup Grieg; "The swan"

149 from "The Carnival of the Animals" by Charles Camille Saint-Saëns; "Reflets dans l'eau" by Claude

150 Debussy; "Almost a fantasy" by Ludwig van Beethoven). This stimulus had bandwidth of 16 kHz, with most

- intensity between 100 and 1000 Hz; while the amplitude levels were 70 and 80 dB re 1 μ Pa, respectively.
- b) Rain sound (RS): a twenty-minute audio file recreating the sound of falling rain. The acoustic
- 153 characteristics were similar to those of CM in terms of overall bandwidth and intensity. For its very nature,
- the spectral characteristics of RS were rather different from those of CM, the former being characterized by

155 large bandwidth impulsive signals with very limited amplitude variations in the frequency band, as opposed 156 to the frequency modulated harmonic sounds with amplitude variations in the frequency band typical of CM. 157 Therefore, RS was included to assess whether any effect of CM was generically due to the provision of an 158 acoustic enrichment regardless of its complexity or rather if it was specific to a complex and varied acoustic 159 stimulus;

c) Photographs of natural landscapes (PH): a twenty-minute video file constituted by 20 natural images of
natural terrestrial environments, each of which was displayed for one minute on the TV monitor. Dolphins
had never been exposed to this type of stimuli before and the inclusion of this condition allowed us to assess
whether any effect of CM was due to the nature (e.g. visual vs acoustic) of a novel enrichment.

d) Floating objects (FO): it consisted of the provision of buoys of different size and shape, volleyballs, and

165 'jellyfish' (a floating head with several plastic arms) for twenty minutes. This form of enrichment was

already in use in the park. Therefore, dolphins were familiar with it and the condition was included as a

167 complex/varied, but non-novel form of enrichment, to assess whether effects of CM were due to novelty.

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169 2.4 Experimental design

Provision of enrichments and observation of the dolphins' behaviour occurred on 56 non-consecutive days.
On any given day of study, only one of the two groups of dolphins was observed. Observations were
performed in a one-hour interval per day, from 09:00 to 10:00 A.M. The hour interval was divided in a preenrichment (20 minutes), an enrichment (20 minutes) and a post-enrichment (20 minutes) phase. In the
enrichment phase, the group under observation was exposed to only one of the different types of enrichment
described above. The order with which enrichments were provided was randomized and homogeneously
distributed across the study period.

Each group was subjected to each of the experimental conditions on seven days, for a total of 28 days ofobservations for each group and 56 observation days in total.

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180 2.5 Behavioural monitoring system and analysis

181 Two camcorders were used to record the behaviour of animals inside the experimental pool during the study.

182 One (HDR-AS50, Sony, Minato, Tokyo, Japan) was positioned behind the rectangular window, allowing an

underwater view of most of the pool (about 3% of the pool's volume was not within the camera's field of 183 view). The second (HERO4, GoPro, San Mateo, CA, USA) was positioned above the experimental pool, 184 185 allowing the recording of behaviours above the water surface (e.g. jump, breathe, etc.). Behavioural data collection from videos was performed with a focal animal, continuous sampling method using the Observer 186 XT software (Ver.12.5, Noldus, Gröeningen, The Netherlands), according to the ethogram reported in Table 187 s2, which was specifically realized for this experiment by taking several published studies as sources of 188 189 behaviours and definitions (Galhardo et al., 1996; Marten et al., 1996; Samuels and Gifford, 1997; Müller et 190 al., 1998; Connor et al., 2000; Miles and Herzing, 2003; Sekiguchi and Kohshima, 2003; Connor et al., 191 2006a; Holobinko and Waring, 2010; Jensen et al., 2013; Clegg et al., 2015, 2017a, 2017c; Dudzinski and Ribic, 2017; Hanna et al., 2017; Serres and Delfour, 2017; Clegg and Delfour, 2018b; Winship and 192 193 Eskelinen, 2018; Moreno and Macgregor, 2019; Serres et al., 2020, 2021). Collected data was used to 194 calculate relative duration of expression (% of observation time) for behavioural states and the frequency (events/hr) for behavioural events. Separate variables were obtained for the three phases (pre-enrichment, 195 enrichment, post-enrichment), subtracting the time the dolphins were not in view from the total observation 196 197 time. Inter-observer reliability was assessed on data collected by a second operator on 25% of videos and 198 was revealed to be good (intra-class correlation coefficient > 0.75 for all behaviours).

