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Kaloko-Honokōhau National Historical Park**

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Sunscreen pollution is abated during the COVID-19 “Anthropause” of 2020 in two U.S. National Parks: Cape Lookout National Seashore and Kaloko-Honokōhau National Historical Park

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1 **Abstract**

2

3 From March to June 2020, governments across the world imposed lockdowns in an attempt to
4 reduce the transmission of COVID-19 during the early phase of the pandemic. This period of
5 time in which human activity slowed worldwide has been coined the “Anthropause”. The goal of
6 this study was to determine if sunscreen pollution abated during the Anthropause and to identify
7 the severity of the pollution when tourism/recreation recovered at two coastal units of the U.S.
8 National Park System: Kaloko-Honokōhau National Historical Park in Hawai‘i, U.S.A. and Cape
9 Lookout National Seashore in North Carolina, U.S.A. Active ingredients of sunscreen products
10 were measured in water and sand samples at both locations, including oxybenzone, octinoxate,
11 octocrylene, octisalate, homosalate, and relevant breakdown products of some of these
12 ingredients. A risk assessment was conducted on Anthropause and post-Anthropause
13 contaminant levels for both locations to determine if there was a threat reduction during the
14 Anthropause, and whether tourism recovery in the post-Anthropause period served as a threat to
15 coastal wildlife. Both national park units exhibited an almost absolute reduction in the levels of
16 sunscreen contamination during the Anthropause period, a striking commonality considering the
17 geographic expanse separating the parks. Once travel restrictions were lifted, a large influx of
18 tourists ensued at both locations, resulting in a relatively sudden and dangerous increase in the
19 levels of sunscreen chemical pollution. This study supports the argument that unmanaged
20 tourism is a source of coastal sunscreen pollution that poses a threat to the localized continuity of
21 species populations and biodiversity, especially to coral reefs and fisheries.

22

23 **Keywords:** oxybenzone, National Park, sunscreen pollution, risk assessment, Anthropause,
24 COVID-19

25

26 **1. Introduction**

27
28 From March to June 2020, governments across the world imposed lockdowns in an attempt to
29 reduce the transmission of COVID-19 during the early phase of the pandemic. This period of
30 time in which human activity slowed worldwide has been coined, the “Anthropause” (Rutz et al.,
31 2020). The effect of the Anthropause was apparent in tourist hotspots (Zielinski and Botero,
32 2020; Ormazza-González et al., 2021; Kough et al., 2022; Moustafa and Mansour, 2022). As
33 travel ground to a halt, tourist destinations were among those to record the largest declines in
34 human activity with concomitant improvements in local environmental conditions (Dutta et al.,
35 2020; Manenti et al., 2020; Oarani, 2020; Thomson et al., 2020; Zambrano-Monserrate et al.,
36 2020; Callejas et al., 2021; Soto et al., 2021; Bertucci et al., 2023). Thus, the “pause” serves as
37 an opportune natural experiment for testing the relationship between tourism and environmental
38 degradation and assessing the effectiveness of visitation policies in reducing contamination (Rutz
39 et al., 2020; Gaiser et al., 2022).

40
41 Over the past 60 years, the exponential increase in tourism has spurred environmental
42 degradation and loss of biodiversity, with coral reefs among the most globally impacted
43 ecosystems (Gardner et al., 2003; Gil et al., 2015; Partelow et al., 2015; Downs et al., 2022a; Lin
44 et al., 2023). Sunscreen pollution is one of the symptoms of unsustainable tourism and coastal
45 development (Sánchez-Quiles and Tovar-Sánchez, 2014; Tovar-Sánchez et al., 2019; Casas-
46 Beltrán et al., 2020; Casas-Beltrán et al., 2021; Downs et al., 2022b, 2022c). Tourism-associated
47 sunscreen pollution contaminates the entire range of marine habitats, from Arctic and Antarctic
48 coastlines and Norwegian Fjords to temperate beaches, as well as coral reefs (Langford and
49 Thomas, 2008; Bratkovics et al., 2015; Downs et al., 2016; Dominguez-Morueco et al., 2020;
50 Garcia-Pimentel et al., 2023). Tourism and water-recreational activities also contaminate
51 limnological systems with sunscreen pollutants, with major sources that include activities such as
52 river tubing, paddle boarding, kayaking/canoeing, and other water sports (Balmer et al., 2005;
53 Cuderman and Heath, 2007; Mandaric et al., 2017; Dominguez-Morueco et al., 2020; Rand et al.,
54 2020; Newham et al., 2021; Harjung et al., 2022; Homem et al., 2023). As a consequence of
55 more than a decade of research on its impacts, tourism-associated sunscreen pollution is no
56 longer an “emerging concern” but a managed pollutant that is being mitigated through legislative
57 rules or executive policies that ban or restrict the presence of offending sunscreen chemicals
58 (Downs et al., 2022a). What remains unclear is how much of a driver tourism versus other
59 potential sources is on sunscreen pollution, regardless of policy status.

60
61 Sunscreen chemicals reach levels in some aquatic environments that they pose a localized
62 threat to biodiversity, especially in conjunction with other stressors, such as climate change
63 factors, microplastic pollution, and other kinds of pollutants (Alonso et al., 2015; Pont et al.,
64 2004; Le Bihanic et al., 2020; Wijgerde et al., 2020; Zhou et al., 2023). In fish, oxybenzone,
65 octocrylene, octisalate and other sunscreen chemicals induce a range of acute toxicities, from
66 genotoxicity and developmental deformities to impacting reproductive fitness over multiple
67 generations (Kim et al., 2014; Almeida et al., 2019; Yan et al., 2020; Zhou et al., 2022; Zhao et
68 al., 2023). Fish seem exceedingly susceptible to the endocrine-disrupting nature of many of these
69 sunscreen chemicals, with impacts such as skeletal deformations, feminization and sex change of
70 males to females, thyroid-axis pathologies, and adverse alterations to critical social behaviors
71 (Ziarrusta et al., 2018; Simons et al., 2022; Xie et al., 2022; Tao et al., 2023). For aquatic
72 invertebrates, these chemicals elicit similar multi-generational toxicities as seen in vertebrates,
73 but are also documented to interfere with aragonite or shell deposition in species ranging from

74 corals to bivalves (Downs et al., 2016; Coinaldesi et al., 2017; Campos et al., 2019; Lopes et al.,
75 2020; Santonocito et al., 2020; Pham et al., 2022). In all animal taxa, the benzophenones from
76 sunscreens, such as oxybenzone and benzophenone, are noted reproductive and developmental
77 toxicants, and have carcinogenic potential (DiNardo and Downs, 2019; Tao et al., 2020; Downs
78 et al., 2021; Zhang et al., 2021; Ma et al., 2023). Moreover, aquatic algae and plants are
79 extremely susceptible to various sunscreen chemicals, exhibiting growth-inhibition toxicities as
80 low as parts per trillions (Mao et al., 2017; Zhong et al., 2019; Xing et al., 2022; Yang et al.,
81 2023).

82
83 Ecotoxicological and risk assessment research on terrestrial species and habitats has lagged
84 far behind studies of aquatic systems, but has recently gained prominence because of the impact
85 sunscreen chemicals have had on hydroponics and irrigated agriculture (Downs et al., 2022b;
86 Gautam et al., 2022). Independent groups in China, Mexico, Portugal, and Spain noticed a
87 significant reduction in crop yields when R1 recycled-waters from municipal waste-water
88 treatment facilities (WWTF) were used for irrigation (Sabourin et al., 2009; Chen et al., 2017;
89 Cabrera-Peralta and Peña-Alvarez, 2018; Zhong et al., 2020; Ramos et al., 2021). Experimental
90 and reductive investigations established that one of the main categories of toxicants to plants
91 were sunscreen chemicals, especially oxybenzone, avobenzone, and octocrylene (Chen et al.,
92 2017; Zhong et al., 2020). This finding is consistent with the historical usage of these chemicals
93 (Downs et al., 2021). These findings led to research regarding sunscreen-chemical impacts to
94 agricultural outcomes when biosolids from a WWTF were used as soil amendments (Wang and
95 Kannan, 2017; Gautam et al., 2022; Li et al., 2022). New research shows that sunscreen chemical
96 contamination and toxicities extend to migratory birds, sea turtles, and even amphibians (Fin et
97 al., 2017; Molins-Delgado et al., 2017; Li et al., 2022; Li et al., 2023). Concern over sunscreen
98 pollution has become so great that even indirect routes of contamination, including atmospheric
99 deposition from saerosolized sunscreen volatile organic carbons (VOCs), surf aerosolization, and
100 various means of discharge from WWTF are now identified as critical targets for mitigation
101 efforts (Teplitz et al., 2018; Pegoraro et al., 2020; Bayati et al., 2021; Franklin et al., 2021;
102 Pendergraft et al., 2021; Franklin et al., 2022).

103
104 Several environmental monitoring studies have suggested the value of using the Anthropause
105 to establish baseline data for determining the impact of tourism to wildlife receptors and natural
106 resources (Coll, 2020; Chakraborty et al., 2021). Research on the Anthropause has been a crucial
107 factor in identifying the lack of effectiveness of current conservation management policies
108 regarding tourism/recreational activities and intensity, allowing for resource management to
109 adjust policies to increase effectiveness, and providing a measure for policy effectiveness (Bates
110 et al., 2020; Callejas et al., 2021; Ben-Haddad et al., 2022). For example, using the Anthropause
111 as a baseline, it was shown that tourism-associated chemical pollution was responsible for the
112 high-levels of genotoxicity seen in near-shore populations of sergeant-major reef fish (de Lima e
113 Silva et al., 2022). Schofield and co-workers also used the Anthropause to demonstrate that
114 tourism served as a major disturbance to sea turtle nesting (critical or essential habitat) and
115 recommended that a refuge zone be established to facilitate increased turtle aggregation in this
116 critical habitat (Schofield et al., 2021). In other studies, fish densities in ecologically important
117 habitat structures improved significantly during the Anthropause, but subsequently declined as
118 tourism recovered (Bertucci et al., 2023). Ghost crab populations on tourist beaches were
119 recognized to be severely reduced because of the Anthropause differential, concluding that "...
120 urban beaches should not be exclusively managed for recreational purposes." (Costa et al.,
121 2022).

