Ultraviolet radiation affects invasibility of lake ecosystems by warmwater fish

Andrew J. Tucker[1], tuckera2@muohio.edu

Craig E. Williamson[1], craig.williamson@muohio.edu

Kevin C. Rose[1], rosekc@muohio.edu

James T. Oris[1], orisjt@muohio.edu

Sandra J. Connelly[2], sandra.connelly@rit.edu

Mark H. Olson[3], mark.olson@fandm.edu

David L. Mitchell[4], dmitchel@mdanderson.org

[1] *Dept of Zoology, Miami University Oxford, OH 45056*

[2]*Dept of Biological Sciences, Rochester Institute of Technology, Rochester NY 14623*

[3]*Dept of Biology, Franklin and Marshall College, Lancaster, PA 17604*

[4]*The University of Texas MD Anderson Cancer Center, Smithville, TX 78957*

Submitted as an *Article*, 28 manuscript pgs.

Tables: 1

Figures: 3

Correspondence should be addressed to:

Andrew Tucker, 212 Pearson Hall, Oxford, OH 45056

Ph: 484.686.8408, fax: 513.529.6900, email: tuckera2@muohio.edu

Abstract

Predicting where species invasions will occur remains a substantial challenge in ecology, but identifying factors that ultimately constrain the distribution of potential invaders could facilitate successful prediction. Whereas ultraviolet radiation (UVR) is recognized as an important factor controlling species distribution and community composition, the role of UVR in a habitat invasibility context has not been explored. Here we examine how underwater UVR can regulate warmwater fish invasion. In Lake Tahoe CA/NV established populations of exotic bluegill sunfish (*Lepomis macrochirus*) are currently limited to turbid, low UVR embayments. An *in situ* incubation experiment that manipulated incident UVR exposure of larval bluegill, combined with an assessment of UVR exposure levels in nearshore habitats around Lake Tahoe, demonstrates that UVR can mediate habitat invasibility. Our findings suggest that the susceptibility to invasion by UVR sensitive species may increase in transparent aquatic systems threatened by declining water quality, and they highlight the importance of abiotic factors as regulators of invasion risk in ecosystems.

Keywords: ultraviolet radiation, habitat invasibility, aquatic invasion, abiotic factors, DNA dosimeters

INTRODUCTION

The proliferation of invasive species is one of the most important anthropogenic impacts in freshwater systems (Naiman et al. 1995). The problem is largely a byproduct of human development, with its tendency to deconstruct biogeographic barriers (Rahel 2007) and fundamentally alter the biotic and abiotic components of environments that foster distinct populations of plants and animals and regulate the susceptibility of habitats to invasion. Consequently, habitat invasibility is generally thought to be high in areas characterized by extensive human impact. For example, among California, USA watersheds the number of non-native fish species is positively correlated with anthropogenic landscape-level changes related to watershed disturbance and altered hydrology (Marchetti et al. 2004). Reservoirs are also a notable example of how human activity may promote invasion (Havel et al. 2005). These examples highlight important factors that are likely to control invasibility in some habitats but they are driven by more traditional notions of human impact, such as the stabilization of flow regimes related to habitat alteration or the influence of a high degree of environmental variability through time. Whereas changes in water transparency with anthropogenic disturbance are widely recognized in aquatic habitats, little attention is given to how such disturbances can mediate exposure to damaging wavelengths of ultraviolet radiation (UVR). Here we demonstrate the potential importance of UVR exposure as a factor controlling habitat invasibility of a warmwater fish in Lake Tahoe.

Lake Tahoe is a sub-alpine lake in the northern Sierra Nevada range spanning the California/Nevada border. The lake is renowned for its deep blue water and high transparency, afforded by the combination of great depth, small watershed to lake area ratio, and granitic

71 geology of the basin (Jassby et al. 1994). However, the transparency has decreased over time
72 with the average annual Secchi transparency declining from 31 m in 1968 to 21 m by 1998
73 (Jassby et al. 1999). During this same thirty year interval a number of non-native warmwater
74 fish species established populations in some portions of Lake Tahoe (Reuter and Miller 2007).

75 The establishment of these warmwater species may be directly related to the significant
76 changes in water transparency observed in recent decades. For example, larval bluegill sunfish
77 (*Lepomis macrochirus*) perish within a single day when exposed to incident UVR at the surface
78 of transparent lakes (Williamson et al. 1999). Yet the requirement for warmer spawning
79 temperatures constrains bluegill nests to the shallow surface waters in the littoral zone of lakes
80 and rivers (Kitchell et al. 1974). Thus the transparency of the water as well as the depth and
81 location of nests are critical determinants of reproductive success in bluegill (Olson et al. 2006).
82 Currently, the only well-established bluegill populations in Lake Tahoe are limited to sites in the
83 southern end of the basin characterized by extensive development and in close proximity to some
84 of the lake's largest tributaries (Kamerath et al. 2008). Water transparency at these sites is low
85 and may explain their suitability for the UVR-sensitive bluegill. Our primary aim was to
86 explicitly test the hypothesis that UVR controls the suitability of nearshore habitats for the
87 earliest life history stages of exotic bluegill.