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200 2.5 Statistical analysis

201 Statistical analysis was aimed at determining the effects of the provision of the CM on the dolphin's 202 behaviour. A preliminary data analysis showed that the provision of FO, for its very nature, determined an 203 almost exclusive expression of interaction with the objects, which resulted in the concurrent lack of 204 expression of most other behaviours other than interest in enrichment. This would have resulted in a 205 significant effect of the provision of FO on all behavioural variables, impacting on our possibility to observe 206 subtler effects of the other types of enrichments (for which no interaction with the enrichment was possible, 207 thus not directly competing with other behaviours). Therefore, we performed two types of analysis: one 208 assessing variations in behaviour between the pre-enrichment phase and the enrichment phase, which was limited to data obtained in the CM, RS and PH conditions. The other analysis assessing variations in 209 behaviour between the pre-enrichment and the post-enrichment phases, in which data from all four 210

enrichment conditions were used. Only behavioural states expressed for $\ge 1\%$ of observation time and events expressed with a frequency ≥ 5 events/hr were included.

213 In both cases, a Generalized Estimating Equations model was used. The dependent variable was the duration or the frequency of behaviour for states and events, respectively. The model included the type of enrichment, 214 the phase and their interaction as fixed factors. The model included also the dolphins name as a random term, 215 to account for the repeated measurement from the same subjects. Post-hoc comparisons were performed 216 217 when a significant effect of the enrichment, the phase or of the phase*enrichment interaction were found. In 218 the latter case, comparisons were performed between phases, within enrichment type and sequential 219 Bonferroni corrections were applied to such comparisons. The software SPSS (ver. 27, IBM, Armonk, NY, 220 USA) was used for all statistical analysis. The level of statistical significance for tests was set at 0.01. Data 221 are reported as mean±SD for descriptive analysis or as estimated mean±SE for inferential statistics. 222 223 2.6 Ethical approval The study was conducted in accordance with relevant legislation for research involving animals, and 224 225 according to the type of procedure used, no formal ethical approval was required. 226 3. Results 227 228 229 3.1 Descriptive analysis 230 Descriptive parameters of behaviours expressed by dolphins on study days is reported in Table s3. Dolphins 231 were almost always visible. Chasing/escaping and lateral swim were expressed for less than 1% of the time 232 and agonistic behaviour, spy-hop, nodding and production of air bubbles were expressed with a lower frequency than 5 events/hr; these behavioural variables were therefore not included in the analysis assessing 233

- the effect of the enrichment. Interest in the enrichment was expressed for 75.1±24.1% of time (enrichment
- 235 phase) in FO, for 8.8±8.2 in CM, for 8.6±7.6 in PH, and for 5.1±4.4 in RS.
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- 237 *3.2 Effect of the presence of the enrichment*

Results of the GEE model indicating the effect of the phase and type of enrichment on behaviours expressedby dolphins before and during the presence of the enrichment are reported in Table 1.

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The frequency of breathing was affected by the mere presence of enrichment, regardless of its nature, increasing from 103.3±6.3 to 108.7±7.1 breathing/hr from pre- to the enrichment phase. Several other behaviours were significantly affected by the interaction between phase and enrichment. Specifically, the presence of CM determined an increase in the relative duration of synchronous swimming (Fig. 1A) and interest in dolphin (Fig. 1B) and in the frequency of touch (Fig. 1C); no such differences were found when RS or PH were provided.

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248 *3.3 Post-exposure effects of enrichment*

Table 2 reports results of the GEE model indicating the effect of the phase and type of enrichment onbehaviours expressed by dolphins before and after being exposed to the enrichment.