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The goal of this study was to determine if sunscreen pollution abated during the Anthropause and to identify the severity of the pollution when tourism/recreation recovered at two coastal units of the U.S. National Park System: Kaloko-Honokōhau National Historical Park in Hawai‘i, U.S.A. and Cape Lookout National Seashore in North Carolina, U.S.A. These two sites were selected because the U.S. National Park Service had concerns about the implications of mass tourism and the impact to two of the major coastal park units. The Hawaii site represented a Pacific coast location while the North Carolina site represented an Atlantic location. Active ingredients of sunscreen products were measured in water and sand samples at both locations, including oxybenzone, octinoxate, octocrylene, octisalate, homosalate, and relevant breakdown products of some of these ingredients. A risk assessment was conducted on Anthropause and post-Anthropause contaminant levels for both locations to determine if there was a threat reduction during the Anthropause, and whether tourism recovery in the post-Anthropause period served as a threat to coastal wildlife. To this end, shoreline sand samples were collected at both National Park units in May 2020 and then again in August 2021. Our results contribute to the baseline information on the linkages between tourism/recreation and coastal natural resources, which can be used to facilitate the development of effective, evidence-based management policies to ensure conservation of biodiversity, as well as sustainable use by tourism (Zieleniski and Botero, 2020; Armstrong et al., 2022; Procko et al., 2022).

143 **2. Materials and Methods**

144

145 *2.1 Sample Locations and Collections*

146

147 *2.1.1 Sample Locations*

148

149 Seawater and beach-sand samples were collected from two U.S. National Park units: Kaloko-
150 Honokōhau National Historical Park on the Island of Hawai‘i in the State of Hawai‘i (U.S.A.;
151 **Figures 1A and 1B**) and Cape Lookout National Seashore in the southern Outer Banks region of
152 the State of North Carolina (U.S.A.; **Figure 1C**). Samples were collected along shoreline of the
153 ‘Ai‘ōpio Fishtrap within Kaloko-Honokōhau National Historical Park (**Figure 1B**). For Cape
154 Lookout National Seashore, two separate locations were sampled (**Figure 1C**). One location is
155 the western end of the island of Shackleford Banks (**Figures 1C and 1D**) and the other is on the
156 island of South Core Banks that is adjacent to the Cape Lookout Lighthouse facility (**Figures 1C**
157 **and 1E**). Coordinates for all sampling sites are listed in **Supplemental Table 1** and, in most
158 cases, were determined using a Garmin WAAS-enabled GPS. Accuracy for all locations was
159 within three meters.

160

161 *2.1.2 Sampling time points.*

162

163 At Kaloko-Honokōhau National Historical Park, water samples were collected in April 18, 2018
164 (15:30-15:48 HST), November 11, 2019 (11:47 HST), May 8, 2020 (16:00-17:00 HST), and
165 August 18, 2021 (sand samples were collected from 09:06 to 09:24, and water samples were
166 collected from 14:10 to 14:21 HST). Water samples were collected during the outgoing tide
167 because concentrations are the highest at this time period (unpublished data).

168

169 At Cape Lookout National Seashore, sand samples from both the South Core Banks and
170 the Shackleford Banks sites were collected in May 14, 2020 (Figure 1C). Water and sand
171 samples were collected July 25, 2021, at the South Core Banks site. Water and sand samples
172 were collected July 24, 2021 at the Shackleford Banks site. Water samples were collected during
173 the outgoing high tide and sand samples were collected between 2-4 pm, when tourist density on
174 the beaches is greatest.

175

176 *2.1.3 Sampling Methods*

177

178 Spatulas for sand collection and sample vessels were precleaned following EPA Method 815-R-
179 05-004.

180

181 Water samples were collected using either precleaned one-liter amber glass bottles with
182 Teflon-lined lids (I-Chem, 300 series, VWR) or precleaned 5 mm PFE-bags with seals, at a depth
183 of 30 cm below the surface of the water. This depth reflects the measurement of the
184 contaminants in the water column versus what is found on the water surface.

185

186 Sand samples were collected by measuring a 10 x 10 cm area on the surface of the beach,
187 and a cleaned spatula was used to collect the top 0.5 cm of surface sand into a precleaned 100
188 mL amber jar with a Teflon-lined lid (Environment Express). Spatulas were only used once to
189 prevent cross-contamination.

190

191 After collection, both water and sand samples were placed in a cooler until transferred to
192 a freezer between -20°C to -30°C. Samples were shipped frozen to the laboratory and remained
193 in a -30°C freezer until processing.

194

195 *2.2 Analytical standard and chemicals*

196 The following analytical standards (> 97% purity) were used: 2-Hydroxy-4-
197 methoxybenzophenone (oxybenzone (BP3), dihydroxy benzophenone (benzophenone-1, BP1), 5-
198 benzoyl-4-hydroxy-2-methoxybenzene-1-sulfonic acid (benzophenone-4, BP4), 4-
199 hydroxybenzophenone (4HB), 4,4'-dihydroxy benzophenone (4DHB), 3-(4-tert-butylphenyl)-1-
200 (4-methoxyphenyl)propane-1,3-dione (avobenzene,AVO), drometrizole (UVP), dimethyl
201 benzotriazole (DMBZT), octocrylene (OC), 3,3,5-trimethylcyclohexyl 2-hydroxybenzoate
202 (homosalate, HMS), 2-ethylhexyl salicylate (octisalate,OS), and BP-13C were purchased from
203 Sigma-Aldrich (Darmstadt, Germany). Benzophenone-2 (BP-2), 2,2'-dihydroxy-4-
204 methoxybenzophenone (DHMB, benzophenone-8), ethyl p-aminobenzoic acid (benzocaine,
205 EtPABA), octinoxate (ethylhexyl methoxycinnamate), and 1H-benzotriazole (BZT) were
206 purchased from Merck (Darmstadt, Germany). 4-Methylbenzylidene camphor was obtained from
207 Dr. Ehrenstorfer (Augsburg, Germany). MeBZT was acquired from TCI (Zwijndrecht, Belgium).
208 Then isotopically labelled standards BP3-¹³C, BP3-d5, 4MBC-d4 and BZT-d4 were obtained
209 from CDN isotopes (Quebec, Canada).

210 Methanol (MeOH), acetonitrile (ACN), dichloromethane (DCM), ethyl acetate (EtAc),
211 formic acid and HPLC-grade water were purchased from J.T. Baker (Deventer, The Netherlands)
212 and were all ≥99.9% purity and included Certificates of Analysis. Aluminum oxide (≥99.9%
213 purity), used as a clean-up agent in the pressurized liquid extraction (PLE) cells, was obtained
214 from Merck (Darmstadt, Germany). Nitrogen and argon gasses (purchased from Air Liquid –
215 Barcelona, Spain) were of 99.999% purity. Glass fiber filters (1 µm) and nylon membranes (0.45
216 µm) from Whatman International Ltd. (Maidstone, UK) and syringe filters supplied by Dionex
217 Corporation (Sunnyvale, CA, USA) were used.

218

219 Stock solutions of the UV-filters and internal standards were prepared at 100 mg/L in
220 MeOH and stored in the dark at -20 °C. Separate working standard solutions with UV-filters and
221 internal standards were prepared at 10 mg/L in MeOH, stored in the dark at -20 °C, and refreshed
222 weekly.

223 *2.3 Sample pre-treatment and analytes extraction*

224 The optimized and validated analytical methods used for the multiresidue determination of the
225 target compounds and isotopically labelled standards in the standard solutions and in the sample
226 extracts, were based on previous procedures developed in our laboratories (Gago-Ferrero et al.,
227 2011; Downs et al., 2022b) and expanded for new compounds as described in Downs et al.,
228 2022c.

229 *2.3.1 Sand*

230

231 The extraction of the selected analytes from the sediment samples was carried out according to
232 the expanded method based on Rodrigues et al. (2021). Benzophenone (BP), Tinosorb M
233 (methylene bis-benzotriazolyl tetramethylbutylphenol; CAS# 103597-45-1), and oxybenzone
234 (CAS# 131-57-7) were purchased from Sigma-Aldrich (Lyon, France). Tinosorb S (bis-

235 ethylhexyloxyphenol methoxyphenyl triazine; CAS# 187393-00-6), avobenzone (butyl
236 methoxydibenzoylmethane; CAS# 70356-09-1), homosalate (CAS# 118-56-9), octisalate
237 (ethylhexyl salicylate; CAS# 118-60-5) and octocrylene (CAS# 6197-30-4) were kindly provided
238 by Pierre Fabre Laboratories. Butyloctyl salicylate (CAS# 190085-41-7) was obtained from
239 Innospec Active Chemicals. Octinoxate (ethylhexyl methoxycinnamate; CAS# 5466-77-3) was
240 obtained from Accustandard (Cat# ALR144N).

241 The standard additions method was used to measure the concentration of UV filters in
242 sand, as described previously (Rodrigues et al., 2021). Briefly, the extraction solutions were
243 MeOH containing 2000, 500, 200, 150, 100, 50, and 0 ng/mL of each UV filter. Exact
244 concentrations were calculated from the concentrations of each compound in the stock solution.
245 Extraction was performed on sand samples (seven samples, ~2 g of sand per each) each with a
246 MeOH (2 mL), and the concentration of UV filters in the supernatant was measured by direct
247 injection using an Ultra High Performance liquid chromatography-high resolution mass
248 spectrometer (UHPLC-HRMS) and calculation of the intersection of the regression line with the
249 x-axis. After analysis, the solvent and the interstitial water were removed by evaporation with a
250 GeneVac HT-4X. The exact mass of dry sand gave the initial mass of water in the sand and the
251 total volume of supernatant (2-mL MeOH + interstitial water), allowing for correction of the
252 concentrations in supernatant and in sand. The limits of quantification (LOQ) are provided in
253 **Supplemental Table 2**.

254 The recovery rates were calculated from spiked sand samples, which were extracted with
255 MeOH using the same protocol as for extraction of natural sand samples (Rodrigues et al., 2021).
256 The recovery rates are reported in **Supplemental Table 3**.

257 2.3.2 Water

258
259 The extraction of the 20 aromatic hydrocarbon ultraviolet filters (parent and transformation
260 products) from the water samples followed the expanded method based on Downs et al. (2021).

261 2.4 LC-MS analysis

262 2.4.1 HPLC- MS/MS analysis of water

263 The chromatographic separation and tandem-mass spectrometry detection for water analysis was
264 performed as described in Downs et al, (2022b and c).

265
266 Several methodological blanks were included (reagent blanks were run by processing
267 HPLC water in the same way as samples and procedural blank without matrix or sample). A
268 surrogate standard (BP-13C) as evaluation of the extraction performance and IS for the
269 quantification of the target analytes (**Supplemental Table 4**).

270
271 We use isotopically labeled standards to spike the samples. These isotopic compounds
272 are not used commercially and can be differentiated in both the chromatograms and with their
273 mass:charge ratios. We use these isotopic standards to determine the %recovery of our extraction
274 method – a measure of quantitation accuracy.

