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Evaluating benthic recovery decades after a major oil spill in the Laurentian Great Lakes

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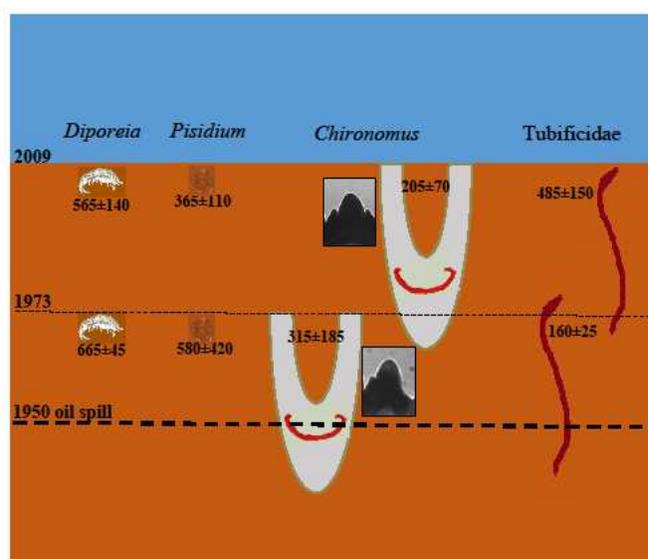
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1 **ABSTRACT:** The long-term effects of oil spills on freshwater organisms have been little
2 studied. In 1950, a large oil spill (10 million L) covered the harbor area of Parry Sound, Ontario,
3 the deepest port in the Laurentian Great Lakes. Ecological impacts were not studied at the time,
4 but 25 years later three-quarters of the *Chironomus cucini* larvae (Insecta, Diptera,
5 Chironomidae) living in the harbor area were reported to be deformed. We returned six decades
6 after the spill and found that the frequency of deformities had returned to background levels, and
7 that the community of burrowing invertebrates has largely recovered. By dating sediment cores
8 and measuring the depth distribution of oils, we conclude that, although the oil persists six
9 decades after the spill, sufficient uncontaminated sediment has covered the oil thereby putting it
10 out of reach of most burrowing animals. Provided that the sediment remains undisturbed, the
11 buried oil is unlikely to exert further negative effects on the biota in spite of the fact that it will
12 likely persist for centuries.

13 **TOC (Figure 3 of manuscript):**



14

15 Introduction

16 Large oil spills in marine systems have made names such as Amoco Cadiz (France 1978), Exxon
17 Valdez (Alaska 1989), and Deepwater Horizon (Gulf of Mexico 2010) infamous, and the effects
18 of such spills have been extensively documented.¹ Oil spills in fresh waters, while not of this
19 scale, can have major impacts in the short term,² whereas their long-term impacts and the
20 persistence of oil in freshwater ecosystems are unknown. There is an urgent need for such
21 information given the increasing rate at which oil sands and other terrestrial sources of oil are
22 being exploited and their products transported over great distances by pipelines, trains and ships.

23 In 1950, one of the largest oil spills in the history of the Laurentian Great Lakes occurred in
24 Parry Sound, Ontario, Canada, when the rupture of an oil storage tank spilled 10 million L of
25 heavy bunker oil into Parry Sound Harbor.³ To put this spill in perspective, a very large oil spill
26 into the Kalamazoo River, Michigan, involved less than half this amount (≈ 4 million L; Detroit
27 Free Press, 23 July 2011). Response measures after the 1950 Parry Sound spill were limited to a
28 few booms, ineffective attempts to burn-off the oil, and the use of sand to sink the oil to the lake
29 bottom.³ Two and a half decades after the spill, sediment collected in the profundal zone of Parry
30 Sound Harbor contained substantial quantities of oil.⁴ Furthermore, three-quarters of the
31 *Chironomus cucini* (Webb 1969) larvae (Insecta, Diptera, Chironomidae) collected from these
32 sediments had deformed mouthparts.⁴ The results of many studies have evoked contaminants
33 such as oil as the cause of high proportions of deformities in field populations of *Chironomus*
34 larvae^{4,5,6,7} and this supposition is supported by the results of laboratory experiments.^{8,9,10}

35 Six decades after the 1950 oil spill, we revisited Parry Sound where we collected large
36 numbers of *Chironomus cucini* larvae to determine if the frequency of larval deformities had
37 declined in the intervening decades. We also collected other invertebrates to determine if benthic

38 community composition had changed since it was measured in the 1970s. Lastly, we collected
39 cores that allowed us to determine the depth of sediment that has been deposited since the oil
40 spill, as well as to measure the vertical distribution of oil in the sediments and describe the
41 history of the chironomid populations at the contaminated site. Overall, these measurements
42 should allow us to relate the depth-distribution of oil in the sediment to the history and present
43 state of benthic invertebrate populations in the impacted area. Given the lack of long-term
44 studies on the impact of oil spills on freshwater biota, our data should be useful for estimating
45 response times for benthic recovery in comparable systems.

46 MATERIALS AND METHODS

47 **Study site and sampling rationale.** Parry Sound Harbor (45° 20' 0" N, 80° 2' 0" W;
48 surface area ~3 km²) is located at the eastern extremity of Parry Sound, a large bay (50 km²) on
49 the eastern shore of Georgian Bay, Lake Huron. Beginning in the 19th century, this major port
50 received contaminants from activities such as shipping, forestry, and sewage from the town of
51 Parry Sound.^{3,4} In 1950, the rupture of a shore-side storage tank released ~10 million L of heavy
52 bunker oil that covered much of the harbor and spread to the Sound itself.³ Two and a half
53 decades later (1973), three-quarters of the *Chironomus cucini* larvae collected from harbor
54 sediments were deformed.⁴

55 To determine if insects were still deformed 6 decades after the oil spill, we collected *C.*
56 *cucini* larvae in 2006 and 2009 from the station (station 1; 22-m depth) originally sampled by
57 Hare and Carter⁴ (station 8; see map in Hare and Carter⁴). In 2009, we also collected *C. cucini* at
58 an additional station situated ~0.7 km to the east (station 2; 24-m depth) as well as sediment
59 cores and grab samples for the determination of benthic community structure and the