88 We were also interested in understanding what controls the UVR transparency of nearshore
89 habitats in Lake Tahoe. The decline in visible light transparency has been attributed to increases
90 in both biological (*i.e.*, phytoplankton and detritus) and inorganic (*i.e.*, terrestrial sediment)
91 particulate matter (Swift et al. 2006) resulting largely from human impacts in and around the
92 basin related to eutrophication (Goldman 1988) and stream bank erosion (Byron and Goldman

1989). However, the attenuation of UVR in freshwater lakes is strongly regulated by chromophoric dissolved organic matter (CDOM) (Morris et al. 1995, Williamson et al. 1996). CDOM may be especially important in nearshore habitats where fish spawning occurs, since CDOM inputs are likely to be concentrated in those areas. For example, in Lake Tahoe stream water inputs of CDOM are approximately 10x higher than CDOM levels offshore where most of the long term transparency monitoring has been conducted (Swift 2004). An understanding of the mechanisms underlying UVR transparency in Lake Tahoe could enable us to better understand how regional and global environmental changes related to the factors that mediate UVR transparency could in turn affect habitat invasibility in this large, highly transparent lake.

METHODS

To test the hypothesis that UVR controls the suitability of nearshore habitats for bluegill invasion we measured UVR exposure at multiple nearshore locations around the perimeter of the lake using DNA dosimeters (Fig. 1). In two of these nearshore locations, we carried out a four day *in situ* incubation experiment that manipulated the incident UVR levels to which larval bluegill were exposed. DNA dosimeters were also deployed with the larval bluegill in these *in situ* incubations as a means for comparing levels of DNA damage in dosimeters with larval bluegill mortality. Standardized DNA damage values obtained from dosimeters incubated alone around the lake were compared to DNA damage values from dosimeters included with larval bluegill to evaluate the potential for larvae to survive in multiple nearshore locations. To assess the relative importance of dissolved organic carbon and chlorophyll as regulators of the UVR environment in nearshore areas of Lake Tahoe we measured levels of these light attenuating components at 13 nearshore sites, including each of the sites where we deployed dosimeters.

Larval Incubation Experiment

Larval yolk sac bluegill were collected from a single nest at approximately 1 m depth in the Tahoe Keys on 17 July 2007. Larvae (n= 5) were placed in Whirl-Pak bags filled with 100 mL of 48 μm filtered lake water to exclude zooplankton. To isolate the effect of UVR between incubation sites, the Whirl-Pak bags were either shielded from incident UVR in Courtgard (CP Films, Inc, Martinsville, VA, USA; http://www.cpfilms.com/) sleeves or exposed to incident UVR in Aclar (Honeywell International, Morristown, NJ, USA; http://www.honeywell.com/) sleeves. Courtgard is a long-wave-pass plastic that transmits PAR (95% 400–800 nm in water) but blocks most UVR (transmits no UV-B 295–319 nm, and only 9% of UV-A 320–400 nm with a sharp wavelength cutoff and 50% transmittance at 400 nm). Aclar is a long-wave-pass plastic that in water transmits both photosynthetically active radiation (PAR) (100% 400–800 nm) and most UVR (98% of UV-B 295–319 nm, 99% UV-A 320–399 nm, with a sharp wavelength cutoff and 50% transmittance at 212 nm). The two incubation sites for the larval exposure experiment included waters with low and high UVR transparencies, that is, the Tahoe Keys and Sand Harbor areas, respectively. Four replicates of each of the UVR shielded and unshielded treatments were deployed at dusk on 17 July at one meter depth in both the high (Sand Harbor) and low (Tahoe Keys) UVR sites and retrieved early on the morning of 21 July. After collection, larvae were examined under a dissecting microscope and scored as live if a heartbeat was observed. The four-day incubation period used here is similar to the time it takes larvae to reach swim-up stage and leave the nest (Gross and MacMillan 1981). Average daily water temperature at 1 meter in both sites was within threshold temperatures for bluegill spawning. All procedures involving animals were in accordance with the policies set forth by Miami University's Institutional Animal Care and Use Committee (IACUC protocol #683).

Both incident and submersible radiometers and DNA dosimeters were deployed to measure incident UVR and water transparency during the incubation. Underwater solar radiation was measured at each site with a BIC profiling UVR-PAR radiometer (Biospherical Instruments, Inc, San Diego, CA, USA; http://www.biospherical.com/). This instrument quantifies incident solar irradiance at three different UVR wavelengths (305, 320, and 380 nm) as well as visible wavelengths of photosynthetically active radiation (PAR, 400-700 nm). Transparency data from BIC profiles were combined with cumulative surface irradiance data measured with a Biospherical Insruments BICLogger, a multichannel, internally recording radiometer of a similar design and specifications to the BIC, to estimate total exposure for the duration of the incubation experiment. Two DNA dosimeters were included with fish in each of the four UVR unshielded bags and in two of the four UVR shielded bags at each site during larval fish incubations.

Logistic regression analysis of a 2^3 factorial design was performed using SAS v. 9.1 to test for main effects of site, UVR+ or UVR- microcosm, and species on larval survival. Larval largemouth bass were also incubated but are not discussed here because of limited replication.

DNA dosimeters

The DNA dosimeters were 10 mm diameter by 40 mm long quartz cuvettes filled with 0.4 ml of raw salmon testes DNA solution diluted to 100 µg/mL in double distilled water and sealed on each end with silicone stoppers and parafilm. DNA in dosimeters accumulates damage as a function of UVR exposure, and the frequency of cyclobutane pyrimidine dimers (CPD's) per mb of DNA, the most common photoproduct, were estimated using radioimmunoassay (RIA) as described by Mitchell (Mitchell 1996, Mitchell 1999).