275 2.4.2 UHPLC-(ESI)-Orbitrap MS analysis of sand

276 The chromatographic separation of all analytes is described in Downs et al., 2022b and was
277 performed in an Acquity UHPLC C18 column (100 Å, 1.8 µm, 2.1 ×100 mm) with a guard
278 column containing the same material, using an Acquity ultra-high-performance liquid
279 chromatograph (UHPLC) (Waters Corporation, Milford, Ma, USA) coupled to a Q-Exactive
280 Orbitrap mass spectrometer (Thermo Scientific, Waltham, Ma, USA).

281
282 All samples were measured in duplicate and the reported value corresponds to the mean
283 of the two determinations. All the compounds were quantified and confirmed with the two most
284 intense transitions by the isotope dilution approach.

285
286 LODs, LOQs and recovery rates are listed elsewhere of analytes and the average recovery
287 rates are the same as found in (Downs et al, 2022b).

288 2.5. Risk Analysis

289 As described in previous papers (Downs et al., 2022b, 2022c), the European Commission
290 guidance was used to calculate risk quotients (RQs) for oxybenzone and octocrylene in both
291 water and sand samples (European Commission 2003; European Medicines Agency 2006;
292 Dussault et al., 2008; Hernando et al., 2006; Downs et al., 2021; Downs et al., 2022b).

293
294 It is praxis to use an AF of 1000 for acute toxicity data and 100 for chronic toxicity data
295 (Belanger et al., 2021). To calculate RQs, the following equation was used: $RQ =$
296 $(MEC)/(PNEC, NOEC, LC_{50} \text{ or } EC_{50}) \times 1000$. Toxicity reference values were obtained from the
297 published literature (**Tables 2-4, 5-7**) (Chapman et al., 2009; Dussault et al., 2008; Environment
298 Canada, 2013; Belanger et al., 2021; Jung et al., 2021).

299
300 The criteria for Levels of Concern for organisms in ecosystems for interpreting the RQs
301 is described in Downs et al., 2022b and 2022c. A color scheme is used for ease of visualization
302 of the Levels of Concern for this methodology (Downs et al., 2021a; **Tables 1 and 2,**
303 **Supplemental Table 5**). Red boxes represent RQ values greater than 1, indicating an
304 unacceptable risk requiring immediate action, and $RQ \geq 1$ is the standard criteria for the Level of
305 Concern within the European Commission framework. Orange boxes represent values between
306 0.5 and 1.0, a moderate concern for an acute impact. Yellow boxes represent values between 0.1
307 and 0.49, indicating a lower risk of impact. White boxes indicate no concern of danger with
308 values below 0.1.

309

310 **3. Results**

311 *3.1.1 Water Samples from Kaloko-Honokōhau National Historical Park.*

312

313 Sunscreen target analytes in water samples collected from 2018 to 2021 are listed in **Table 1A**.
314 Site W3 was sampled from 2018 to 2021. Nine out of the 17 target UVFs analyzed were
315 detected at least in one water sample (**Table 1A**). Oxybenzone levels ranged from 5,150 ng/L to
316 35,207 ng/L in 2018-2019, but were below the limit of quantification during the Anthropause.
317 Oxybenzone levels remained at this unquantifiable level in 2021. Avobenzone, octisalate,
318 homosalate and octocrylene at site W3 did not follow this behavior. In 2018-2019, these
319 sunscreen compounds were quantifiable in the µg/L concentrations, but were not quantifiable
320 during the Anthropause sampling period. All target compounds (except oxybenzone) were
321 detected several-fold higher in 2021 compared to 2018-2019 (**Table 1A**). During the
322 Anthropause sampling, only Site W5 had measurable quantities of a single sunscreen compound
323 – 85 ng/L oxybenzone.

324

325 *3.1.2. Sand Samples from Kaloko-Honokōhau National Historical Park.*

326

327 Only one sample from site S4 was collected (2018) before the Anthropause time-period. Site S4
328 is a popular area for beachgoers to dwell during their visit. In 2018, common sunscreen
329 compounds were detected in concentrations of parts per billion, while the breakdown product of
330 octocrylene, benzophenone, was detected at 60 ng/g (**Table 1B**). During the Anthropause
331 sampling period, all five beach sites had quantifiable levels of octocrylene (mean = 57 ng/g, Std
332 deviation = 36.7 ng/g), but no measurable concentration of the other six sunscreen compounds
333 (**Table 1B**). In 2021, oxybenzone was not detected at any of the beach sites. Octocrylene (mean
334 = 27,485 ng/g, Std deviation = 4,414 ng/g) and avobenzone (mean = 3,700 ng/g, Std Deviation =
335 5,666 ng/g; median = 1,333 ng/g) were measurable at all four 2021-beach sites. Homosalate
336 (mean = 22,102 ng/g, Std deviation = 21,029 ng/g; median = 18,860 ng/g) and octisalate (mean =
337 12,393 ng/g, Std deviation = 12,583; median = 9,814 ng/g) were measurable at sites S2-S4, but
338 were not detectable at site S1(**Table 1B**).

339

340 *3.1.3. Water samples from South Core Banks and Shackleford Banks, Cape Lookout National*
341 *Seashore area*

342

343 Water samples were not collected during the Anthropause period, but were collected during the
344 2021 sampling period. Sampling occurred in the proximity of a high density of swimmers for
345 Site CW1, but Site CW2 had only 22 swimmers south of the collection site. At the time of
346 sample collection, current flow was moving south to north. At the South Core Banks sites, Site
347 CW1 had extremely high levels of avobenzone, octisalate, octocrylene, and oxybenzone (**Table**
348 **2A**). At site CW1, it was observed that individuals were standing in the water and applying
349 aerosol sunscreen to 90% of their body, then quickly entering into the water.

350

351 The site SW1 sample at the Shackleford Banks area (**Figure 1D, Table 2A**) was
352 collected in the midst of ~80 people in the water in a small cove that contained a centralized
353 eddy. It was also observed that at least two individuals sprayed aerosol sunscreen onto their
354 bodies while standing in the water. Several individuals on watercraft anchored next to shore
355 were also spraying aerosol sunscreen to their bodies, and the spray was drifting onto the cove's
356 water-surface.

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3.1.4. Sand samples from South Core Banks and Shackleford Banks, Cape Lookout National Seashore area

Sand samples collected during the Anthropause period at the South Core Banks site (**Figures 1C, 1E**) have no quantifiable levels of target sunscreen chemicals (**Table 2B**). During this period, the commercial ferry services to the islands were not operating transport to either of the islands. During the sampling period of 2021, levels of avobenzone, homosalate, octisalate, octocrylene, and oxybenzone were all measurable in sand samples, though concentrations ranged by as much as 12-fold (**Table 2B**). Benzophenone was measurable at site C1 and site C4 (**Table 2B**).

Sand samples collected during the Anthropause period at the Shackleford Bank site (**Figures 1C, 1D**) had no quantifiable levels of target sunscreen chemicals with the exception of Site 4, where 10 ng/g of octinoxate was measured (**Table 2C**). During the 2021 sampling period, levels of six of the seven target sunscreens chemicals were measurable, the exception being octinoxate (**Table 2C**).

3.2. Risk Analysis

3.2.1. Risk quotients for water samples Kaloko-Honokōhau National Historical Park.

During the 2018-2019 pre-Anthropause period, RQs for oxybenzone concentrations in water samples exhibited a severe threat to the range of taxonomic receptors found at Kaloko-Honokōhau (**Table 3**). This result is in striking contrast to the RQs for all the water samples during the 2020 Anthropause period, except for site W5. For 2020 W5, the measurement of oxybenzone posed a severe threat to coral receptors, lower threat levels to arthropods and molluscs, and a moderate to severe threat to fish and photosynthetic taxa (**Table 3**). Risk quotients for oxybenzone in water samples in 2021 was “0” because oxybenzone was always below the limit of detection, i.e., not detected.

Octocrylene RQs in water samples from 2018-2021 showed a different trend than that of oxybenzone (**Table 3**); the RQs for the four major taxonomic groups exhibited a severe threat. During the Anthropause period, RQ values were “0” or not calculable because octocrylene was not detected. In 2021, with the return of crowds using oxybenzone-free sunscreens, octocrylene levels in W2 and W3 had $RQ > 1$, indicating a severe threat to the four taxonomic groups.

3.2.2. Risk quotients for sand samples at Kaloko-Honokōhau National Historical Park.

The single sand sample collected in 2018 had a $RQ > 1$ for oxybenzone, indicating a severe threat to the two terrestrial taxonomic groups (**Table 4**). Sand samples collected in 2020-2021 had an RQ of > 0 , indicating no threat from oxybenzone because oxybenzone was not detected in any of the water samples.

In contrast, the RQ values for octocrylene were “0” in all the sand samples collected from 2018-2021, except for site S5 (**Table 4**). Tourists are prohibited from lounging at site S5 by a park-specific policy of the U.S. National Park System.

3.2.3. Risk quotients for water samples at South Core Banks, Cape Lookout National Seashore

405
406 Oxybenzone risk quotients for water samples collected from South Core Banks (**Figure 1C-1E**)
407 during the 2021 sampling period had RQ values of more than 1 for both sampling locations,
408 with the exception was for site CW2, where the sea urchin and mussel where $RQ = 0.9$ (**Table 5**).
409

410 Octocrylene was not detected at site CW2 (**Figure 1E**) resulting in an RQ of 0. Sites
411 CW1 and SW1 had RQ values far exceeding “1” (**Table 5**).
412

413 3.2.4. Risk quotients for sand samples at South Core Banks, Cape Lookout National Seashore 414

415 During the Anthropause, neither oxybenzone nor octocrylene were detected at sites C1-C3
416 (**Figure 1 E, Table 1**), resulting in a $RQ = 0$ (**Table 6**). All sand samples for the sites collected
417 in 2021 had $RQ > 1$, indicating a severe threat to localized wildlife.
418

419 3.2.5. Risk quotients for sand samples at Shackleford Banks, Cape Lookout National Seashore 420 421

422 As with South Core Banks, none of the sand samples collected during the Anthropause contained
423 oxybenzone or octocrylene, resulting in a $RQ = 0$ (**Table 7**). It should be noted that Site S2 was
424 not collected during the 2020 sampling time point, but only during the 2021 sampling. For
425 oxybenzone, all five sites predominantly exhibited an $RQ > 1$, with the exception of sites S2 and
426 S5 for two of the parameters of *Eisenia fetida* (**Table 7**). Site S3 had an $RQ = 0$ for octocrylene;
427 this site was immediately in front of the latrines, and away from the beach zone where tourists
428 repose on the beach.
429

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432 4. Discussion

433
434 Both national park units exhibited an almost absolute reduction in the levels of sunscreen
435 contamination during the Anthropause period, a striking commonality considering the
436 geographic expanse separating the parks. Once travel restrictions were lifted, a large influx of
437 tourists ensued at both locations, resulting in a relatively sudden and increase in dangerous levels
438 of sunscreen chemical pollution. This study supports the argument that unmanaged tourism is a
439 source of coastal sunscreen pollution that poses a threat to the localized continuity of species
440 populations and biodiversity, especially to coral reefs and fisheries (Downs et al., 2022c; Pei et
441 al., 2023).