60 measurement of contaminants. Historical and current values are compared to those for a
61 reference station in Parry Sound (14-m depth; sampled in 2009) that is located beyond the reach
62 of the oil spill (station 34 in Hare and Carter⁴). All stations sampled are in the profundal zone,
63 and larvae of *C. cucini* are reported to be limited to this zone in Parry Sound.⁴ *Chironomus*
64 *cucini* are recognizable as members of the *salinarius* group of *Chironomus* species by the fact
65 that larvae lack ventral and lateral tubules on their abdomen.¹¹ *Chironomus cucini* was reported
66 to be the only *salinarius*-group *Chironomus* species present in the hypolimnion of Parry Sound
67 Harbor,⁴ and this identification has since been confirmed using a combination of morphological,
68 cytological and genetic techniques.¹²

69 **Sediment cores.** We collected two sediment cores (A and B; length 50 cm) at station 1
70 using a gravity corer. To avoid sediment displacement during transport, the ends of the core
71 samples were plugged using floral foam. In the laboratory, the polyvinyl-chloride core tubes
72 (6.7-cm diameter) were cut lengthwise into halves using a miniature circular saw and a wire was
73 used to separate the sediment lengthwise into two halves. For ²¹⁰Pb dating and ¹³⁷Cs
74 measurements, half of core A was cut into 0.5-cm lengths for the first 10 cm and then into 1 cm
75 lengths for the remainder of the core. These fresh samples were weighed, freeze dried, weighed
76 again and ground to a powder of which a minimum of 1 g was placed into a pre-weighed vial and
77 counted for 24 h in an ultra-low-background, high-purity, germanium, gamma spectrophotometer
78 (ORTEC). Counts were treated and data interpreted according to Appleby¹³ and Couture et al.¹⁴

79 Total lead (Pb) measurements were obtained using an ITRAX X-ray fluorescence scanner
80 (Cox Analytical Systems). Measurements were made at intervals of 100 μm and the resulting
81 data were treated using ImageJ (<http://rsbweb.nih.gov/ij/>).

82 Total greases and oils were measured in subsamples collected at 0.5-cm intervals along one
83 of the longitudinal halves of core B. To glass vials we added 4 g of homogenized sediment, 0.8 g
84 of anhydrous magnesium sulfate (MgSO_4), and 16 mL of hexane. Vials were held in an
85 ultrasonic bath for 10 min, shaken for 10 min, and set aside to allow the MgSO_4 to precipitate.
86 The supernatant was pipetted through a Millex 0.8- μm filter into a preweighed glass vial and
87 then evaporated in a rotary evaporator at a temperature above that of hexane volatilization (69
88 $^\circ\text{C}$). The containers were held overnight in a desiccator and then weighed to obtain the weight of
89 total greases and oils in the each sample; this value was divided by the weight of the whole
90 sample to obtain the concentration of total greases and oils.

91 **Trace metals in sediments and larvae.** Since trace metals are reported to cause deformities
92 in *Chironomus* larvae,^{8,10} we measured cadmium (Cd), copper (Cu), nickel (Ni), thallium (Tl)
93 and zinc (Zn) in surface sediments and in larvae collected in 2009 at station 1 in Parry Sound
94 Harbor and at the reference site. At each site, oxic sediment was collected by scraping a small
95 plastic spatula along the surface (0-0.5 cm) of intact Ekman grab (15 x 15 x 23 cm) samples and
96 then remaining sediment was sieved through a 0.5-mm mesh-aperture net to concentrate *C.*
97 *cucini* larvae. In the laboratory, larvae were held in lake water for 4 days to allow them to
98 evacuate their gut contents. Fifteen well-depurated fourth-instar larvae were chosen from each
99 site and their head capsules were detached and preserved in alcohol for later examination for
100 deformities. Larval bodies and sediment samples were frozen at $-20\text{ }^\circ\text{C}$ on pieces of pre-weighed
101 acid-washed Teflon sheeting held in acid-washed 1.5 mL Eppendorf tubes then freeze dried and
102 weighed. We then added Omni Trace nitric acid (100 μL per mg of sample) to each larval and
103 sediment sample as well as to samples of similar weight of certified reference materials (MESS-3
104 and PACS-2 for sediments and TORT-2 (lobster hepatopancreas) and bovine liver for biological

105 samples). After five days at room temperature, hydrogen peroxide (60 μ L of Trace Select Ultra
106 per mg of samples) was added and samples were held for a further two days. Samples were
107 topped up with ultra-pure water to a volume of 1 mL per mg of sample. Trace metals were
108 measured by inductively coupled plasma – mass spectrometry; values of certified standards fell
109 within the range of certified values.¹⁵

110 **Collection and study of benthic invertebrates.** In 2009, we collected 10 Ekman grab (15 x
111 15 x 23 cm) samples at each of stations 1 and 2 to measure the composition of the extant benthic
112 community. Samples were sieved using a 0.5-mm mesh-aperture net and preserved in 10%
113 formalin to which we added a vital stain (Rose Bengal) to facilitate recovery of the invertebrates.
114 We compared the population densities obtained with those from samples collected in 1973
115 (triplicate samples collected by Ekman grab in July).¹⁶

116 To retrieve subfossil chironomid head capsules, we cut a longitudinal half of core A into 5-
117 mm long slices that were sieved through a 100- μ m mesh-aperture sieve. Material retained was
118 examined in a Bogorov dish under a microscope and head capsules found were mounted in
119 Canada balsam on microscope slides.

120 In 2006 and 2009, we collected large numbers of *Chironomus cucini* larvae at station 1 in
121 Parry Sound Harbor. In 2009, *C. cucini* larvae were also collected at station 2 in Parry Sound
122 Harbor and at the reference site. Larvae were collected by Ekman grab, the contents of which
123 were sieved using a 0.5-mm mesh-aperture net and held in lake water at field temperatures for
124 transport back to the laboratory where larvae were removed and preserved in 10% formalin. To
125 measure deformities, the head capsule of each larva was removed and held in 10% potassium
126 hydroxide (KOH) for 30 minutes to digest soft tissues. The KOH was neutralized by holding the

127 head capsule in concentrated acetic acid for 30 seconds and then it was transferred to 95%
128 ethanol before being placed in Canada balsam on a microscope slide where it was cut laterally to
129 separate it into dorsal and ventral halves. The mandibles and antennae were removed and all
130 parts were covered with cover glasses and the slide was left to dry at room temperature for
131 several weeks. Several parts of the larval head capsule were found to be deformed including the
132 mentum, mandibles, pectin epipharyngis and the antennae as illustrated in Fig. S1.