Dosimeters (n=2) alone were deployed for approximately four days during the week of 15 July to 20 July 2007 at 1 and 2 m depths at ten sites around Lake Tahoe, including the two larval incubation sites. Dosimeters were placed in Whirl-Pak bags filled with 100 mL of 48 μm filtered lake water and then inserted into Aclar sleeves. The DNA damage values obtained from dosimeters incubated alone over four day periods were standardized for depth and deployment duration for comparison to DNA damage values from dosimeters that were included in the larval fish incubations. Standardization was accomplished by estimating UVR 320 nm exposure for a depth (1m) and a time (2.5 exposure days) equivalent to that of the larval fish incubations, and then estimating a DNA damage value (CPDs / MB DNA) from a DNA damage versus 320 nm exposure relationship derived from the dosimeter and exposure data collected over the one week period from 15 July to 20 July 2007.

DOC and Chlorophyll a analysis

Water samples were collected in pre-rinsed 1 L polyethylene bottles from within the mixed layer. Water used in DOC analysis was filtered through pre-ashed 25 mm 0.7 μm Whatmann GFF filters within 8 hours of sample collection using a glass frit. The filtered sample was stored in the cold and dark in 40ml glass bottles until analysis. The DOC samples were analyzed with a Shimadzu TOC- V_{CPH} analyzer within one week post sampling. For chlorophyll *a*, 100 ml of the water sample was filtered through pre-ashed 25mm 0.7μm Whatmann GFF filters within 8 hours of collection and the filter was immediately frozen until chlorophyll analysis. Chlorophyll *a* extraction was completed with an acetone-methanol mixture and chlorophyll *a* concentration was completed via fluorometry within one month of sample collection. UVR attenuation was also measured at each site with the BIC profiling radiometer, and diffuse attenuation coefficients (K_d) were calculated for each site from the slope of the natural log relationship of UVR irradiance

versus depth. Using SAS v. 9.1 we performed a likelihood ratio test to compare models that predicted $K_{d\ 320nm}$ from DOC and/or Chl *a* values.

RESULTS

For the larval incubation experiment, exposure to 320 nm radiation in unshielded treatments was nearly 40x higher in the Sand Harbor site (22.65 kJ m^{-2}) compared to the Tahoe Keys site (0.60 kJ m^{-2}, Fig. 2A). The mean DNA damage levels, measured in DNA dosimeters as the frequency of cyclobutane pyrimidine dimers (CPDs), at the Sand Harbor site were more than 30x higher than those measured at the Tahoe Keys site (729 vs 22 CPDs/mb DNA, Fig. 2B). Larval survival was inversely related to UVR exposure with 84% of larvae surviving in unshielded microcosms in the low UVR site and only 11% survival in the high UVR site (Fig. 2C). For bluegill in unshielded UVR microcosms, there was a statistically significant effect of site on larval fish survival (PROC LOGISTIC; $p<0.0001$). In the UVR-shielded microcosms, larval survival was high (90-100%) at both sites. DNA damage measured in the dosimeters also increased with increasing UVR transparency across the ten sample sites (Fig. 3). In 7 of the 10 sample sites DNA damage levels were higher than those measured at the Tahoe Keys, where bluegill survival was high. Indeed, the majority of sites showed DNA damage levels above the threshold for larval survival (Figs. 2B and 3), implying high potential UVR-induced mortality in bluegill at most sample sites.

The 1 % attenuation depths, that is the depth where 320 nm UVR reaches 1% of surface irradiance, show the wide range of UVR transparency of nearshore sites in Lake Tahoe (Table 1). UVR (320 nm) transparency of the near shore sites was strongly dependent upon DOC (K_{d320nm} = (b0*DOC^b1); R^2 = 0.81). However, a model that included both DOC and chlorophyll

204 *a* (K_{d320nm} = (b0*DOC^b1) + (b2*Chl)) was the best predictor of UVR attenuation (R^2= .98) for
205 the sites sampled (LR x2=11.2, df=1, p=.0008).

206 DISCUSSION

207 In this study, dosimeters of raw DNA in solution were used as tools to assess potential
208 UVR effects on larval bluegill by relating DNA damage levels in dosimeters with larval fish
209 mortality. The observed levels of DNA damage in the dosimeters support the hypothesis that
210 UVR is a potent force contributing to the suitability of nearshore habitats for successful bluegill
211 reproduction. Current UVR conditions were substantial enough to reduce reproductive success
212 of bluegill in the majority of nearshore sites sampled. Both DOC and chlorophyll *a* were
213 important regulators of variation in the UVR environment in nearshore areas of Lake Tahoe.
214 This suggests that effective regulation of chlorophyll and DOC inputs could stem future declines
215 in UVR transparency in Lake Tahoe and in turn help mediate habitat invasibility.