442
443 One major difference between the two sites was the absence of oxybenzone in the post-
444 Anthropause-period in Kaloko-Honokōhau National Historical Park. This absence is most likely
445 a direct result of the implementation of the State of Hawai‘i Act 104 (2018), which banned the
446 sale and distribution of sunscreen products containing oxybenzone or octinoxate and went into
447 effect on January 1, 2021 (Downs et al., 2022b). Octocrylene contamination during the
448 Anthropause at this location may have come from local residents who were able to access the
449 beach areas even when the park was officially closed (**Table 1B**; State of Hawai‘i, 2020). It
450 should be noted that at these two locations, the major proximate source for sunscreen chemical
451 contamination are swimmers, and that fluvial sewage discharges from homes or municipalities
452 are not present within the boundaries of the National Park units and the nearest municipal
453 discharge is over 7 km way with prevailing current going away from Shackelford Banks.

454
455 The high RQ values for Kaloko-Honokōhau NHP for water and beach areas indicate that
456 actions to mitigate sunscreen pollution are warranted. Besides the threat to the taxonomic
457 categories included in this risk analysis, the threat to other endangered species (including those
458 on the IUCN Red List and those protected under the U.S. Endangered Species Act) should be
459 considered. Sea turtles accumulate sunscreen compounds and exhibit associated pathologies
460 (Cocci et al., 2020; Cocci et al., 2022; Guevara-Melendez et al., 2023). Juvenile green sea turtles
461 (*Chelonia mydas*) frequently haul-out onshore to bask along the shoreline of the ‘Ai‘ōpio
462 Fishtrap at Kaloko-Honokōhau NHP. Because these chemicals are readily absorbed through the
463 skin, the concentrations of sunscreen contamination at this location may prove a serious threat
464 both in and out of the water. Food sources in this area (macroalgae and turf algae) could also be
465 contaminated with sunscreen chemicals and could be another route of exposure for these turtles
466 as has been found in macroalgae and sea grasses elsewhere (Cunha et al., 2015; Agawin et al.,
467 2022; Xing et al., 2022; García-Márquez et al., 2023).

468
469 Both national park units provide important sea turtle habitat. Kaloko-Honokōhau NHP is
470 a feeding and resting area for juvenile green turtles. The Cape Lookout National Seashore is a
471 critical nesting habitat for loggerhead, Kemp’s Ridley, green, and leatherback sea turtle species.
472 Sunscreen contamination can occur on sands immediately adjacent to sea turtle nesting sites at
473 Cape Lookout (**Supplemental Figure 1**), either via drift from surf-action or from aerosol spray
474 discharges, as well as from urination of tourists and their canine pets (C. Downs, personal
475 observation). We observed both signs of human urination (done out of ignorance with confirmed
476 admission by the perpetrators) and canines urinating on the sea turtle marker poles during our
477 field sampling. Canine urine can potentially be a source of contamination because of market
478 pressure to use canine sunscreen on pets, licking their owners' sunscreen-laden skin, and from
479 drinking from plastic dishes or on canine chew-toys which often contain oxybenzone as a UV

480 stabilizer (Asimakopoulos et al., 2016; Karthikraj et al., 2020; American Kennel Club, 2023).
481 Temperature is a major driver of sea turtle sex ratio (Mrosovsky and Provancha, 1989; Tanner et
482 al., 2019; Hays et al., 2023). For a broad range of vertebrates, oxybenzone can cause
483 feminization of juveniles, effect change in primary and secondary sex ratios, and alter embryonic
484 developmental fates (Bae et aal., 2016; Santamaria et al., 2020; Xu et al., 2021; Moreira et al.,
485 2022; Bai et al., 2023). In Florida, reports of sex ratios in sea turtle nests are 99 females:1 male,
486 even in areas/years where average temperatures during nesting incubations are not above the
487 critical threshold (U.S. NOAA, 2023b). Could sunscreen pollution be a contributing factor to
488 skewed sex ratios? Monitoring data collected for 2020-2021 nest hatching during the
489 Anthropause in Cape Lookout and other marine parks may provide some insight.

490
491 The 2014-2017 global coral bleaching event caused extensive mortality to the park's
492 coral reefs (McCutcheon and McKenna, 2021). The RQ analysis indicates that live coral
493 recovery may be affected by sunscreen contamination, particularly if bleaching events associated
494 with temperature increases become more frequent with the progression of global climate change.
495 Increase in sea surface temperatures can aggravate the toxicities of oxybenzone, avobenzone, and
496 octocrylene suggesting that the compound stressors may exacerbate localized population declines
497 (Wijgerde et al., 2020; Bordalo et al., 2023; He et al., 2023; Vuckovic et al., 2022; Morais et al.,
498 2023; He et al., 2023; Zhang et al., 2024). Mitigation of sunscreen pollution will be necessary for
499 coral reef recovery and conservation efforts (Downs et al., 2022a; Pastorino, 2024).

500
501 High RQ values for the Cape Lookout National Marine Seashores barrier islands indicate
502 a threat to both the marine and beach ecosystems. The botanical diversity of the beach dunes on
503 these islands provides critical ecosystem services for preventing coastal erosion. Oxybenzone,
504 octocrylene, avobenzone, and octinoxate are toxic to vascular plants (Zhong et al., 2020; Santo et
505 al., 2023; Li et al., 2023). Benzophenone, the breakdown product of octocrylene, is an herbicide
506 patented by both Monsanto and the Rohm & Haas Company (Downs et al., 2021). The major
507 metabolite of benzophenone, 4-hydroxybenzophenone, is a powerful estrogenic endocrine
508 disruptor (Carstensen et al., 2023). Ecotoxicological studies should be conducted using native
509 plants species from this area to generate a relevant ecological risk assessment for these chemicals
510 on their impact to native floral species, and their threat to the ecological-services structures they
511 provide (Feagin et al., 2013; Feagin et al., 2019). As a result of sunscreen chemical toxicity,
512 decreased botanical root densities in these dunes may make them more susceptible to erosion
513 from storm action (Li et al., 2023). Rising sea levels with increasingly intense storms threaten
514 North Carolina's barrier islands; therefore, it is important to address factors that decrease
515 resilience to coastal erosion (Sweet et al., 2022; Balaguru et al., 2023).

516
517 Although plants can act as keystone species for building-up and sustaining dune
518 landscapes, ghost crabs (Genus *Ocypode*) contribute to the bioerosion and aeration of dune
519 structures, as well as beach sedimentary turnover and mixing (Hughes et al., 2014). They are a
520 vital component of the beach landscape food web, acting as an important food source for birds
521 and mammals (Wolcott, 1978). Ghost crabs are an indicator-species for the health condition of
522 coastal dunes impacted by tourism (Schlacher et al., 2010; Noriega et al., 2012; Schlacher et al.,
523 2016). With high concentrations of sunscreen chemicals in both national park units, it begs the
524 question of whether sunscreen ingredients could be the etiological drivers for tourism-associated
525 declines of ghost crab demographics and detrimental changes in social behaviors, especially
526 because oxybenzone, octocrylene, benzophenone, and avobenzone are renowned endocrine
527 disruptors (Campos et al., 2019; Muñoz-González and Martínez-Guitarte, 2020; Boyd et al.,

528 2021). Decapods are susceptible to estrogenic endocrine disruption, especially to estrogenic-axis
529 disruption which will inappropriately induce vitellogenin production in males, exhibit significant
530 shifts in sex ratios in decapod populations, and display a reduction in population persistence
531 (Brian, 2005; Lye et al., 2008; Matozzo et al., 2008; Park et al., 2014; Langston, 2020).
532 Furthermore, oxybenzone and octocrylene's degradate, benzophenone, are genotoxic and the
533 high concentration of these chemicals on beach sands may explain why high levels of
534 genotoxicity in ghost crabs were associated with tourist/recreational beaches (de Lima de Silva et
535 al., 2002; Ma et al., 2023). Ghost crabs are not the only decapod species under threat. The
536 Barden Inlet Blue Crab Spawning Sanctuary is within the boundaries of the Cape Lookout
537 National Seashore, between Shackelford Banks and South Core Banks. This Sanctuary is
538 immediately adjacent to the Cape Lookout sampling site, and rests within the pollution plume of
539 this site. The tourism's sunscreen pollution plume in this area could potentially impact this
540 spawning habitat.

541
542 Many species of migratory birds subsist on ghost crabs, and their principle feeding
543 grounds are coastal beaches. Storks, harriers, gulls, eagles, kestrels, and other carnivorous bird
544 species were shown to have excessive amounts of oxybenzone and its metabolites and
545 octocrylene accumulating in eggs (Molins-Delgado et al., 2017; Oró-Nolla et al., 2021).
546 Octocrylene was detected in 100% of the bird egg samples from the variety of species studied;
547 therefore, benzophenone (a known carcinogen and reproductive toxicant) may also be a
548 contaminant (Downs et al., 2021). Ghost crabs' primary habitat is regularly visited by tourists
549 and local recreationists; their shoreline habitat is highly contaminated with sunscreen pollutants.
550 Could a diet of ghost crabs inhabiting sunscreen-polluted beaches be a major factor for egg
551 contamination and subsequent hatching success? The bioaccumulation and biomagnification of
552 these sunscreen chemicals need to be assessed in wildlife settings, starting with a rigorous survey
553 of contaminant loads in ghost crabs from various locations.

554
555
556 The beaches of Kaloko-Honokōhau are designated Critical Habitat for the recovery of
557 monk seals under the Endangered Species Act (Federal Register, 2015). Hawaiian Monk seals
558 (*Neomonachus schauinslandi*) may spend about 1/3 of their time resting on beaches in the main
559 Hawaiian Islands (Wilson et al., 2017). In Kaloko-Honokōhau, seals have been recorded to haul
560 out and rest at some of the same locations the samples were collected. In mammals, oxybenzone,
561 avobenzone, benzophenone and octocrylene can be readily absorbed through the skin and
562 detected in blood samples after two weeks from a single acute exposure (Matta et al., 2019;
563 Matta et al., 2020; Downs et al., 2021; Pilli et al., 2021). Increasing exposure concentration often
564 translates into increased body absorption, and with no vehicle formulation to protect or mitigate
565 the rate of absorption, exposure directly from sand may result in a very high rate of absorption
566 (Kurul and Hekimoglu, 2001; Gonzalez et al., 2006; Do et al., 2022). Octocrylene has been
567 shown to be carried at a high body-burden concentration in cetaceans, and octocrylene
568 contamination can be passed directly from mother to neonate via breastmilk, suggesting a similar
569 phenomenon in pinnipeds (Gago-Ferrero et al., 2013; Alonso et al., 2015). Octocrylene can
570 directly reduce Vitamin D production (Abdi et al., 2022). Octocrylene accumulation from beach-
571 reposed seals may affect Vitamin D seasonal variance, and result in deficiencies at vulnerable
572 times during reproductive development as well as immunocompetence (Wilske, 1993; Atkinson
573 and Gilmartin, 1992; Wilske and Arnbom, 1996). Monk seals may also consume high levels of
574 microplastics (Pietroluongo et al., 2022; McIvor et al., 2023). Marine plastic debris is a
575 concentrator of sunscreen chemicals, thus consumption of marine plastics in sunscreen polluted

576 waters may increase exposure risk and body burdens to pinnipeds that haul out on shore
577 (O'Donovan et al., 2020; Achar et al., 2021; Cui et al., 2022). Research on contaminant
578 accumulation in phocid seals is necessary to better understand this threat.