133 **RESULTS AND DISCUSSION**

134 **Sediment Chronology.** To estimate the chronology of events in Parry Sound Harbor and to
135 determine the depth of sediment that had accumulated since the oil spill, we measured ^{210}Pb
136 activity in a sediment core collected at station 1. The cumulative curve of unsupported ^{210}Pb
137 activity (not shown) indicated that sedimentation rates in Parry Sound Harbor have varied
138 somewhat over time. Using the Constant Rate of Supply model,^{13,14} sediment depth was related
139 to the year in which the sediment was deposited (Fig. 1). The reliability of the resulting ^{210}Pb
140 profile (Fig. 1) is supported by the fact that peak ^{137}Cs values occurred in the early 1960s (Fig.
141 2a) when inputs of fallout from atmospheric nuclear testing would have been at a maximum.¹³
142 Further support for the ^{210}Pb dates comes from the relative concentrations of total Pb that
143 increase sharply through the 1920s, reach a maximum between the 1950s and the 1980s, and
144 then decline up to the present day (Fig. 2b). Such a pattern is coherent with the use of Pb as an
145 additive in gasoline.¹⁷

146 Concentrations of total greases and oils in sediment were below the detection limit at
147 sediment depths greater than 6.5 cm and less than 4 cm, whereas between these depths

148 concentrations ranged from 75 to 175 mg/kg (Fig. 2c). The large depth range of the oil suggests
149 that there was some mixing of oil and previously deposited sediment at the time of the spill.

150 Measurements of ^{210}Pb , ^{137}Cs , total Pb and total greases and oils were consistent in
151 suggesting that ≈ 4.5 cm of sediment has accumulated at station 1 in Parry Sound Harbor since
152 the 1950 oil spill. The relationship between sediment depth and ^{210}Pb dates (Fig. 1) is consistent
153 with a published value for mean sedimentation rates (0.78 mm per year) in the near-shore waters
154 of Georgian Bay,¹⁸ since, at this rate, ≈ 4.5 cm of sediment would have accumulated between
155 1950 and 2009. The upper boundary of sediment rich in oil was also found at a depth of ≈ 4.5 cm
156 (Fig. 2c), which suggests that oil from the 1950 spill has remained buried below this depth in the
157 sediment of Parry Sound Harbor.

158 **Trace metals in sediments and in *Chironomus cucini*.** We measured the trace metals Cd,
159 Cu, Ni, Tl and Zn in sediments and larvae of *C. cucini* to determine if these contaminants could
160 be responsible for deformities (as illustrated in Fig. S1). Concentrations of all trace metals,
161 except copper, were significantly higher in sediments from Parry Sound Harbor (station 1, 2009)
162 than at a reference site (Table 1), which is explained by the harbor's history as a centre of
163 transport and industry for the region.³ In contrast, larvae from station 1 and the reference site
164 showed no significant differences in their concentrations of most trace metals (Table 1;
165 exceptionally, cadmium concentrations were marginally higher in larvae from Parry Sound
166 Harbor). These results suggest that metals alone cannot explain the biological effects measured
167 in Parry Sound Harbor. Indeed, much higher concentrations of Cd, Cu, Ni and Zn have been
168 measured in *Chironomus* larvae from lakes in a mining area near Parry Sound (Sudbury,
169 Ontario) and these larvae did not have elevated levels of deformities (¹⁹; Proulx and Hare,

170 unpublished). Likewise, laboratory experiments have shown that high incidences of deformities
171 in *Chironomus* larvae collected from contaminated lake sediments could not be explained by the
172 high lead content of the sediments.²⁰ Although trace metals are reported to cause deformities in
173 *Chironomus* larvae in the laboratory,^{8,9,10,21} caution should be used when inferring cause and
174 effect relationships between high concentrations of trace metals in field sediments and
175 incidences of larval deformities. Measurements of trace metal concentrations in burrowing
176 insects such as *Chironomus* would provide better estimates of metal bioavailability,^{22,23} which
177 could then be compared to the prevailing incidence of deformities. In conclusion, our data
178 suggest that the trace metals we measured are not the cause of deformities in *C. cucini* larvae,
179 however, we cannot rule out synergistic interactions between trace metals and oil in Parry Sound
180 Harbor.

181 **Changes in benthic community composition over time.** Benthic communities have been
182 severely impacted by oil spills in both marine^{24,25} and freshwater²⁶ ecosystems. Their recovery is
183 reported to be more rapid in shallow, hard, substrates subject to strong currents than in deeper
184 regions characterized by soft sediments in both marine^{24,25} and freshwater²⁶ systems. Thus
185 recovery of the benthic community in the cold, deep water, soft muds that we studied is likely to
186 relatively slow.

187 We compared densities of the numerically-dominant benthic taxa collected both 23 years
188 after the 1950 oil spill (in 1973)¹⁶ and 59 years after the spill (in 2009) to determine if they had
189 changed in the intervening decades. Note that the value of these comparisons is limited by the
190 small sample sizes in 1973 ($n = 3$)¹⁶ and the lack of data from intervening years.

191 Densities of the amphipod crustacean *Diporeia hoyi* S. I. Smith, 1874, and the sphaeriid
192 mollusk *Pisidium* spp. were not significantly different in 1973 and 2009 (Fig. 3), which suggests
193 that populations of these taxa were well-established 23 years after the oil spill. Larvae of
194 *Chironomus cucini* were also present at station 1 in similar densities in 1973 and in 2009 (Fig.
195 3), which suggests that its population density, if the same at the time of the spill, had recovered
196 by 1973. However, the high incidence of deformities reported for this species in the mid-1970s,⁴
197 suggests continued contact with oily sediment at this time (as discussed below). Mean densities
198 of Tubificidae were lower in 1973 than in 2009 (Fig. 3), which suggests that populations of these
199 deep burrowing, sediment feeding, worms were affected by the spill but may not have recovered
200 by 1973.

201 We hypothesized that the recovery of a given benthic taxon in Parry Sound Harbor will
202 depend on the depth to which it burrows and feeds, with species living in near-surface sediments
203 recovering first as uncontaminated sediment gradually buries the spilled oil.

204 Given that the amphipod crustacean *Diporeia hoyi* (Fig. 3) feeds on freshly deposited
205 sediment and burrows little below the sediment surface,^{27,28} this species was likely one of the
206 first to recover from the oil spill. The sphaeriid mollusk *Pisidium* spp. (Fig. 3) should be one of
207 the next taxa to colonize station 1 since it lives within the first 5 mm of sediment where it is
208 reported to filter interstitial water to feed on micro-organisms.²⁹ Judging from the similarity
209 between the densities of these two taxa in 1973 and 2009, we assume that if they were affected
210 by the 1950 spill they had recovered sometime prior to 1973. By this date \approx 2-2.5 cm of
211 uncontaminated sediment should have covered the oil (Fig. 1), which should be sufficient for
212 these shallow-burrowing taxa (Fig. 3).