216 Our study was motivated by a framework for predicting species invasion that highlights the
217 importance of identifying the specific abiotic factors that will ultimately constrain distribution in
218 an invaded range. Current approaches for predicting habitat invasion tend to rely on correlating
219 species' distribution with selected habitat parameters that implicitly incorporate biotic constraints
220 on distribution. These biotic constraints may not always be present in an uninvaded range
221 (Kearney and Porter 2004). It has been argued that a more powerful approach is to identify
222 specific abiotic factors with demonstrable fitness consequences for an organism, and then map
223 the fitness consequences (*e.g.*, survival or reproduction) at various levels of the abiotic factor
224 onto the landscape (Kearney 2006). This kind of approach is fundamental if we wish to improve
225 our confidence in extrapolating species' potential distributions to novel circumstances under

226 climate change scenarios, and it could be especially useful for predicting invasions in systems
227 where a specific factor regulating invasion (*e.g.*, UVR) is closely tied to a global change element
228 (*e.g.*, climate driven changes in DOC). In our study we have accomplished the crucial first step
229 in this approach by demonstrating that UVR is a key abiotic factor with the potential to constrain
230 the reproductive success of bluegill in Lake Tahoe. By identifying some of the key mechanisms
231 underlying UVR transparency we have also increased our understanding of how regional and
232 global environmental changes related to the factors that mediate UVR transparency could in turn
233 affect habitat invasibility in this lake. We suspect that this framework and our results could be
234 directly relevant to other transparent lakes.

235 Whereas few lakes are as highly transparent as Lake Tahoe, estimates from DOC
236 measurements in North American lakes indicate that UVR transparency is relatively high
237 throughout western, northwestern, and southeastern portions of the USA (Williamson et al.
238 1996). For example, based on modelling the relationship between DOC concentration and UVR
239 attenuation, the depth to which 1% of 320 nm UVR surface irradiance penetrates is greater than
240 1 m in 75% of lakes sampled in the western USA. About 25% of these lakes exhibit 1% UVR
241 depths greater than 4.75 m. This is noteworthy because bluegill generally nest at depths less than
242 4 m (Carlander 1977), and other studies have demonstrated significant UVR effects on
243 reproductive success of temperate fish species (including bluegill) in the eastern USA in lakes
244 with a 1% UVR depth not in excess of 4.9 m (Huff et al. 2004, Olson et al. 2006).

245 The DOC concentration in most of the transparent lakes referenced above is quite low (*i.e.*
246 < 1 mg/L), suggesting that even small changes in DOC could significantly reduce current UVR
247 levels in these lakes (Williamson et al. 1996). Although there are no specific predictions for

248 future DOC concentrations in western and southeastern US lakes, widespread and strong trends
249 of generally increasing DOC concentrations have been observed in lakes and rivers elsewhere
250 (Evans et al. 2005, Monteith et al. 2007). Therefore it is reasonable to consider the potential for
251 substantial changes in UVR transparency, and consequently habitat invasibility in these
252 transparent lakes. Just as relevant and better documented in high elevation transparent lakes like
253 Tahoe are trends of increased algal growth and reduced water clarity as a consequence of
254 increased nitrogen deposition (Jassby et al. 1994, Jassby et al. 1995, Sickman et al. 2003). These
255 trends, documented in the western USA, are predicted to continue across that region (Lamarque
256 et al. 2005). Chlorophyll has a proportionately greater effect on UVR attenuation in low DOC
257 systems (Laurion 2000, Sommaruga and Augustin 2006). Consequently variations in
258 chlorophyll levels, like changing DOC concentrations, have the potential to modify transparency
259 in very low DOC lakes. This in turn could facilitate the establishment of exotic species in
260 formerly unsuitable habitats.

261 One critical question pertinent to the role of UVR in mediating habitat invasibility in
262 transparent lakes is whether adult bluegill are able to respond to these selective pressures by
263 reducing UVR exposure through either nesting deeper or shifting their spawning time to coincide
264 with periods of decreased water transparency. For Lake Tahoe, this seems an unlikely
265 possibility. First, in this study the 1 m depth, the seasonal timing of our experiments, and the 4
266 day duration of the incubation were consistent with actual nest depths and nesting times in Lake
267 Tahoe. Moreover, later spawning times, coincident with increasing water temperatures that
268 might allow bluegill to nest at greater depths, are unlikely to decrease UVR exposure because
269 UVR transparency (320 nm) actually increases on the order of 20 – 90% from spring to summer
270 as allochthonous inputs decrease in the nearshore (Rose et al. in press). On the other hand

accelerated spawning phenologies that could potentially enable bluegill to benefit from decreased water transparency earlier in the growing season are likely constrained by thermal conditions required for spawning. Bluegill are reported to spawn at temperatures from 15.6° C to 32° C, with optimum spawning temperatures in the range of 21° C to 24°C (Wallus and Simon 2008). Surface water temperatures measured at an index site in May 2007 and 2008 never exceeded 11.1° C, well below the minimum spawning temperature. Even in June the maximum surface water temperature over this two year period was 17.2° C (unpublished data), still below the optimal spawning temperature for bluegill. Thus the primary opportunity for invasion is likely to be in the shallow nearshore embayments where both water temperatures are high enough and transparency to UVR is low enough to permit adult spawning and larval survival.