579
580 It is important to note that the water samples we collected were at least 30 cm below the
581 surface of the water. In Shackleford Bank, the water sample was collected about 10 meters from
582 the narrow swash zone, in and amongst over 120 swimmers in the water within 20 meters of the
583 collection point. Kaloko-Honokōhau is a protected embayment that is the remnants of a cultural
584 Hawaiian fishpond – it had no active swash zone on any of the collection days. Concentrations of
585 many of the sunscreen compounds were above the solubility in fresh water or salt water (36 ppt
586 salinity). This is because water column loading of organic compounds can increase the solubility
587 of these slightly hydrophobic and polyaromatic compounds (Witt, 1995; Dong et al., 2012;
588 Nasher et al., 2013; Abdel-Safy and Mansour, 2016; Zukausaute et al., 2021; Zheng et al.,
589 2023). Sunscreen compounds, such as oxybenzone and octocrylene, can readily be bound to
590 proteins for increased solubilization and bioavailability (Ao et al., 2015; Ma et al., 2023a; Ma et
591 al., 2023c; Tsurushima et al., 2023). Proteins and acyl-chain compounds are major components
592 of marine flocculence and organic carbon in the marine water column allowing for the increased
593 availability within the length of the water column (Lee et al., 2000; Sun et al., 2019). This is
594 why we see a fraction of the sunscreen compounds on the surface of the water, and can detect
595 sunscreen compounds meters deep in waters amongst corals (Tsui et al., 2017; He et al., 2023).
596 Coral themselves are major sources of organic matter in the water column of coral reefs;
597 including sunscreen solubilizing solvents such as dimethyl sulfoxide and
598 dimethylsulphoniopropionate (Ferrier-Pages et al., 1990; Broadbent and Jones, 2006;
599 Deschaseaux et al., 2015; Jones et al., 2015; Gardner et al., 2016; Gardner et al., 2017).

600
601 Aerosol-spray sunscreen was observed to be an obvious mode of environmental
602 contamination at both locations. Daily onshore winds at Cape Lookout frequently reach 10-20
603 knots (https://www.ndbc.noaa.gov/station_page.php?station=clkn7). Sunscreen drift from
604 aerosol applicators can be significant with as much as 60-95% of the discharge not deposited on
605 human skin. In a laboratory setting with no wind, the amount of aerosol sunscreen lost to the
606 environment and not deposited on the skin is over 61% of the discharge (Broussard et al., 2019).
607 Furthermore, to achieve the SPF protection on the product label, the amount of aerosol sunscreen
608 to be discharged is significantly higher than commonly assumed; a full body application to reach
609 labeled SPF levels would require more than a 90-second-long continuous discharge application
610 (Cancer Council, 2020). The high concentration of sunscreen contamination on surface sands is a
611 testament to this form of environmental contamination. Aerosol applications have also been
612 observed when beachgoers wade into the sea and discharge the product directly into receiving
613 waters (*e.g.*, Sample Site CW1, **Figure 1E, Supplemental Video 1**). In most observed cases, the
614 sunscreen-drift directly contaminates the water as the person applies the aerosol sunscreen.
615 Tourists and local visitors have also been observed to submerge their bodies and swim
616 immediately after sunscreen application of both lotions and aerosol applications without waiting
617 the 15 minutes for the matrix formula to polymerize as indicated by the product's directions for
618 use (personal observations). This failure to use the technology as directed increases the discharge
619 rate of the sunscreen product ingredients into the environment as well as causing "Sunscreen
620 Abuse" which can cause harm to the individual and public health by potentially increasing their
621 risk for UV-associated skin cancer (Autier, 2009; DiNardo and Downs, 2021). Estimates for the
622 amount of sunscreen contamination of a specific location can be calculated based on rigorous
623 surveys of the number of visitors and the percentage of those visitors using aerosol sprays (*e.g.*,

624 Downs et al., 2002c). Regulating the use of aerosol sunscreen sprays should be considered for
625 marine protected areas as a way of mitigating environmental contamination via sunscreen aerosol
626 drift (Logemann et al., 2022).
627

628 **5. Conclusion**

629 During the Anthropause of 2020, the concentrations of UV filters in water and sand samples at
630 two national park units in the U.S. were lower than at times of normal visitation. Because these
631 filters, at the levels present in the collected samples from after the Anthropause, can negatively
632 affect marine life, interventions will be necessary to avoid these deleterious effects. Previous
633 studies have identified several behavioral barriers that may prevent visitors from adopting
634 ecologically friendly sun protection behaviors. A multi-faceted approach, including educational
635 campaigns, behavior change strategies, and park-specific prohibitions would address many of the
636 challenges that parks face regarding sunscreen pollution while maintaining the quality of the
637 visitor experience.

638

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647
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667

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1687 **Figure 1.** Location of water and sand sampling sites. (Panel A) Sampling sites of water and sand
1688 samples within Kaloko-Honokōhau National Historical Park in Hawai'i, U.S.A. (Panel B)
1689 Locations of Kaloko-Honokōhau National Historical Park on the Island of Hawai'i, Hawaii,
1690 U.S.A. (Panel C) Location of the two sampling areas within the Cape Lookout National Marine
1691 Seashore where “D” indicates the Shackleford Banks sampling area and “E” indicates the South
1692 Core Banks sampling area. (Panel D) Location of the sand and water sampling sites at
1693 Shackleford Banks. (Panel E) Location of the sand and water sampling sites in the South Core
1694 Banks sampling area adjacent to the Cape Lookout Light House compound.

1695

1696 **Table 1.** Concentrations of sunscreen chemicals and their metabolites in water (Table 1A) and
 1697 beach sand (Table 1B) individual samples collected within the Kaloko-Honokōhau National
 1698 Historical Park in Hawai'i, U.S.A. Location of individual sample collection is shown in Figure
 1699 1A,B. Samples were collected in 2018-2021. Concentrations are in nanograms per liter. NA =
 1700 Not assayed. <LOQ = Below the Limit of Quantitation. <LOD = Below the Limit of Detection.

Table 1A

Water Samples

Conct.	ng/L	ng/L	ng/L	ng/L	ng/L	ng/L	ng/L	ng/L	ng/L	ng/L	ng/L
Year	2018	2018	2019	2020	2020	2020	2020	2020	2021	2021	2021
Sample location	W3	W5	W3	W1	W2	W3	W4	W5	W1	W2	W3
Avobenzene	898	702	20,710	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	94,000
Benzophenone	54	17	329	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Benzophenone-1	9	9	16	NA	NA	NA	NA	NA	NA	NA	NA
Benzophenone-4	<LOQ	<LOQ	17	NA	NA	NA	NA	NA	NA	NA	NA
Homosalate	220	<LOQ	24,710	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	129,000
Octinoxate	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Octisalate	500	170	8,100	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	65,000
Octocrylene	1,996	2,136	33,527	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	6,000	298,000
Oxybenzone	5,150	7,130	35,207	<LOQ	<LOQ	<LOQ	<LOQ	85	<LOQ	<LOQ	<LOQ

1701

Table 1B

Sand Samples

Conct.	ng/g	ng/g	ng/g	ng/g	ng/g	ng/g	ng/g	ng/g	ng/g	ng/g
Year	2018	2020	2020	2020	2020	2020	2020	2021	2021	2021
Sample location	S4	S1	S2	S3	S4	S5	S1	S2	S3	S4
Avobenzene	4,716	<LOD	<LOQ	<LOD	<LOQ	<LOD	<LOQ	1,678	987	12,137
Benzophenone	60	<LOD	<LOD	<LOD	<LOD	<LOD	72	28	24	110
Homosalate	<LOQ	<LOD	<LOD	<LOD	<LOQ	<LOD	<LOQ	18,860	7,271	50,688
Octinoxate	<LOQ	<LOD	<LOD	<LOD	<LOD	<LOD	<LOQ	<LOQ	<LOQ	<LOD
Octisalate	8,960	<LOD	<LOD	<LOD	<LOD	<LOD	<LOQ	9,814	11,916	29,946
Octocrylene	21,312	78	53	56	98	<LOD	33,493	26,794	10,548	22,860
Oxybenzone	11,735	<LOD	<LOD	<LOD	<LOD	<LOD	<LOQ	<LOQ	<LOQ	<LOQ

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1704 **Table 2.** Concentrations of sunscreen chemicals and their metabolites in individual water and
 1705 beach sand samples collected within the Cape Lookout National Marine Seashore in North
 1706 Carolina, U.S.A. Location of individual sample collection is shown in Figure 1(Panel C-E).
 1707 Table 2A are concentrations in water samples collected from the South Core Bank area (Figure
 1708 1E). Table 2B are concentrations in beach sand samples collected from the South Core Bank area
 1709 (Figure 1E). Table 2C are concentrations in beach sand samples collected at Shackleford Banks
 1710 area (Figure 1D). Samples were collected in 2020-2021. Concentrations are in nanograms per
 1711 liter. NA = Not assayed. <LOQ = Below the Limit of Quantitation. <LOD = Below the Limit of
 1712 Detection.