213 Several lines of evidence suggest that although the population densities of *Chironomus*
214 *cucini* were similar in 1973 and 2009 (Fig. 3), larvae of this species continued to contact oily
215 sediment in the mid-1970s. First, final-instar *Chironomus* larvae are reported to construct tubes
216 to mean depths of ~4 cm in fine sediment,¹⁹ whereas only ≈2-2.5 cm of uncontaminated sediment
217 had been deposited by 1973 (Figs. 1 and 3). Second, at ~2 cm in maximum length, vertically-
218 oriented larvae would have been close to the contaminated zone. Third, *C. cucini* larvae do not
219 feed in the water-column or on surface sediment, but on anoxic sediment below the surface oxic
220 zone.³⁰ Fourth, the high incidence of deformities (Fig. S1) reported for this species two and a
221 half decades after the oil spill,⁴ indicates continued contact with oily sediment at this time (as
222 discussed below).

223 Tubificid oligochaetes (Fig. 3), and the tubificid genera reported to occur at station 1 in
224 Parry Sound Harbor (*Ilyodrilus*, *Limnodrilus*, *Rhyacodrilus*, and *Tubifex*),³¹ are thought to
225 construct vertical burrows in soft sediments and to consume deep sediment in a head-down
226 position.^{32,33} The fact that the mean values of Tubificidae reported for 1973 were lower than
227 those we measured in 2009 (Fig. 3) suggests that populations of these deep burrowing and
228 feeding worms were affected by the spill but may not have fully recovered by 1973.

229 **Temporal changes in the chironomid community.** We also evaluated historical trends in
230 Parry Sound Harbor by studying changes in the chironomid community as revealed by head
231 capsules collected at various depths in a sediment core collected at station 1. In the decade or so
232 following the oil spill (corresponding to a core depth of ≈4 to 4.5 cm), the numbers of the
233 numerically-dominant chironomid groups, that is, the Orthoclaadiinae (mainly
234 *Heterotrissocladius*), the Chironomini (mainly *Chironomus*, *Dicrotendipes*, *Phaenopsectra*, and

235 *Polypedilum*), and the Tanytarsini (mainly *Micropsectra* and *Tanytarsus*) were reduced but
236 returned to their previous densities after this time (Fig. 4, upper panels). Exceptionally,
237 Diamesinae (all *Protanypus*) were absent prior to the oil spill but present thereafter (Fig. 4). The
238 low numbers of this taxon and the lack of ecological data on *Protanypus* make interpretation of
239 this pattern difficult. Larvae of Tanypodinae were in low and varying abundances over the time
240 interval studied (Fig. 4). Overall, the total numbers of subfossil Chironomidae show a minimum
241 near the estimated depth of the buried oil (Fig. 4), which suggests that the chironomid
242 community was decimated by the oil spill but had recovered within a decade or two after the
243 spill.

244 Of the 34 subfossil head capsules of *C. cucini* found at all depths in the sediment cores, only
245 5 were deformed and, of these, 3 were found at the 4-4.5 cm depth interval, that is, near the time
246 of the oil spill.

247 **Temporal changes in deformed *Chironomus cucini*.** Examination of larval head capsules
248 showed that teeth on the mandibles, the mentum and the pecten epipharyngis were subject to loss
249 or deformation, as were the antennae (Fig. S1). Approximately two and a half decades after the
250 oil spill, 77% of the *C. cucini* larvae collected at station 1 in Parry Sound Harbor (Fig. 5A; 85
251 larvae measured) had a deformed mentum, mandibles or both⁴ suggesting that contaminants were
252 still having an effect on this population. At that time, the comparable figure for a reference site
253 outside of Parry Sound Harbor was 2.5% (Fig. 5A; 41 larvae measured). The results of other
254 studies also suggest that high incidences of deformities in *Chironomus* larvae are caused by
255 exposure to contaminants,^{34,35,36} whereas background levels of deformities are reported to be

256 much lower. For example, in nearby Georgian Bay, only 5.3% of *Chironomus* larvae were
257 reported to have a deformed mentum and 1.3% deformed mandibles.³⁷

258 In 2009, the percentage of deformed individuals in Parry Sound Harbor was 12% (of a total
259 of 146 larvae) at both stations 1 and 2 (Fig. 5A). We note that in 2006 a higher percentage of
260 deformed larvae was measured at station 1 (28.5% of 81 larvae), which suggests that the depth of
261 uncontaminated sediments covering the oil is spatially variable (the exact position of the
262 sampling site varied somewhat from year to year). The percentage of *C. cucini* larvae having a
263 deformed mentum or mandibles was very similar at the reference site (9% of 129 larvae) and at
264 the two contaminated sites in Parry Sound Harbor (12% of 103 in 2009; Fig. 5A), which suggests
265 that the majority of the Parry Sound Harbor population is no longer affected by contaminants. At
266 the reference site, the lower percentage of deformed individuals in the mid-1970s (2.5% of 41
267 larvae)⁴ compared to 2009 (9%; Fig. 5A) is likely explained by the fact that the evaluator in 2009
268 (K. Bertrand) categorized a wider range of differences as being deformities that did the evaluator
269 of the earlier samples (L. Hare).

270 We found that deformities were not limited to the mentum and mandibles but that the
271 antennae and the pecten epipharyngis (part of the labium) could also be deformed (Fig. S1). In 3
272 of 4 cases (Fig. 5B), antennal deformities were at least as common as deformities of the mentum
273 and mandible. Individuals with deformities of the pecten epipharyngis were uncommon and were
274 recorded only in Parry Sound Harbor in 2009 (Fig. 5B). The fact that parts other than the
275 mentum and mandibles can be deformed suggests that the value of 77% deformed larvae in the
276 mid-1970s (Fig. 5A) would have been even higher if other head-capsule parts had been
277 examined. For a given part, deformities were further classified according to type (Figs. S1,6).
278 The only clear trend was that antennal deformities in larvae from Parry Sound Harbor mainly

279 involved the loss of segments, whereas those from the reference site had multiple ring organs in
280 the basal segment (Figs. S1,6).

281 The deformities we observed do not appear to have hindered *C. cucini* larvae, since their
282 population density was similar in the mid-1970s and in 2009 and because deformed larvae were
283 able to pupate successfully as determined by the fact that 5 of the 7 pupae that we collected had
284 attached larval head capsules with deformed mouthparts.