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252 The mere exposure to enrichment determined an increase in the relative duration of forward swim 253 $(47.2\pm2.9\%$ to $52.5\pm3.2\%)$, synchronous swim $(9.2\pm2.2\%$ to $16.4\pm3.6\%)$ and in the frequency of breathing 254 $(105.5\pm6.1 \text{ to } 117.4\pm5.1 \text{ breathing/hr})$, regardless of the type of enrichment. Breathing was also more 255 frequent on days when dolphins were exposed to FO (118.9±5.4 breathing/hr) than when exposed to CM 256 (109.7±7.1 breathing/hr), RS (107.6±5.4 breathing/hr) or PH (109.6±5.7 breathing/hr). The duration of 257 synchronous swim was also affected by the type of enrichment, being more expressed on days in which 258 dolphins were exposed to FO (16.1 \pm 3.5%) than when exposed to CM (12.5 \pm 2.9%; P < 0.001) or to PH 259 $(9.1\pm2.0\%; P < 0.001)$; expression on days when exposed to RS $(13.5\pm3.7\%)$ was not different from any of 260 the other enrichments. A significant effect of the phase*enrichment interaction was found for touches, with 261 their frequency increasing from pre- to post-exposure to CM, but not to other enrichments (Fig. 2A). Another significant effect of the phase*enrichment interaction was found from pre- to post- exposure to PH, but not 262 to other enrichments, with a decrease in the duration of circular swimming (Fig. 2B). Although a significant 263 effect of the phase*enrichment interaction was also found for rest, post-hoc analysis revealed no significant 264 effect once corrections for multiple comparisons were applied. 265

266 4. Discussion

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Compared to the baseline period (*i.e.*, before the provision of the enrichment), the administration of classical music resulted in unspecific effects on dolphins' behaviours observed while all enrichment were present and during the 20 minutes after they were removed. Interestingly, an increase of social behaviours was an effect noticed only during and after the provision of classical music.

272 As about the unspecific effects of enrichments, breathing increased when any kind of non-object enrichment 273 was present. In dolphins, as it occurs in many animal species, breathing rate is modulated in response to stress 274 (Clegg and Delfour, 2018a), social behaviors (Serres and Delfour, 2019), activity level (Sekiguchi and 275 Kohshima, 2003) and environmental parameters (Serres et al., 2021). Hence, breathing rate cannot be directly 276 linked to the valence of an emotional status, but most probably to the arousal's intensity (Clegg and Delfour, 277 2018a). The same unspecific effect lasted also after the removal of the enrichments, when an increase of 278 forward and synchronous swimming compared to baseline was also present. This means that regardless of the 279 nature, complexity or novelty of the type of enrichment, dolphins performed more social behaviours and 280 increased their general activity when presented with such stimuli. In line with our results, Clegg and co-authors 281 (2017c) noticed that in bottlenose dolphins synchronous swimming peaked shortly after training and fun sessions and Miller and collaborators (2011) found that general affiliative behaviour and activity levels 282 283 increased after shows. The fact that enrichments have the power to increase activity level and social behaviours 284 is not surprising given their very nature but, to the best of our knowledge, the fact that this effect is maintained 285 in the short term is poorly documented. Moreover, the ability of classical music to evoke unspecific effects, 286 equal to those of other enrichments widely used in bottlenose dolphins, supports its effectiveness as an 287 enrichment.