Table 2A. Concentrations in South Core Banks Water Sample

Concentration	ng/L	ng/L	ng/L
Year	2021	2021	2021
Sample location	CW1	CW2	SW1
Avobenzene	1,209,000	<LOQ	523,000
Benzophenone	2,000	<LOQ	2,000
Benzophenone-1	NA	NA	NA
Benzophenone-4	NA	NA	NA
Homosalate	56,000	<LOQ	941,000
Octinoxate	<LOQ	<LOQ	<LOQ
Octisalate	1,109,000	<LOQ	528,000
Octocrylene	1,950,000	<LOQ	1,304,000
Oxybenzone	1,888,000	3,000	331,000

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Table 2B. Concentrations in South Core Banks Sand Samples

Concentration	ng/g	ng/g	ng/g	ng/g	ng/g	ng/g	ng/g
Year	2020	2020	2020	2021	2021	2021	2021
Sample location	C1	C2	C3	C1	C2	C3	C4
Avobenzene	<LOQ	<LOQ	<LOQ	1,967	595	439	9,987
Benzophenone	<LOQ	<LOQ	<LOQ	54	<LOQ	<LOQ	137
Homosalate	<LOQ	<LOQ	<LOQ	23,578	5,096	3,552	20,584
Octinoxate	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Octisalate	<LOQ	<LOQ	<LOQ	23,692	12,051	4,466	15,560
Octocrylene	<LOQ	<LOQ	<LOQ	14,786	9,499	4,397	25,722
Oxybenzone	<LOQ	<LOQ	<LOQ	9,535	5,073	1,792	22,860

Table 2C. Concentrations in Shackleford Banks Sand Samples

Concentration	ng/g	ng/g	ng/g	ng/g	ng/g	ng/g	ng/g	ng/g	ng/g
Year	2020	2020		2021	2021	2021	2021	2021	2021
Sample location	S1	S3	S4	S5	S1	S2	S3	S4	S5
Avobenzene	<LOQ	<LOQ	<LOQ	<LOQ	375	1,347	<LOQ	2,721	<LOQ
Benzophenone	<LOQ	<LOQ	<LOQ	<LOQ	16	40	<LOQ	15	<LOQ
Homosalate	<LOQ	<LOQ	<LOQ	<LOQ	41,644	7,025	<LOQ	2,874	<LOQ
Octinoxate	<LOQ	<LOQ	<LOQ	10	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Octisalate	<LOQ	<LOQ	<LOQ	<LOQ	2,725	4,325	<LOQ	3,705	<LOQ
Octocrylene	<LOQ	<LOQ	<LOQ	<LOQ	7,179	15,172	<LOQ	10,590	827
Oxybenzone	<LOQ	<LOQ	<LOD	<LOD	2,604	7,408	89	5,066	199

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1716 **Table 3.** Risk Quotient for Acute Toxicity of oxybenzone and octocrylene in water samples
1717 collected at Kaloko-Honokōhau National Historical Park in Hawai'i, U.S.A from 2018-2021
1718 associated with Table 1A for Cnidarian species, invertebrate (non-Cnidarian) species, plant and
1719 algae species, and fish species. Color chart: RED= Severe condition for a potential toxic effect
1720 ≥ 1 ; Orange = Moderate threat condition for a potential toxic effect = 0.5 to 1.0; Yellow= Low
1721 condition of concern 0.49 to 0.1.

Oxybenzone				2018 W3	2018 W5	2019 W3	2020 W1	2020 W2	2020 W3	2020 W4	2020 W5	2021 W1	2021 W2	2021 W3		
SPECIES				5	7	35	0	0	0	0	0.09	0	0	0	Environ. Concen. µg/L	
															References for Toxicity Reference Values	
Coral cells in vitro																
Pocillopora damicornis	coral cells 4hr light	LC ₅₀	8.00	644	891	4,400	0	0	0	0	11.25	0.00	0.00	0.00	Downs et al. 2016	
Stylophora pistillata	coral cells 4hr dark	LC ₅₀	679.00	8	11	52	0	0	0	0	0.13	0.00	0.00	0.00	Downs et al. 2016	
Coral & Cassiopeia Planula (early life stage)																
Cassiopea xamachana	Planula Lethality	NOEC	2.28	2,259	3,127	15,439	0	0	0	0	39.47	0.00	0.00	0.00	Fitt & Hoffman 2020	
Cassiopea frondosa	Planula Lethality	NOEC	2.28	2,259	3,127	15,439	0	0	0	0	39.47	0.00	0.00	0.00	Fitt & Hoffman 2020	
Stylophora pistillata	Planula 24h light	LC ₅₀	139.00	37	51	253	0	0	0	0	0.65	0.00	0.00	0.00	Downs et al. 2016	
Stylophora pistillata	Planula DNA Damage 8h light	NOEC	11.40	452	625	3,088	0	0	0	0	7.89	0.00	0.00	0.00	Downs et al. 2016	
Stylophora pistillata	Planula bleaching 8h light	NOEC	1.14	4,518	6,254	30,877	0	0	0	0	78.95	0.00	0.00	0.00	Downs et al. 2016	
Invertebrates																
Corbicula fluminea	clams 96h filter feeding inhibition	NOEC	315.00	16	23	112	0	0	0	0	0.29	0.00	0.00	0.00	Seoane et al. 2020	
Daphnia magna	neonate crustacean 48h mortality	LC ₅₀	1,090.00	5	7	32	0	0	0	0	0.08	0.00	0.00	0.00	Du et al. 2017	
Daphnia magna	neonate crustacean 24h immobility	EC ₅₀	2,700.00	2	3	13	0	0	0	0	0.03	0.00	0.00	0.00	Molins-Delgado et al. 2016	
Paracentrotus lividus	sea urchin 48 h larval growth (4-arm-pluteus)	EC ₅₀	3,280.00	2	2	11	0	0	0	0	0.03	0.00	0.00	0.00	Paredes et al. 2014	
Mytilus galloprovincialis	mussel embryos 48h % normal D-larvae	EC ₅₀	3,472.59	1	2	10	0	0	0	0	0.03	0.00	0.00	0.00	Paredes et al. 2014	
Siriella armata	mysid, crustacean 24h neonates 96h mortality	EC ₅₀	710.76	7	10	50	0	0	0	0	0.13	0.00	0.00	0.00	Paredes et al. 2014	
Algae and vascular plants																
Arthrospira sp.	cyanobacteria 7d Net photosynthetic rate	NOEC	0.23	22,588	31,272	154,386	0	0	0	0	394.74	0.00	0.00	0.00	Zhong et al. 2019	
Chlorella sp.	green microalgae growth inhibition 20d	NOEC	0.02	225,877	312,719	1,543,860	0	0	0	0	#####	0.00	0.00	0.00	Zhong et al. 2019	
Cucumis sativus	vascular plant 24h photosynthesis	NOEC	570.00	9	13	62	0	0	0	0	0.16	0.00	0.00	0.00	Zhong et al. 2020	
Scenedesmus obliquus	Green algae 4d growth rate	NOEC	250.00	21	29	141	0	0	0	0	0.36	0.00	0.00	0.00	Lee et al. 2020	
Tetraselmis sp.	Marine green algae 7d Growth Inhibition	NOEC	1.00	5,150	7,130	35,200	0	0	0	0	90.00	0.00	0.00	0.00	Thorel et al. 2020	
Fish																
Betta splendens	Adult 28d aggressive opercular display	NOEC	5.00	1,030	1,426	7,040	0	0	0	0	18.00	0.00	0.00	0.00	Chen et al. 2018	
Carassius auratus	Adult 7d increased hepatic oxidative damage	NOEC	2.00	2,575	3,565	17,600	0	0	0	0	45.00	0.00	0.00	0.00	Zhang et al. 2020	
Danio rerio	Embryos 24h Vitellogenin1 Induction	NOEC	104.00	50	69	338	0	0	0	0	0.87	0.00	0.00	0.00	Meng et al. 2020	
Danio rerio	Phenotypic sex ratio Fewer males more females	NOEC	191.00	27	37	184	0	0	0	0	0.47	0.00	0.00	0.00	Kinnberg et al. 2015	
Danio rerio	Gonad maturation in females	NOEC	191.00	27	37	184	0	0	0	0	0.47	0.00	0.00	0.00	Kinnberg et al. 2015	
Danio rerio	Gonad maturation in males	NOEC	388.00	13	18	91	0	0	0	0	0.23	0.00	0.00	0.00	Kinnberg et al. 2015	
Danio rerio	Embryo -24 h Locomotor disruption	NOEC	5.00	1,030	1,426	7,040	0	0	0	0	18.00	0.00	0.00	0.00	Tao et al. 2020	
Oryzias latipes	21 d - vitellogenin in males	NOEC	61.00	84	117	577	0	0	0	0	1.48	0.00	0.00	0.00	Coronado et al. 2008	
Oryzias latipes	21 d % eggs hatching	NOEC	310.00	17	23	114	0	0	0	0	0.29	0.00	0.00	0.00	Coronado et al. 2008	
Poecilia reticulata	DNA Damage COMET Tail Length 96 h	NOEC	0.05	103,000	142,600	704,000	0	0	0	0	#####	0.00	0.00	0.00	Almeida et al. 2019	
Poecilia reticulata	DNA Damage Micronucleus 96 h	NOEC	0.50	10,300	14,260	70,400	0	0	0	0	180.00	0.00	0.00	0.00	Almeida et al. 2019	
Octocrylene																
SPECIES				2	2.1	34	0	0	0	0	0	0	6	298	Environ. Concen. µg/L	
															References for Toxicity Reference Values	
Coral (adult life stage)																
Stylophora pistillata	adult coral photosynthetic Fv/Fm	NOEC	519.00	4	4	65	0	0	0	0	0	0	11.6	574.2	Fel et al. 2018	
Pocillopora damicornis	adult coral polyp retraction	PNEC	500.00	4	4	67	0	0	0	0	0	0	12.0	596.0	He et al. 2019	
Pocillopora damicornis	adult coral polyp retraction	PNEC	150.00	13	14	223	0	0	0	0	0	0	40.0	1,986.7	Stien et al. 2019	
Invertebrates																
Daphnia magna	crustacean < 24 h neonates immobilization 48 h	EC ₅₀	30.00	67	70	1,117	0	0	0	0	0	0	200.0	9,933.3	Boyd et al. 2021	
Daphnia magna	crustacean < 24 h neonates mortality 48 h	LC ₅₀	3,600.00	1	1	9	0	0	0	0	0	0	1.7	82.8	Boyd et al. 2021	
Artemia salina	crustacean nauplii 48 h mortality	LC ₅₀	610.00	3	3	55	0	0	0	0	0	0	9.8	488.5	Thorel et al., 2020	
Ficopomatus enigmaticus	brackish water polychaete larval development	EC ₅₀	14,740.00	0	0	2	0	0	0	0	0	0	0.4	20.2	Sanches et al. 2021	
Mytilus galloprovincialis	mussel embryos 48h % normal D-larvae	NOEC	20.00	100	105	1,675	0	0	0	0	0	0	300.0	14,900.0	Giraldo et al., 2017	
Paracentrotus lividus	sea urchin - 48 h larval growth (4-arm-pluteus)	NOEC	20.00	100	105	1,675	0	0	0	0	0	0	300.0	14,900.0	Giraldo et al., 2017	
Algae and vascular plants																
Tetraselmis sp.	green microalgae, phytoplankton	PNEC	50.00	40	42	670	0	0	0	0	0	0	120.0	5,960.0	Thorel et al., 2020	
Isochrysis galbana	Haptophyta algae 72 h growth inhibition	NOEC	40.00	50	53	838	0	0	0	0	0	0	150.0	7,450.0	Giraldo et al., 2017	
Chlorella vulgaris	green microalgae 72 h growth inhibition	NOEC	3,120.00	1	1	11	0	0	0	0	0	0	1.9	95.5	Pablos et al., 2015	
Fish																
Oryzias latipes	Japanese medaka fecundity 28 d exposure	PNEC	2.50	800	840	13,400	0	0	0	0	0	0	2,400.0	#####	Yan et al., 2020	
Oryzias latipes	Japanese medaka F1 development 14 dph; 28 d FO exposure	PNEC	25.00	80	84	1,340	0	0	0	0	0	0	240.0	11,920.0	Yan et al., 2020	
Danio rerio	24 h embryos of dietary exposed F0 for 47 d	PNEC	3.50	571	600	9,571	0	0	0	0	0	0	1,714.3	85,142.9	Li et al., 2018	

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Table 4. Risk Quotient for Acute Toxicity of oxybenzone and octocrylene in beach sand samples collected at Kaloko-Honokōhau National Historical Park in Hawai'i, U.S.A from 2018, 2020, and 2021 associated with Table 1B for Annelida and Arthropoda species. Color chart: RED= Severe condition for a potential toxic effect ≥ 1 ; Orange = Moderate threat condition for a potential toxic effect = 0.5 to 1.0; Yellow= Low condition of concern 0.49 to 0.1.