285 A persistent population of deformed larvae suggests tolerance to oil. If so, *C. cucini* larvae
286 could be used as sentinels to monitor recovery in the decades following an oil spill, since their
287 tolerance allows them to persist in the presence of buried oil and yet their developmental
288 sensitivity to the oil, as evidenced by deformities, allows for the quantification of contaminant
289 exposure. Six decades after the oil spill, the low background level of deformities suggests that
290 sufficient uncontaminated sediment has accumulated to allow *C. cucini* larvae to burrow and
291 feed without contacting oil-contaminated sediment (Fig. 3).

292 **Extrapolation from the Parry Sound Harbor oil spill.** Our data suggest that biological
293 effects on the benthos were extreme during the first decade or two following the oil spill, likely
294 due to invertebrates contacting the oil either directly or indirectly through volatile components
295 diffusing towards the sediment-water interface. If we assume that the burial of oily sediment
296 allowed recovery of the benthic community, then recolonization likely proceeded according to
297 the depths to which various taxa burrow, with surface-dwelling taxa reestablishing themselves
298 first. At a given site, the greater the sedimentation rate the faster the benthic community will be
299 able to completely recolonize sediments after an oil spill.

300 In the absence of light and at 4° C, the oil at our study site is sufficiently stable that it
301 continues to persist 6 decades after the oil spill. Although, it does not appear to be a current

302 threat to most benthic invertebrates this could change in the future if, for example, sediments
303 were disturbed by dredging or by the arrival of a deep-burrowing invading species. In lakes
304 where bottom sediments are subject to disturbance by wind-induced currents, much longer time
305 frames could be required before oil is safely buried at depth. Such considerations will be
306 important in designing and evaluating the effectiveness of artificial lakes that are created to cover
307 oil-rich tailings, such as those being planned for use in the oil sands of northern Alberta.³⁸ Since
308 our study is the first to address the long-term effects of oil in lakes, it will be useful for
309 evaluating the feasibility of such projects as well as the long-term recovery of freshwater benthic
310 communities impacted by oil.

311 **Supporting Information.**

312 The Supporting Information is available free of charge on the ACS Publications website at
313 DOI xxxx.

314 Figure S1. Deformed and normal structures of final instar *Chironomus cucini* larvae
315 collected from station 1 in Parry Sound Harbor.

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Table 1. Mean (\pm 95% CI) concentrations (nmol/g dry weight) of the trace metals Cd, Cu, Ni, Tl and Zn in sediments and in larvae of *Chironomus cucini* collected in 2009 at a reference site in Parry Sound and in Parry Sound Harbor (station 1). Overlap of confidence intervals indicates no significant difference between means.

Medium / station	Metal: Cd (nmol/g)	Cu (nmol/g)	Ni (nmol/g)	Tl (nmol/g)	Zn (nmol/g)
Sediment - reference	17 \pm 2	578 \pm 294	435 \pm 94	0.8 \pm 0.03	2987 \pm 703
Sediment - Parry Sound Harbor	37 \pm 3	884 \pm 79	703 \pm 99	1.7 \pm 0.03	8883 \pm 1014
<i>C. cucini</i> - reference	23 \pm 6	330 \pm 97	15 \pm 7	0.08 \pm 0.04	842 \pm 272
<i>C. cucini</i> - Parry Sound Harbor	63 \pm 29	217 \pm 127	26 \pm 5	0.04 \pm 0.02	1191 \pm 816

FIGURE CAPTIONS

Figure 1. Changes in sediment age with depth in core A collected at station 1 in 2009 from Parry Sound Harbor, as determined by ^{210}Pb measurements. Error bars are standard deviations (SD).

Figure 2. Depth profiles of variables in cores collected at station 1 in 2009 from Parry Sound Harbor: (a) ^{137}Cs activity ($\pm\text{SD}$, core A); (b) total Pb (core A); (c) total greases and oils (core B). Dates are derived from ^{210}Pb geochronology.

Figure 3. Schematic representation of presumed burrowing and feeding depths of the four numerically-dominant benthic invertebrates at station 1 in Parry Sound Harbor as related to the depths of the 1950 oil spill and to the sediment-water interface in 1973 and 2009 as well as their mean densities ($\pm 95\%$ confidence interval) in these two years. Also pictured is the middle tooth of the mentum of deformed (1973) and normal (2009) *Chironomus cucini* larvae.

Figure 4. Numbers of chironomid head capsules in 0.5-cm slices of core A collected from station 1 in Parry Sound Harbor. The 1950 oil spill corresponds to a sediment depth of ≈ 4.5 cm.

Figure 5. Deformities in *Chironomus cucini* from a reference station and two stations in Parry Sound Harbor. Larvae were collected in either the 1970s (1973 and 1976⁴) or 2009 (this study). a: percentage of larvae having either a deformed mentum or deformed mandibles (numbers examined given above bars). b: percentage of deformed larvae exhibiting various types of deformities (totals can exceed 100%, since individuals can have more than one deformed part).

Figure 6. Percentages of various types of deformities observed in the various parts of deformed *Chironomus cucini* larvae collected at two stations in Parry Sound Harbor (1 and 2) and at a reference station outside of Parry Sound Harbor. Larvae were collected in 2009 from all stations and in 2006 from station 1.