An increase of the relative duration of synchronous swimming, interest in dolphin and the frequency of gentle touches was only observed during the provision of classical music and this increase was maintained in the following 20 minutes for synchronous swimming and touches. In dolphins, increased expression of these affiliative behaviors may represent either a form of social support, thus helping animals to cope with stressors (Waples and Gales, 2002), or an expression of positive emotions (Clegg et al., 2017a, 2017b). The social support function is supported by the fact that affiliative behaviours appear in different stressful events in the 294 dolphin society (Waples and Gales, 2002; Connor et al., 2006a; Serres et al., 2020, 2021). However, this 295 explanation seems unlikely in our case, as no increases in aggressive behaviors or decreases in positive state-296 related behaviours were expressed when classical music was provided, as it is observed under conditions of 297 acoustical stress (Couquiaud, 2005; Serres and Delfour, 2017) or when a novel stimulus is presented (Winship 298 and Eskelinen, 2018). It is more likely that the increased social behaviours observed in this study represent an 299 expression of positive emotions. Connor and co-authors (2006b) described wild bottlenose dolphins' affiliative 300 behaviour and found that synchronous surfacing in males is associated with petting behaviour. Moreover, 301 gentle tactile interactions, analogous to grooming behaviour in terrestrial mammals (Kuczaj et al., 2013), and 302 synchronous swimming (thought to reflect social bonds; Connor et al., 2006a) are some of the more direct 303 indicators of social affiliation (Holobinko and Waring, 2010; Kuczaj et al., 2013). Even though the current 304 knowledge about dolphins' emotions is extremely limited, affiliative behaviours are correlated with positive 305 affective state in a wide range of mammals (Boissy et al., 2007) and this might be the case in bottlenose 306 dolphins as hypothesized in previous studies on dolphins' enrichment (Miller et al., 2011; Clark et al., 2013; 307 Clegg et al., 2017c; Serres et al., 2020). Classical music was the only condition that resulted in an increase in 308 the frequency of gentle touches between dolphins also after its removal. Specific acoustical stimuli may indeed 309 trigger changes in behaviours after their presentation; for instance, a species-specific music increased calming 310 and foraging behaviours in tamarins (Snowdon and Teie, 2010), while in chimpanzees instrumental music increased affiliative behaviours in both males and females, "easy-listening" music decreased aggressive 311 312 behaviours in males (Videan et al., 2007) and a mix of music (classical music among others) decreased 313 agitated/aggressive behaviours and increased social inactive/relaxed behaviours (Howell et al., 2003). The 314 short-term effects previously described on other mammals are in line with the social behaviours observed in 315 the current study, and, as previously described, the expression of affiliative behaviours may be linked to 316 positive affective states (Boissy et al., 2007).

The question is why only classical music stimulated these social interactions while rain sound (anotheracoustical enrichment) or photographs of natural landscapes (another novel enrichment) did not?

As for rain sound, the reasons of its (lack of) effect must be sought for in the characteristics of the sound. Music can indeed trigger different kind of behavioral responses in both human and non-human animals (for a review, see Snowdon, 2021) and acoustic features of auditory stimuli can trigger specific emotional states in 322 humans (Juslin and Laukka, 2003; Bresin and Friberg, 2011). However, it is still unclear what specific traits 323 of music are responsible for the variety of responses toward music observed in animals. One possible 324 explanation of the different effect of classical music and rain observed in the present study, is that only music 325 activates the endogenous opioids system, a central component in social motivation and pleasure perception 326 (Tarr et al., 2014) that is related to social bonding in many mammals and birds. Moreover, dolphins may perceive classical music as consonancy and rhythmicity. Consonant music is hypothesized to be "therapeutic" 327 328 for most uses with animals (Snowdon, 2021), and human infants (Trainor and Heinmiller, 1998), infant 329 chimpanzee (Sugimoto et al., 2010) and birds (Chiandetti and Vallortigara, 2011) preferred consonant 330 compared to dissonant music. Furthermore, in humans, synchronization of movement to a beat increases trusts and cooperation (Tarr et al., 2014) and is associated with positive emotions (Trost et al., 2017). Musicality 331 increases the number of simple relationship and the quality of existing relationships, underlying a coevolved 332 333 system for social bonding. The capacity to perceive and move to a beat is a core component of musicality, rare 334 among vertebrates (Savage et al., 2021). Interestingly, the capacity to adjust to and synchronize behaviour to the beat appears to be limited to vocal learning species, which also includes dolphins. 335