It is also important to note that we have used the most severe response metric (*i.e.* mortality) in evaluating the role of UVR for regulating habitat suitability for larval bluegill invasion. Consequently, our study likely underestimates the full extent of UVR induced effects on larvae when considered in terms of the interactions of sub-lethal effects with sources of background mortality in developing larvae and other 'life history bottlenecks' that young fish face. For example, UVR exposure impedes larval growth in a variety of fish species (Hunter et al. 1979, Winckler and Fidhiany 1996, Vehniainen et al. 2007) and body size in young fish, including bluegill, is a critical determinant of over-winter survivorship and mortality due to predation (Cargnelli and Gross 1996, Belk and Hales 1993). Other potential sub-lethal UVR effects that may ultimately reduce bluegill survival include diminished immune system function and increased incidence of infectious disease resulting from 'sunburn' (Salo et al. 1998, Novak 1999), developmental anomalies that might increase susceptibility to predators (Vehniainen et al. 2007), indirect trophic mediated UVR effects on food availability (Williamson et al. 1994,

294 Zagarese and Williamson 2001), or phototoxic effects (Bullock and Roberts 1979, Oris and
295 Geisy 1987).

296 It is unclear to what extent UVR may play a role in the invasion ecology of other invasive
297 species or other life history stages. We contend that it could have relevance for any UVR
298 sensitive species that is constrained to shallow water environments by, *e.g.*, requirements for
299 warmer spawning temperatures in clear, cold-water lakes. For older more tolerant and mobile
300 life history stages other biotic and/or abiotic factors (*e.g.* food availability or habitat structure)
301 likely play a more important role in determining habitat suitability. However, we have
302 emphasized the earliest life history stages here for two reasons. First, early life history stages are
303 less pigmented, less mobile, and thus likely to be less well-defended against UV damage.
304 Second, in a biological invasion context the naturalization and eventual invasion of a species in a
305 novel environment depends critically on the ability of that species to establish self-perpetuating
306 populations through successful reproduction (Richardson et al. 2000). Whereas other among-
307 habitat characteristics may be important in regulating species invasions, we have shown that for
308 these critical early life history stages UVR alone is an adequate determinant of habitat suitability
309 and thus a potential regulator of habitat invasibility.

310 The extent to which UVR ultimately controls bluegill invasion in Lake Tahoe or any other
311 system will depend upon the potential for these organisms to adapt to local conditions. It is
312 possible for example, that constitutive levels of maternally derived photoprotective compounds
313 (PPCs) could increase in larval fish spawned in high UVR environments, thereby increasing
314 UVR tolerance and the ability to spread into new habitats. High UV environments tend to
315 stimulate PPC synthesis by algae and bacteria. These can be transferred in food chains and

316 accumulated at higher trophic levels by organisms that have such capability, which may in turn
317 be enhanced by UV exposure (e.g., copepods (Tartarotti et al. 2004, Moeller et al. 2005,
318 Tartarotti and Sommaruga 2006); coral reef fish (Zamzow 2004)).

319 Environmental stress is often considered a driver of adaptation during invasion and it has
320 been demonstrated that abiotic conditions can select for adaptive genotypes in invasive species
321 (Lee et al. 2007). Our data suggest that UVR can similarly act as a selective force in highly
322 transparent systems, and the potential for the development of more resistant genotypes could be
323 tested. Future research concerning the role of UVR in controlling biological invasion should
324 consider these and other possibilities. Nevertheless, we have shown that for the current bluegill
325 population in Lake Tahoe UVR is a potent stressor that mediates habitat suitability for larval fish
326 in nearshore areas and therefore controls habitat invasibility.

327 Further efforts to quantify the effect of abiotic controls on the growth, survival, and
328 reproduction of organisms and to map those effects onto the landscape will help us to more
329 accurately predict the full potential of species invasion in imperiled environments. Knowledge of
330 the particular levels of important abiotic factors that reduce the fitness of non-natives could also
331 enable us to manage abiotic conditions in habitats for the prevention of species invasion (Alpert
332 et al. 2004). In lakes, for example, one goal might be to establish and manage UVR transparency
333 thresholds that prevent the establishment of non-native species by inhibiting successful
334 reproduction. We suggest that future studies in highly transparent aquatic ecosystems consider
335 UVR and other abiotic habitat features as important factors controlling habitat invasibility and
336 invasion risk.

337

Acknowledgements

We would like to thank Geoff Schladow, Monika Winder, Sudeep Chandra and Marcy Kamerath for logistical support. We are also grateful to Anne Liston and the staff of the Tahoe Environmental Research Center for their assistance. Thanks also to Neil Winn for GIS mapping assistance. This work was supported in part by Miami University's Field Workshop Program (field expenses), NSF IRCEB 0210972 (UV radiometers) and NIEHS Center grant ES07784 (DNA analysis).

Author contributions

D.L.M. performed the radioimmunoassay for DNA dosimeters. K.C.R. collected and analyzed data and deployed DNA dosimeters and larval fish. J.T.O. and S.J.C. deployed DNA dosimeters. A.J.T. collected and analyzed data, deployed DNA dosimeters and larval fish, and wrote the paper with C.E.W. M.H.O., A.J.T., C.E.W., J.T.O., S.J.C., and K.C.R. were involved in study design. All authors discussed the results and commented on the manuscript.

Literature Cited

Alpert, P., E. Bone, and C. Holzapfel. 2004. Invasiveness, invasibility and the role of environmental stress in the spread of non-native plants. Perspectives in Plant Ecology, Evolution and Systematics 3: 52-66.

Belk, M.C., and L.S. Hales. 1993. Predation-induced differences in growth and reproduction of bluegills (*Lepomis macrochirus*). Copeia 1993: 1034-1044.

Belzile, C., and W.F. Vincent. 2002. Contribution of absorption and scattering to the attenuation of UV and photosynthetically available radiation in Lake Biwa. Limnology and Oceanography 47:95-107.

Bullock, A.M., and R.J. Roberts. 1979. Indication of UDN like lesions in salmonids by exposure to ultraviolet light in the presence of phototoxic agents. Journal of Fish Diseases 2: 439-441.