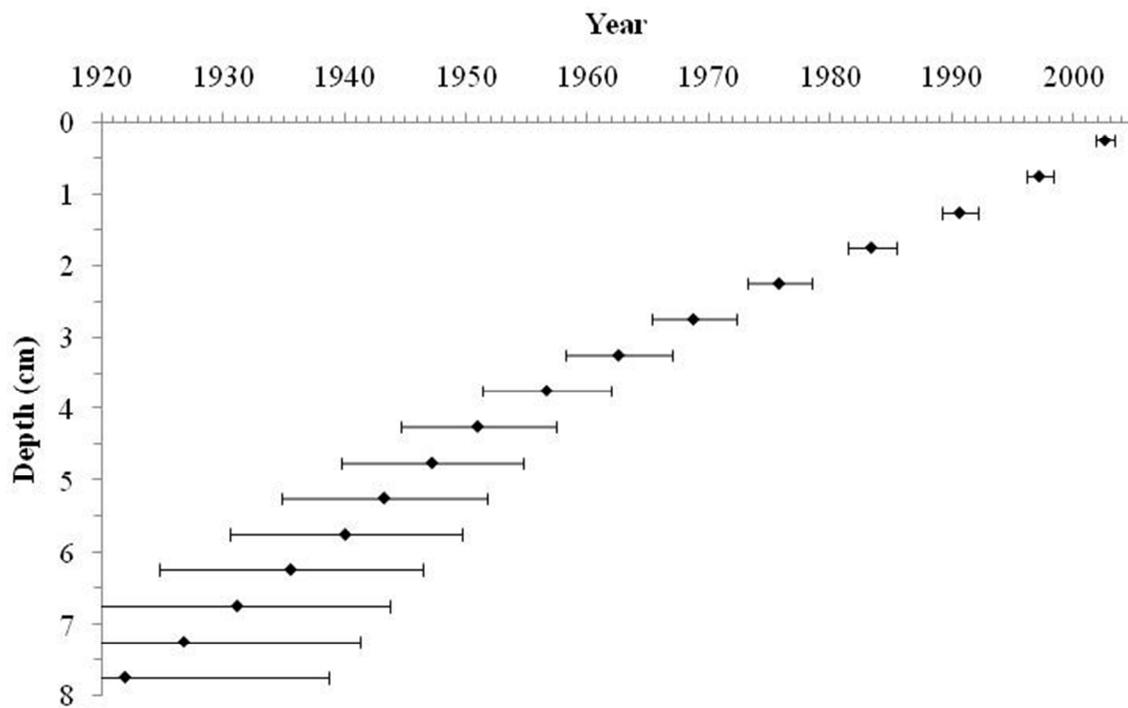


Figure 1.

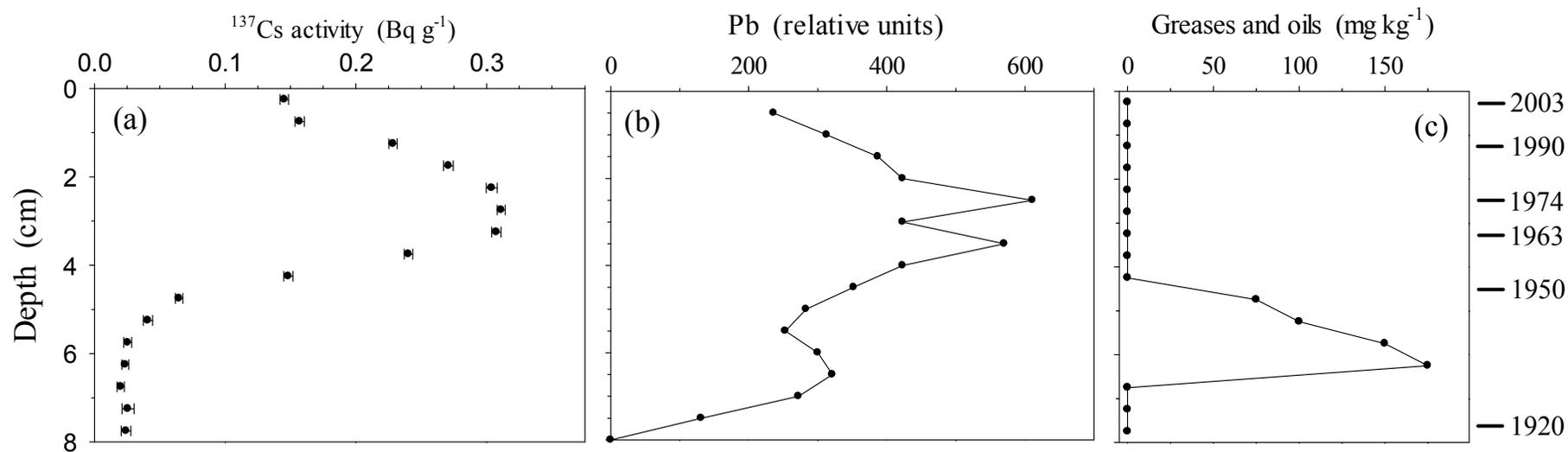


Figure 2.