336 Vocalizations in dolphins are commonly expressed during social affiliative behaviour (Mello and Amundin, 337 2005; Janik, 2013; De Moura Lima, 2017) and detection of conspecifics sounds could trigger behavioral responses in seals, whales and dolphins' species (Deecke, 2006; Barluet de Bauchesne et al., 2021). In this 338 scenario, the possibility that acoustical cues from music could induce affiliative behaviour has to be considered, 339 340 as this sensory channel is used in association with context-specific social behaviours. In addition, the more 341 complex pattern of classical music may be closer to the variety in the characteristics of dolphins' vocalizations, 342 compared to the repetitive pattern of the rain sound. However, albeit not significantly, the rain sound audio 343 file tended to increase touches after the end of the presentation of the stimuli (P=0.02; Fig. 2A), while 344 photographs and floating objects were far to reach statistical significance (P=0.74, P=0.64 respectively). 345 Therefore, whether this is due to a generic effect of auditory stimuli leading to an afterward increase in social 346 behaviours, or to some specific characteristics of (classical) music, remains an open question.

The lack of effect on social behaviours when the photographs of natural landscapes were provided is likely due to the content and characteristics of our visual stimulus. First, the relative slowness by which images changed on the monitor and the fact that only landscape was represented may have been too less stimulating to induce an increase in activity while presented. Second, the absence of conspecifics or other moving animals represents a context that is not proper for the expression of social behaviors (Platt and Novak, 1997; Bloomsmith and Lambeth, 2000; Hanna et al., 2017; Winship and Eskelinen, 2018). Regardless of that, it should be highlighted that our visual and musical enrichment were both novel to the dolphins which underlines how the effects on social behavior induced by classical music in this phase cannot be explained by novelty.

355 Although beyond the aim of this study, it should be noted that photographs presented on a monitor were the 356 only enrichment significantly decreasing circular swimming after the end of their provision. To note, circular 357 swimming could be categorized as stereotypical behaviour (Mason, 1991) even if no clear link was found 358 between circular swimming and good or poor welfare (Mason and Latham, 2004). The fact that only 359 photographs have been able to reduce this unwanted behaviour is difficult to explain, but it is possible that this 360 is due to the limited expression of this behaviour and the small size of our sample. In fact, circular swimming tended to decrease also after classical music enrichment albeit in a not significant way (P=0.06; Fig. 2B). If 361 362 this will be confirmed, being a novel enrichment, a feature shared by classical music and video, could be the reason of this effect. 363

364 An unexpected result was the observation that, dolphins' swimming and breathing behaviors were more 365 expressed in days when floating objects were used as enrichment than in days when other enrichments were used, regardless of the experimental phase. Since this enrichment was the only one already known by the 366 367 animals, it is likely that dolphins anticipated the provision of floating objects. In fact, we cannot exclude that 368 the employees, despite having been instructed not to do so, might have unintentionally carried out activities 369 that were associated with the provision of the known enrichment. Anticipatory behaviours analogous to the 370 behaviors that would have followed have been already observed in dolphins before shows (Jensen et al., 2013) 371 and before different human-controlled situations (Clegg et al., 2018). Accordingly, anticipatory behaviours 372 expressed by our dolphins (*i.e.*, synchronous swimming and breathing) were increased after floating objects 373 provision.

- 374
- 375 5. Conclusion
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The present experiment identified classical music as an effective form of enrichment for bottlenose dolphins. 377 To the best of our knowledge, this is the first study looking at auditory enrichment for this species and for 378 379 dolphins under human care in general. Generalization of the results would require the replication of similar experiments in other dolphinaria and possibly in other dolphin species. Nonetheless, the results provide 380 several relevant inputs, for both application of music as an enrichment and for further research. The specific 381 382 effects of music on the enhancement of affiliative behaviour suggests that this enrichment could be 383 particularly useful in situations in which positive social interactions must be facilitated, including for 384 instance modifications in the social composition of the group, or exposure to stressors or other situations that may lead to increased conflict. An aspect that certainly warrants further investigations is the mechanisms by 385 which classical music enhances social behavior. Studies in this sense should determine if the effect is 386 specific to classical music or it is shared by other musical genres, and what specific features of music are 387 responsible for the effect. In relation to the latter aspect, an intriguing hypothesis that should be explored is 388 389 that behaviour could be synchronized to musical rhythm in dolphins, as a characteristic shared only by vocal 390 learning species. Such investigation could be also extended to other highly social, non-vocal-learning 391 species, such as dogs or primates, to highlight phylogenetic aspects whereby musical features of auditory 392 stimuli became linked to the expression of social behavior. 393 6. Conflicts of interest 394

- 395
- 396 The authors report no declarations of interest.
- 397

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399

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9. Tables

Table 1. Results of the Generalized Estimating Equations, indicating the effect of the phase, the type of
enrichment and their interaction on behaviours expressed by dolphins before and during the presence of the
enrichment. The analysis did not include data collected when the floating objects was provided as

611 enrichment.