Oxybenzone				2018 S4	2020 S1	2020 S2	2020 S3	2020 S4	2020 S5	2021 S1	2021 S2	2021 S3	2020 S4	Environ. Concen. ng/g
SPECIES				Toxicity Reference Value ng/g										References for Toxicity Reference Values
Annelida														
<i>Eisenia fetida</i>	Mortality, 14 days	LC50	364,000	32	0	0	0	0	0	0	0	0	0	Gautam et al 2022
<i>Eisenia fetida</i>	Growth rate, 14 days	NOEC	196,000	60	0	0	0	0	0	0	0	0	0	Gautam et al 2022
<i>Eisenia fetida</i>	Coccon production, 56 days	PNEC	18,200	645	0	0	0	0	0	0	0	0	0	Gautam et al 2022
<i>Eisenia fetida</i>	Reduction in Juvenile formation, 56 days	PNEC	3,640	3224	0	0	0	0	0	0	0	0	0	Gautam et al 2022
<i>Hediste diversicolor</i>	Avoidance behavior, 24 h	NOEC	10,000	1174	0	0	0	0	0	0	0	0	0	Clergeaud et al 2022
<i>Hediste diversicolor</i>	Survival rate, 28 days	NOEC	10,000	1174	0	0	0	0	0	0	0	0	0	Clergeaud et al 2022
Arthropoda														
<i>Sericostoma vittatum</i>	Feeding inhibition, 6 days	PNEC	1,775	6611	0	0	0	0	0	0	0	0	0	Campos et al 2017a
<i>Sericostoma vittatum</i>	Carbohydrates content, 6 days	PNEC	1,775	6611	0	0	0	0	0	0	0	0	0	Campos et al 2017a
<i>Sericostoma vittatum</i>	Oxidative stress & neurotoxicity, 6 days	NOEC	3,550	3306	0	0	0	0	0	0	0	0	0	Campos et al 2017a
<i>Sericostoma vittatum</i>	Glutathione levels, 6 days	PNEC	725	16186	0	0	0	0	0	0	0	0	0	Campos et al 2017a
<i>Chironomus riparius</i>	Larval growth inhibition, 28 days	PNEC	375	31293	0	0	0	0	0	0	0	0	0	Campos et al 2017b
<i>Chironomus riparius</i>	Delayed development, females, 28 days	PNEC	1,705	6883	0	0	0	0	0	0	0	0	0	Campos et al 2017b
<i>Chironomus riparius</i>	Delayed development, male, 28 days	NOEC	3,410	3441	0	0	0	0	0	0	0	0	0	Campos et al 2017b
<i>Chironomus riparius</i>	Decrease in adult bodyweight, 28 days	PNEC	1,705	6883	0	0	0	0	0	0	0	0	0	Campos et al 2017b
<i>Chironomus riparius</i>	Egg rope hatching	PNEC	1,000	11735	0	0	0	0	0	0	0	0	0	Campos et al 2019
<i>Chironomus riparius</i>	Emergence and developmental rate, 28 Days	PNEC	2,000	5868	0	0	0	0	0	0	0	0	0	Campos et al 2019

Octocrylene				2018 S4	2020 S1	2020 S2	2020 S3	2020 S4	2020 S5	2021 S1	2021 S2	2021 S3	2020 S4	Environ. Concen. ng/g
SPECIES				Toxicity Reference Value ng/g										References for Toxicity Reference Values
Annelida														
<i>Hediste diversicolor</i>	Survival rate, 28 days	NOEC	10,000	2,131	8	5	6	10	0	3,349	2,679	2,679	2,286	Clergeaud et al 2022
<i>Hediste diversicolor</i>	Avoidance behavior 24 hr	NOEC	10,000	2,131	8	5	6	10	0	3,349	2,679	2,679	2,286	Clergeaud et al 2022
Arthropoda														
<i>Chironomus riparius</i>	Larval growth inhibition, 28 days	PNEC	1,165	18,294	8	45	48	84	0	28,749	22,999	22,999	19,622	Campos et al 2017b
<i>Chironomus riparius</i>	Developmental delay of male and female, 28 days	NOEC	2,330	9,147	8	23	24	42	0	14,375	11,500	11,500	9,811	Campos et al 2017b
<i>Chironomus riparius</i>	Adults bodyweight, 28 days	NOEC	3,410	6,250	8	16	16	29	0	9,822	7,857	7,857	6,704	Campos et al 2017b

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1730 **Table 5.** Risk Quotient for Acute Toxicity of oxybenzone and octocrylene in water samples
1731 collected at South Core Banks within the Cape Lookout National Marine Seashore, North
1732 Carolina U.S.A in 2021 associated with Table 2A for Cnidarian species, invertebrate (non-
1733 Cnidarian) species, plant and algae species, and fish species. Color chart: RED= Severe
1734 condition for a potential toxic effect ≥ 1 ; Orange = Moderate threat condition for a potential toxic
1735 effect = 0.5 to 1.0; Yellow= Low condition of concern 0.49 to 0.1.

Oxybenzone				2021 Cape LO CW1	2021 Cape LO CW2	2021 Shackleford SW1	Environ. Concen. µg/L
				1950	3	331	
SPECIES	Toxicity Reference Value µg/L			References for Toxicity Reference Values			
				Coral & Cassiopeia Planula (early life stage)			
Cassiopeia xamachana	Planula Lethality	NOEC	2.3	855,263	1,316	145,175	Fitt & Hoffman 2020
Cassiopeia frondosa	Planula Lethality	NOEC	2.3	855,263	1,316	145,175	Fitt & Hoffman 2020
Stylophora pistillata	Planula bleaching 8h light	NOEC	1	1,710,526	2,632	290,351	Downs et al. 2016
Invertebrates							
Corbicula fluminea	clams 96h filter feeding inhibition	NOEC	315	6,190	10	1,051	Seoane et al. 2020
Daphnia magna	neonate crustacean 48h mortality	LC ₅₀	1,090	1,789	3	304	Du et al. 2017
Daphnia magna	neonate crustacean 24h immobility	EC ₅₀	2,700	722	1.1	123	Molins-Delgado et al. 2016
Paracentrotus lividus	sea urchin 48 h larval growth (4-arm-pluteus)	EC ₅₀	3,280	595	0.9	101	Paredes et al. 2014
Mytilus galloprovincialis	mussel embryos 48h % normal D-larvae	EC ₅₀	3,473	562	0.9	95	Paredes et al. 2014
Siriella armata	mysid, crustacean 24h neonates 96h mortality	EC ₅₀	711	2,744	4	466	Paredes et al. 2014
Algae and vascular plants							
Arthrospira sp.	cyanobacteria 7d Net photosynthetic rate	NOEC	0.23	8,552,632	13,158	1,451,754	Zhong et al. 2019
Chlorella sp.	green microalgae growth inhibition 20d	NOEC	0.02	85,526,316	131,579	14,517,544	Zhong et al. 2019
Cucumis sativus	vascular plant 24h photosynthesis	NOEC	570	3,421	5	581	Zhong et al. 2020
Scenedesmus obliquus	Green algae 4d growth rate	NOEC	250	7,800	12	1,324	Lee et al. 2020
Tetraselmis sp.	Marine green algae 7d Growth Inhibition	NOEC	1.00	1,950,000	3,000	331,000	Thorel et al. 2020
Fish							
Betta splendens	Adult 28d aggressive opercular display	NOEC	5	390,000	600	66,200.00	Chen et al. 2018
Carassius auratus	Adult 7d increased hepatic oxidative damage	NOEC	2	975,000	1,500	165,500.00	Zhang et al. 2020
Danio rerio	Embryos 24h Vitellogenin1 Induction	NOEC	104	18,750	29	3,182.69	Meng et al. 2020
Danio rerio	Phenotypic sex ratio Fewer males more females	NOEC	191	10,209	16	1,732.98	Kinnberg et al. 2015
Danio rerio	Gonad maturation in females	NOEC	191	10,209	16	1,732.98	Kinnberg et al. 2015
Danio rerio	Gonad maturation in males	NOEC	388	5,026	8	853.09	Kinnberg et al. 2015
Danio rerio	Embryo ~24 h Locomotor disruption	NOEC	5	390,000	600	66,200	Tao et al. 2020
Oryzias latipes	21 d % eggs hatching	NOEC	310	6,290	10	1,068	Coronado et al. 2008
Poecilia reticulata	DNA Damage COMET Tail Length 96 h	NOEC	0.05	39,000,000	60,000	6,620,000	Almeida et al. 2019
Poecilia reticulata	DNA Damage Micronucleus 96 h	NOEC	0.50	3,900,000	6,000	662,000	Almeida et al. 2019