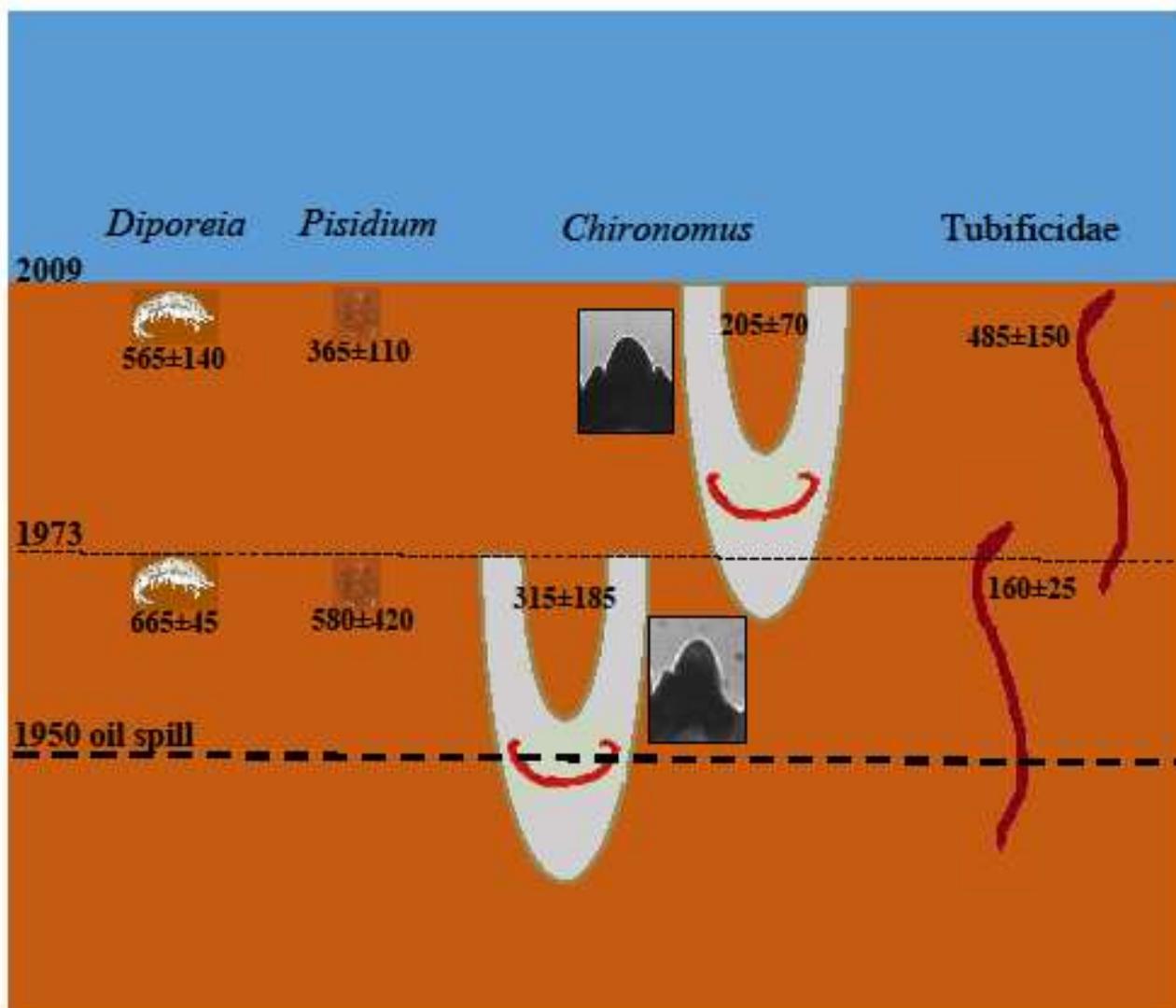
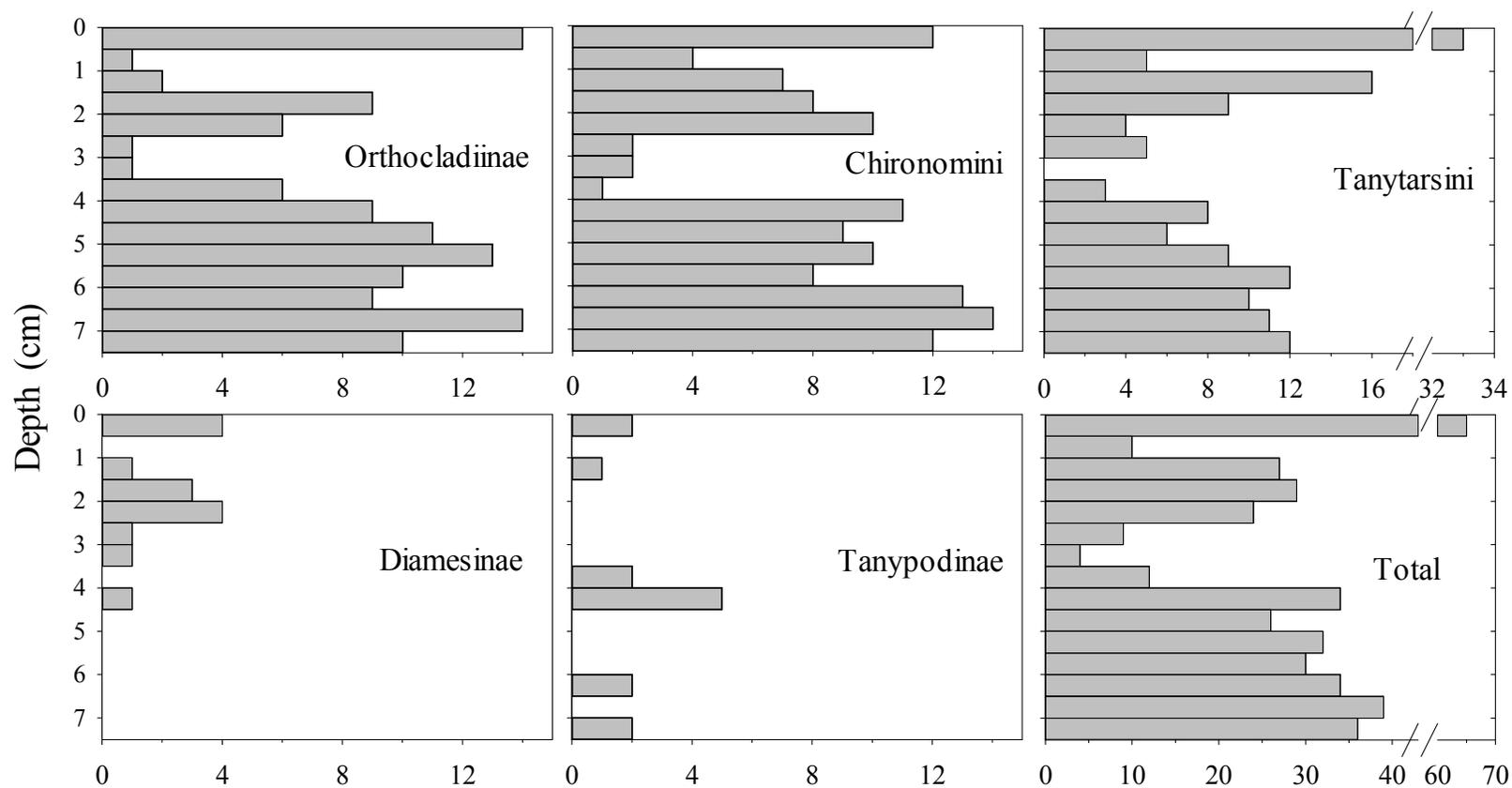


Figure 3.



Numbers of subfossil chironomid head capsules per depth interval

Figure 4.