Behaviour	Behaviour	Phase	Enrichment	Phase*Enrichment
type				
States	Forward swim	$X^2 = 1,88; P = 0.170$	$X^2 = 0.77; P = 0.680$	$X^2 = 5.57; P = 0.062$
(% of time)	Inverted swim	$X^2 = 2.02; P = 0.155$	$X^2 = 5.97; P = 0.050$	$X^2 = 2.61; P = 0.271$
	Synchronous swim	$X^2 = 12.60; P < 0.001$	$X^2 = 10.16; P = 0.006$	$X^2 = 30.86; P < 0.001$
	Circular swim	$X^2 = 4.87; P = 0.027$	$X^2 = 1.44; P = 0.486$	$X^2 = 0.41; P = 0.813$
	Interest in dolphin	$X^2 = 0.81; P = 0.366$	$X^2 = 7.51; P = 0.023$	$X^2 = 17.05; P < 0.001$
	Rest	$X^2 = 3.27; P = 0.070$	$X^2 = 7.58; P = 0.023$	$X^2 = 2.33; P = 0.311$
Events	Breathing	$X^2 = 10.80; P = 0.001$	$X^2 = 0.13; P = 0.936$	$X^2 = 0.16; P = 0.920$
(events/hr)	Touch	$X^2 = 2.62; P = 0.105$	$X^2 = 18.03; P < 0.001$	$X^2 = 17.01; P < 0.001$

Behaviour	Behaviour	Phase	Enrichment	Phase*Enrichment
type				
States	Forward swim	$X^2 = 10.00; P = 0.002$	$X^2 = 3.09; P = 0.377$	$X^2 = 6.73; P = 0.081$
(% of time)	Inverted swim	$X^2 = 0.09; P = 0.762$	$X^2 = 8.98; P = 0.029$	$X^2 = 4.51; P = 0.211$
	Synchronous swim	$X^2 = 17.5; P < 0.001$	$X^2 = 12.55; P = 0.006$	$X^2 = 8.62; P = 0.035$
	Circular swim	$X^2 = 1.24; P = 0.265$	$X^2 = 19.45; P < 0.001$	$X^2 = 16.26; P = 0.001$
	Interest in dolphin	$X^2 = 1.59; P = 0.207$	$X^2 = 3.60; P = 0.308$	$X^2 = 6.57; P = 0.087$
	Rest	$X^2 = 2.98; P = 0.084$	$X^2 = 72.61; P < 0.001$	$X^2 = 14.10; P = 0.003$
Events	Breathing	$X^2 = 40.75; P < 0.001$	<i>X</i> ² = 13.86; P = 0.003	$X^2 = 3.00; P = 0.391$

 $X^2 = 5.81; P = 0.016$

 $X^2 = 4.03; P = 0.258$ $X^2 = 23.59; P < 0.001$

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Table 2. Results of the Generalized Estimating Equations, indicating the effect of the phase, the type of

615 enrichment and their interaction on behaviours expressed by dolphins before and after the presence of the

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Touch

(events/hr)

10. Figure captions

Figure 1. Estimated means±SE of the relative duration of synchronous swim (A), interest in another dolphin (B) and touches of another dolphin (C) expressed by dolphins before and during the exposure to classical music (CM), rain sound (RS) and photographs of natural landscapes (PH). Generalized Estimating Equations. Figure 2. Estimated means±SE of the frequency of touches of another dolphin (A) and of the duration of circular swim (B) expressed by dolphins before and after the exposure to classical music (CM), rain sound (RS), photographs of natural landscapes (PH) and floating objects (FO). Generalized Estimating Equations.