Byron, E.R., and C.R. Goldman. 1989. Land use and water quality in tributary streams of Lake Tahoe, CA/NV. Journal of Environmental Quality 18: 84-88.

Cargnelli, L.M., and M.R. Gross. 1996. The temporal dimension in fish recruitment: birth date, body size, and size-dependent survival in a sunfish (bluegill: *Lepomis macrochirus*). Canadian Journal of Fisheries and Aquatic Sciences 53: 360-367.

Carlander, K.D. 1977. *Handbook of Freshwater Fishery Biology*. (The Iowa State University Press, Ames, Iowa).

Evans, C.D., D.T. Monteith, and D.M. Cooper. 2005. Long-term increases in surface water dissolved organic carbon: Observations, possible causes and environmental impacts. Environmental Pollution 137: 55–71.

Goldman, C.R. 1988. Primary productivity, nutrients, and transparency during the early onset of eutrophication in ultra-oligotrophic Lake Tahoe, CA/NV. Limnology and Oceanography 33: 1321-1333.

Gross, M.R., and A.M. MacMillan. 1981. Predation and the evolution of colonial nesting in bluegill sunfish (*Lepomis macrochirus*). Behavioral Ecology and Sociobiology 8: 163-174.

Havel, J.E., C.E. Lee, and M.J. VanderZanden. 2005. Do reservoirs facilitate invasions into landscapes? BioScience 55: 518-525.

Huff, D.D., G. Grad, and C.E. Williamson. 2004. Environmental constraints on spawning depth of yellow perch: The roles of low temperature and high solar ultraviolet radiation. Transactions of the American Fisheries Society 133: 718-726.

Hunter, J.R., J.H. Taylor, and H.G. Moser. 1979. Effect of ultraviolet radiation on eggs and larvae of the northern anchovy, *Engraulis mordax*, and the Pacific mackerel, *Scomber japonicas*, during the embryonic stage. Photochemistry and Photobiology 29: 325-338.

Jassby, A.D., C.R. Goldman, and J.E. Reuter. 1995. Long-term change in Lake Tahoe (California-Nevada, U.S.A.) and its relation to atmospheric deposition of algal nutrients. Archiv fuer Hydrobiologie 135: 1-21.

Jassby, A.D., C.R. Goldman, J.E. Reuter, and R.C. Richards. 1999. Origins and scale dependence of temporal variability in the transparency of Lake Tahoe, California-Nevada. Limnology and Oceanography 44: 282-294.

Jassby, A.D., J.E. Reuter, R.P. Axler, C.R. Goldman, and S.H. Hackley. 1994. Atmospheric desposition of nitrogen and phosphorus in the annual nutrient load of Lake Tahoe (California-Nevada). Water Resources Research 30: 2207-2216.

Kamerath, M., S. Chandra, and B.C. Allen. 2008. Distribution and impacts of warm water invasive fish in Lake Tahoe, USA. Aquatic Invasions 3: 35-41.

Kearney, M. 2006. Habitat, environment, and niche: what are we modelling? Oikos 115: 186-191.

Kearney, M., and W.P. Porter. 2004. Mapping the fundamental niche: physiology, climate, and the distribution of a nocturnal lizard. Ecology 85: 3119-3131.

Kitchell, J.F., *et al.* 1974. Model of fish biomass dynamics. Transactions of the American Fisheries Society 103: 786-798.

Lamarque, J.F., *et al.* 2005. Assessing future nitrogen deposition and carbon cycle feedback using a multi-model approach. Part 1. Analysis of nitrogen deposition. Journal of Geophysical Research 110.

Laurion, I., M. Ventura, J. Catalan, R. Psenner, and R. Sommaruga. 2000. Attenuation of ultraviolet radiation in mountain lakes: factors controlling the among- and within-lake variability. Limnology and Oceanography 45: 1274-1288.

Lee, C.E., J.L. Rmefert, and Y-M Chang. 2007. Response to selection and evolvability of invasive populations. Genetica 129:179-192.

Marchetti, M.P., T. Light, P.B. Moyle, and J.H. Viers. 2004. Fish invasions in CA watersheds: testing hypotheses using landscape patterns. Ecological Applications 14: 1507-1525.

Mitchell, D.L. 1996 in *Technologies for Detection of DNA Damage and Mutations*, ed Pfeifer G (Plenum, New York, New York), pp 73-83.

Mitchell, D.L. 1999 in *Methods in Molecular Biology: DNA Repair Protocols*, ed Henderson DS (Humana, Totowa, New Jersey), pp 165-175.

Moeller, R.E., S. Gilroy, C.E. Wiliamson, G. Grad, and R. Sommaruga. 2005. Dietary acquisition of photoprotective compounds (mycosporine-like amino acids, carotenoids) and acclimation to ultraviolet radiation in a freshwater copepod. Limnology and Oceanography 50: 427-439.

Monteith, D.T., *et al.* 2007. Dissolved organic carbon trends resulting from changes in atmospheric deposition chemistry. Nature 450: 537-541.

Morris, D.P., *et al.* 1995. The attenuation of solar UV radiation in lakes and the role dissolved organic carbon. Limnology and Oceanography 40: 1381-1391.

Naiman, R.J., J.J. Magnuson, D.M. McKnight, and J.A. Stanford. 1995. *The Freshwater Imperative: A Research Agenda*. (Island Press, Washington DC).

Nowak, B.F. 1999. Significance of environmental factors in aetiology of skin diseases of teleost fish. Bulletin of the European Association of Fish Pathologists 19: 290-292.