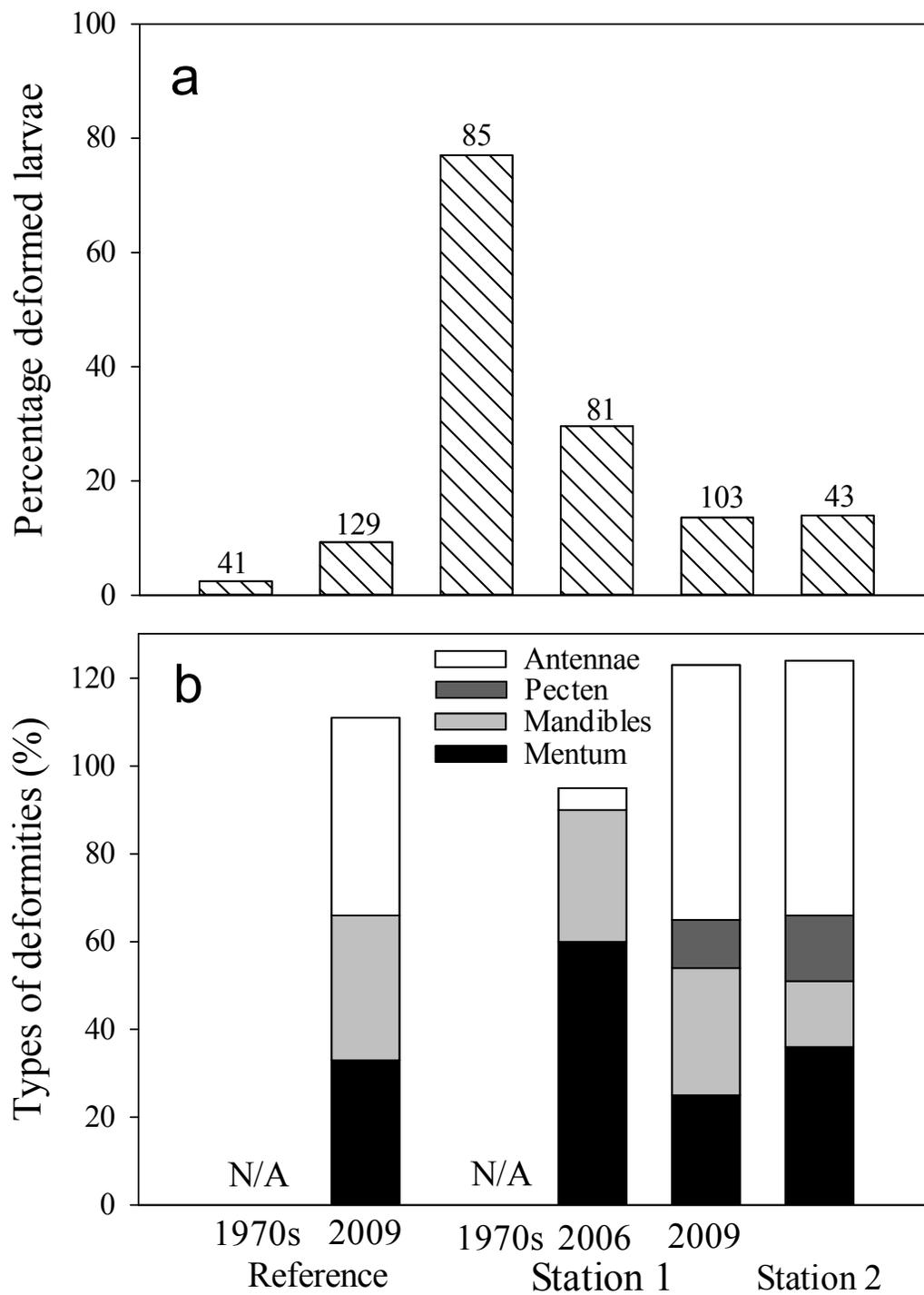


Figure 5.

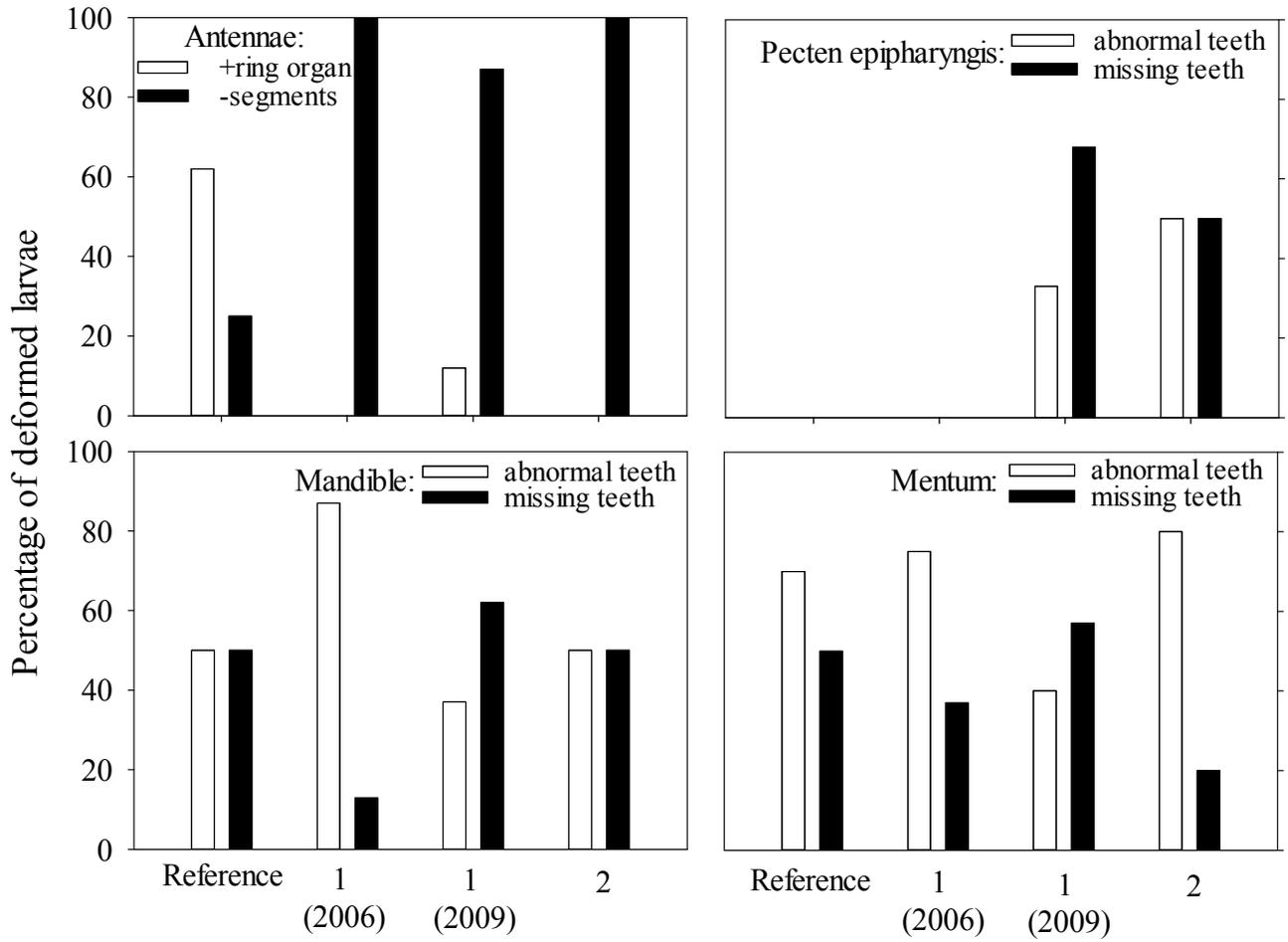


Figure 6.