Octocrylene				2021 Cape LO CW1	2021Cape LO CW2	2021 Shackleford SW1	Environ. Concen. µg/L
				1950	0	1304	
SPECIES	Toxicity Reference Value µg/L			References for Toxicity Reference Values			
				Invertebrates			
Daphnia magna	crustacean < 24 h neonates immobilization 48 h	EC ₅₀	30	65,000	0	43,467	Boyd et al. 2021
Daphnia magna	crustacean < 24 h neonates mortality 48 h	LC ₅₀	3,600	542	0	362	Boyd et al. 2021
Artemia salina	crustacean nauplii 48 h mortality	LC ₅₀	610	3,197	0	2,138	Thorel et al., 2020
Ficopomatus enigmaticus	brackish water polychaete larval development	EC ₅₀	14,740	132	0	88	Vieira Sanches et al. 2021
Mytilus galloprovincialis	mussel embryos 48h % normal D-larvae	NOEC	20	97,500	0	65,200	Giraldo et al., 2017
Paracentrotus lividus	sea urchin - 48 h larval growth (4-arm-pluteus)	NOEC	20	97,500	0	65,200	Giraldo et al., 2017
Algae and vascular plants							
Tetraselmis sp.	green microalgae, phytoplankton	PNEC	50	39,000	0	26,080	Thorel et al., 2020
Isochrysis galbana	Haptophyta algae 72 h growth inhibition	NOEC	40	48,750	0	32,600	Giraldo et al., 2017
Chlorella vulgaris	green microalgae 72 h growth inhibition	NOEC	3,120	625	0	418	Pablos et al., 2015
Fish							
Oryzias latipes	Japanese medaka fecundity 28 d exposure	PNEC	2.5	780,000	0	521,600	Yan et al., 2020
Oryzias latipes	Japanese medaka F1 development 14 dph; 28 d F0 exposure	PNEC	25	78,000	0	52,160	Yan et al., 2021
Danio rerio	24 h embryos of dietary exposed F0 for 47 d	PNEC	3.5	557,143	0	372,571	Li et al., 2018

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Table 6. Risk Quotient for Acute Toxicity of oxybenzone and octocrylene in beach sand samples collected at South Core Banks within the Cape Lookout National Marine Seashore, North Carolina U.S.A in 2021 associated with Table 2B for Annelida and Arthropoda species. Color chart: RED= Severe condition for a potential toxic effect ≥ 1 ; Orange = Moderate threat condition for a potential toxic effect = 0.5 to 1.0; Yellow= Low condition of concern 0.49 to 0.1.

Oxybenzone				2020 C1	2020 C2	2020 C3	2021 C1	2021 C2	2021 C3	2021 C4	
				0	0	0	9535	5073	1792	22860	Environ. Concen. ng/g
SPECIES			Toxicity Reference Value ng/g								References for Toxicity Reference Values
Annelida											
<i>Eisenia fetida</i>	Mortality, 14 days	LC50	364,000	0	0	0	26	14	5	63	Gautam et al 2022
<i>Eisenia fetida</i>	Growth rate, 14 days	NOEC	196,000	0	0	0	49	26	9	117	Gautam et al 2022
<i>Eisenia fetida</i>	Coccon production, 56 days,	PNEC	18,200	0	0	0	524	279	98	1,256	Gautam et al 2022
<i>Eisenia fetida</i>	Reduction in Juvenile formation, 56 days	PNEC	3,640	0	0	0	2,620	1,394	492	6,280	Gautam et al 2022
<i>Hediste diversicolor</i>	Avoidance behavior, 24 h	NOEC	10,000	0	0	0	954	507	179	2,286	Clergeaud et al 2022
<i>Hediste diversicolor</i>	Survival rate, 28 days	NOEC	10,000	0	0	0	954	507	179	2,286	Clergeaud et al 2022
Arthropoda											
<i>Sericostoma vittatum</i>	Feeding inhibition, 6 days	PNEC	1,775	0	0	0	5,372	2,858	1,010	12,879	Campos et al 2017a
<i>Sericostoma vittatum</i>	Carbohydrates content, 6 days	PNEC	1,775	0	0	0	5,372	2,858	1,010	12,879	Campos et al 2017a
<i>Sericostoma vittatum</i>	Oxidative stress & neurotoxicity 6 days	NOEC	3,550	0	0	0	2,686	1,429	505	6,439	Campos et al 2017a
<i>Sericostoma vittatum</i>	Glutathione levels, 6 days	PNEC	725	0	0	0	13,152	6,997	2,472	31,531	Campos et al 2017a
<i>Chironomus riparius</i>	Larval growth inhibition, 28 days	PNEC	375	0	0	0	25,427	13,528	4,779	60,960	Campos et al 2017b
<i>Chironomus riparius</i>	Delayed development females, 28 days	PNEC	1,705	0	0	0	5,592	2,975	1,051	13,408	Campos et al 2017b
<i>Chironomus riparius</i>	Delayed development male, 28 days	NOEC	3,410	0	0	0	2,796	1,488	526	6,704	Campos et al 2017b
<i>Chironomus riparius</i>	Decrease in adult bodyweight 28 days	PNEC	1,705	0	0	0	5,592	2,975	1,051	13,408	Campos et al 2017b
<i>Chironomus riparius</i>	Egg rope hatching	PNEC	1,000	0	0	0	9,535	5,073	1,792	22,860	Campos et al 2019
<i>Chironomus riparius</i>	Emergence and developmental rate 28 Days	PNEC	2,000	0	0	0	4,768	2,537	896	11,430	Campos et al 2019
Octocrylene (OC)											
				0	0	0	14786	9499	4397	25722	Environ. Concen. ng/g
SPECIES			Toxicity Reference Value ng/g								References for Toxicity Reference Values
Annelida											
<i>Hediste diversicolor</i>	Survival rate, 28 days	NOEC	10,000.00	0	0	0	1,479	950	440	2,572	Clergeaud et al 2022
<i>Hediste diversicolor</i>	Avoidance behavior, 24 hr	NOEC	10,000.00	0	0	0	1,479	950	440	2,572	Clergeaud et al 2022
Arthropoda											
<i>Chironomus riparius</i>	Larval growth inhibition, 28 days	PNEC	1,165.00	0	0	0	12,692	8,154	3,774	22,079	Campos et al 2017b
<i>Chironomus riparius</i>	Developmental delay of male and female, 28 days	NOEC	2,330.00	0	0	0	6,346	4,077	1,887	11,039	Campos et al 2017b
<i>Chironomus riparius</i>	Adults bodyweight, 28 days	NOEC	3,410.00	0	0	0	4,336	2,786	1,289	7,543	Campos et al 2017b

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Table 7. Risk Quotient for Acute Toxicity of oxybenzone and octocrylene in beach sand samples collected at Shackelford Banks within the Cape Lookout National Marine Seashore, North Carolina U.S.A in 2020-2021 associated with Table 2C for Annelida and Arthropoda species. Color chart: RED= Severe condition for a potential toxic effect ≥ 1 ; Orange = Moderate threat condition for a potential toxic effect = 0.5 to 1.0; Yellow= Low condition of concern 0.49 to 0.1.

Oxybenzone				2020 S1	2020 S3	2020 S4	2020 S5	2021 S1	2021 S2	2021 S3	2021 S4	2021 S5	
				0	0	0	0	2604	7408	89	5066	199	Environ. Concen. ng/g
SPECIES			Toxicity Reference Value ng/g										References for Toxicity Reference Values
Annelida													
<i>Eisenia fetida</i>	Mortality, 14 days	LC50	364,000	0	0	0	0	7	20	0	13.92	0.55	Gautam et al 2022
<i>Eisenia fetida</i>	Growth rate, 14 days	NOEC	196,000	0	0	0	0	13	38	0	26	1	Gautam et al 2022
<i>Eisenia fetida</i>	Coccon production, 56 days,	PNEC	18,200	0	0	0	0	143	407	5	278	11	Gautam et al 2022
<i>Eisenia fetida</i>	Reduction in Juvenile formation, 56 days	PNEC	3,640	0	0	0	0	715	2,035	24	1,392	55	Gautam et al 2022
<i>Hediste diversicolor</i>	Avoidance behavior, 24 h	NOEC	10,000	0	0	0	0	260	741	9	507	20	Clergeaud et al 2022
<i>Hediste diversicolor</i>	Survival rate, 28 days	NOEC	10,000	0	0	0	0	260	741	9	507	20	Clergeaud et al 2022
Arthropoda													
<i>Sericostoma vittatum</i>	Feeding inhibition 6 days	PNEC	1,775	0	0	0	0	1487	4174	50	2854	112	Campos et al 2017a
<i>Sericostoma vittatum</i>	Carbohydrates content, 6 days	PNEC	1,775	0	0	0	0	1487	4174	50	2854	112	Campos et al 2017a
<i>Sericostoma vittatum</i>	Oxidative stress & neurotoxicity, 6 days	NOEC	3,550	0	0	0	0	734	2087	25	1427	56	Campos et al 2017a
<i>Sericostoma vittatum</i>	Glutathione levels, 6 days	PNEC	725	0	0	0	0	3592	10218	123	6988	274	Campos et al 2017a
<i>Chironomus riparius</i>	Larval growth inhibition, 28 days	PNEC	375	0	0	0	0	6944	19755	237	13509	531	Campos et al 2017b
<i>Chironomus riparius</i>	Delayed development, females, 28 days	PNEC	1,705	0	0	0	0	1527	4345	52	2971	117	Campos et al 2017b
<i>Chironomus riparius</i>	Delayed development, male, 28 days	NOEC	3,410	0	0	0	0	764	2172	26	1486	58	Campos et al 2017b
<i>Chironomus riparius</i>	Decrease in adult bodyweight, 28 days	PNEC	1,705	0	0	0	0	1527	4345	52	2971	117	Campos et al 2017b
<i>Chironomus riparius</i>	Egg rope hatching	PNEC	1,000	0	0	0	0	2604	7408	89	5066	199	Campos et al 2019
<i>Chironomus riparius</i>	Emergence and developmental rate, 28 Days	PNEC	2,000	0	0	0	0	1302	3704	45	2533	100	Campos et al 2019
Octocrylene													
				2020 S1	2020 S3	2020 S4	2020 S5	2021 S1	2021 S2	2021 S3	2021 S4	2021 S5	
				0.00	0.00	0.00	0.00	7179	15172	0	10590	827	Environ. Concen. ng/g
SPECIES			Toxicity Reference Value ng/g										References for Toxicity Reference Values
Annelida													
<i>Hediste diversicolor</i>	Survival rate, 28 days	NOEC	10,000	0	0	0	0	718	1,517	0	1,059	83	Clergeaud et al 2022
<i>Hediste diversicolor</i>	Avoidance behavior, 24 hr	NOEC	10,000	0	0	0	0	718	1,517	0	1,059	83	Clergeaud et al 2022
Arthropoda													
<i>Chironomus riparius</i>	Larval growth inhibition, 28 days	PNEC	1,165	0	0	0	0	6,162	13,023	0	9,090	710	Campos et al 2017b
<i>Chironomus riparius</i>	Developmental delay of male and female, 28 days	NOEC	2,330	0	0	0	0	3,081	6,512	0	4,545	355	Campos et al 2017b
<i>Chironomus riparius</i>	Adults bodyweight, 28 days	NOEC	3,410	0	0	0	0	2,105	4,449	0	3,106	243	Campos et al 2017b

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