Olson, M.H., M.R. Colip, J.S. Gerlach, and D.L. Mitchell. 2006. Quantifying ultraviolet radiation mortality risk in bluegill larvae: effects of nest location. Ecological Applications 16: 328-338.

Oris, J.T., and J.P. Geisy. 1987. The photo-induced toxicity of polycyclic aromatic hydrocarbons to larvae of the fathead minnow (*Pimephales promelas*) Chemosphere 16: 1395-1404.

Rahel, F.J. 2007. Biogeographic barriers, connectivity and homogenization of freshwater faunas: it's a small world after all. Freshwater Biology 52: 696-710.

Reuter, J.E., and W.W. Miller. 2000 in *Lake Tahoe Watershed Assessment Volume II: Appendix J*, eds Murphy, D.D., and C.M. Knopp (U.S. Department of Agriculture-Forest Service, Pacific Southwest Research Station).

444 Richardson, D.M., *et al*. 2000. Naturalization and invasion of alien plants: concepts and
445 definitions. Diversity and Distributions 6: 93-107.

446 Rose, K.C., C.E. Williamson, SG. Schladow, M. Winder, and J.T. Oris. In press. Patterns of
447 spatial and temporal variability of UV transparency in Lake Tahoe, CA/NV. Journal of
448 Geophysical Research (in press)

449 Salo, H.M., M. Tuula, S. Aaltonen, E. Markkula, and I. Jokinen. 1998. Ultraviolet B irradiation
450 modulates the immune system of fish (*Rutilus rutilus*, Cyprinidae) I. Phagocytes. Photochemistry
451 and Photobiology 67: 433-437.

452 Sickman, J.O., J.M. Melack, and D.W. Clow. 2003. Evidence for nutrient enrichment of high
453 elevation lakes in the Sierra Nevada, California. Limnology and Oceanography 48: 1885-1892.

454 Smith, R.E.H., J.A. Furgal, M.N. Charlton, B.M. Greenberg, V. Hiriart, and C. Marwood. 1999.
455 Attenuation of ultraviolet radiation in a large lake with low dissolved organic matter
456 concentrations. Canadian Journal of Fisheries and Aquatic Sciences 56: 1351-1361.

457 Sommaruga, R., and G. Augustin. 2006. Seasonality in UV transparency of an alpine lake is
458 associated to changes in phytoplankton biomass. Aquatic Sciences- Research Across Boundaries
459 68: 129-141.

460 Swift, T.J. 2004, thesis, *The Aquatic Optics of Lake Tahoe CA-NV*. (University of California
461 Davis.

462 Swift, T.J., *et al.* 2006. Water clarity modeling in Lake Tahoe: Linking suspended matter
463 characteristics to Secchi depth. Aquatic Sciences 68: 1-15.

464 Tartarotti, B., G. Baffico, P. Temporetti, and H.E. Zagarese. 2004. Mycosporine-like amino
465 acids in planktonic organisms living under different UV exposure conditions in Patagonian lakes.
466 Journal of Plankton Research 26: 753-762.

467 Tartarotti, B., R. Sommaruga. 2006. Seasonal and ontogenetic changes of mycosporine-like
468 amino acids in planktonic organisms from an alpine lake. Limnology and Oceanography 51:
469 1530-1541.

470 Vehniainen, E.R., J.M. Hakkinen, and A.O.J. Oikari. 2007. Fluence rate or cumulative dose?
471 Vulnerability of larval northern pike (*Esox lucius*) to ultraviolet radiation. Photochemistry and
472 Photobiology 83: 444-449.

473 Wallus, R., and T.P. Simon. 2008. *Reproductive biology and early life history of fishes in the*
474 *Ohio river drainage.* (CRC Press, Boca Raton, Florida).

475 Williamson, C.E., B.R. Hargreaves, P.S. Orr, and P.A. Lovera. 1999. Does UV play a role in
476 changes in predation and zooplankton community structure in acidified lakes? Limnology and
477 Oceanography 44: 774-783.

478 Williamson, C.E., R.S. Stemberger, D.P. Morris, T.M. Frost, and S.G. Paulsen. 1996. Ultraviolet
479 radiation in North American lakes: Attenuation estimates from DOC measurements and
480 implications for plankton communities. Limnology and Oceanography 41: 1024-1034.

481 Williamson, C.E., H.E. Zagarese, P.C. Schulze, B.R. Hargreaves, and J. Seva. 1994. The impact
482 of short-term exposure to UV-B on zooplankton communities in north temperate lakes. Journal
483 of Plankton Research 16: 205-218.

484 Winckler, K., and L. Fidhinay. 1996. Significant influence of UVA on the general metabolism in
485 the growing ciclid fish, *Cichlasoma nigrofasciatum*. Journal of Photochemistry and Photobiology
486 33: 131-135.

487 Zagarese, H.E., and C.E. Williamson. 2001. The implications of solar UV radiation exposure for
488 fish and fisheries. Fish and Fisheries 2: 250-260.

489 Zamzow, J.P. 2004. Effects of diet, ultraviolet exposure, and gender on the ultraviolet
490 absorbance of fish mucus and ocular structures. Marine Biology 144: 1057-1064.

491

492

493

494

495

496

497

498

499

500

501

502

503

504

505 Table 1. Attenuation depths for 320 nm UVR.

506

Site Number	$Z_{1\%320nm}$ (m)	DOC (mg/L)	Chl *a* (µg/L)
1	0.4	1.77	2.47
2	1.3	1.24	12.20
3	1.1	1.00	144.70
5	8.6	0.66	0.95
6	14.0	0.53	1.81
7	17.8	0.58	0.42
8	18.6	0.58	0.58
9	28.8	0.53	0.32
10	30.3	0.51	0.52

507 508 509 510 511 512 513 514 515 516

517 $Z_{1\%\ 320nm}$, is the depth where 320 nm UVR reaches 1% of surface irradiance. Site numbers
518 correspond to those plotted in Figures 1 and 3. The 1% attenuation depths were estimated from
519 the diffuse attenuation coefficient K_d as: $Z_{1\%} = 4.605\ K_d^{-1}$ where $K_d(\lambda, z) = [\ln (I_0\ I_z^{-1}]\ Z^{-1}$, Z is
520 depth (m), and I is down-welling irradiance for a given wavelength (λ) at the surface (I_0) and at
521 depth Z (I_z). Also shown are dissolved organic carbon (DOC) and chlorophyll *a* values for each
522 of the near shore sample sites (excluding site 4 where only UVR data were collected).

523

524

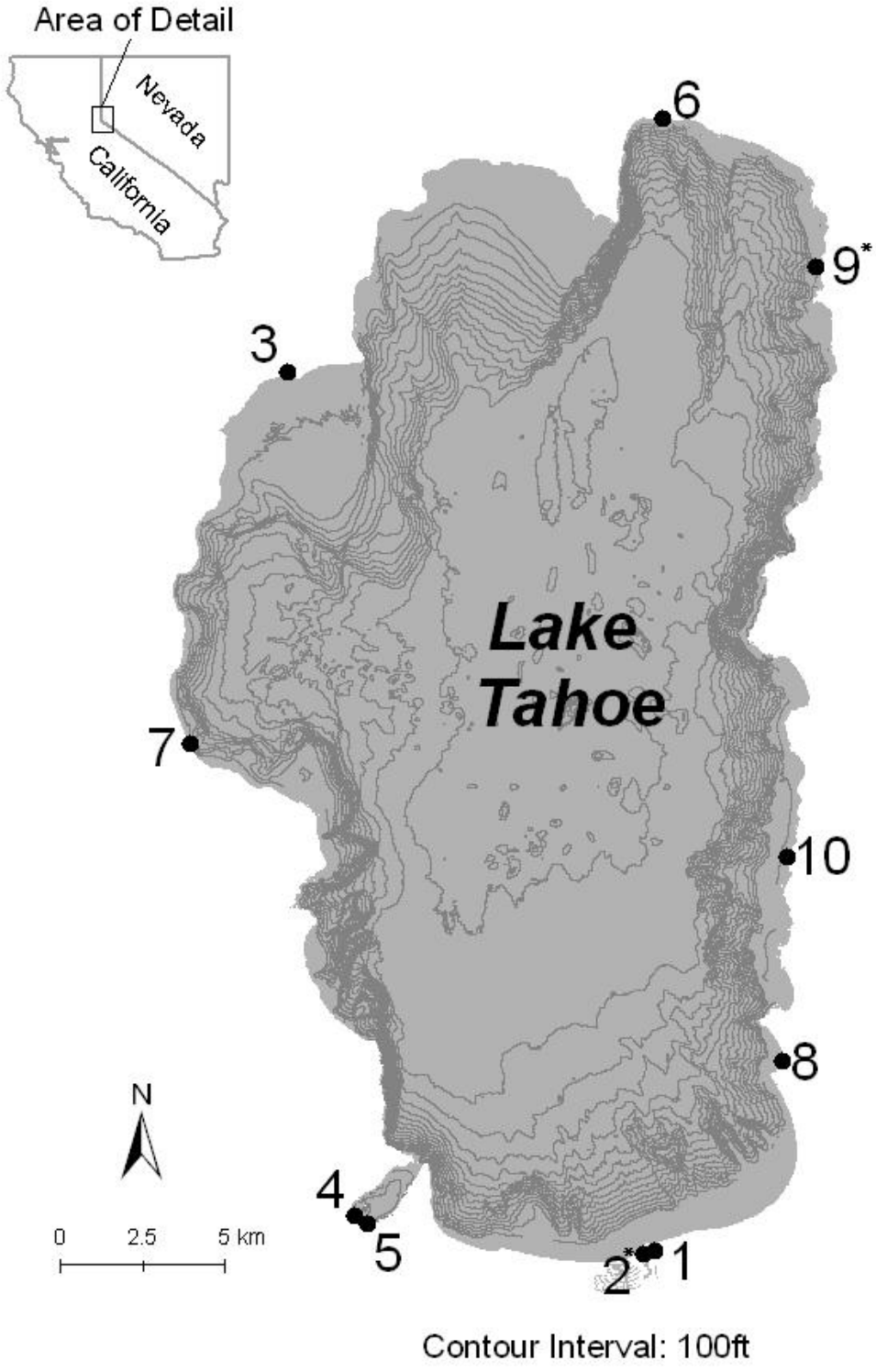
525

526

Figure 1. Map of Lake Tahoe indicating the location of nearshore sites where DNA dosimeters were deployed. Site numbers correspond to those plotted in Figure 3 and suggest that nearshore areas throughout Lake Tahoe are currently unsuitable for bluegill nesting due to their high UV transparency. Sites where larval incubations were deployed are indicated (*).

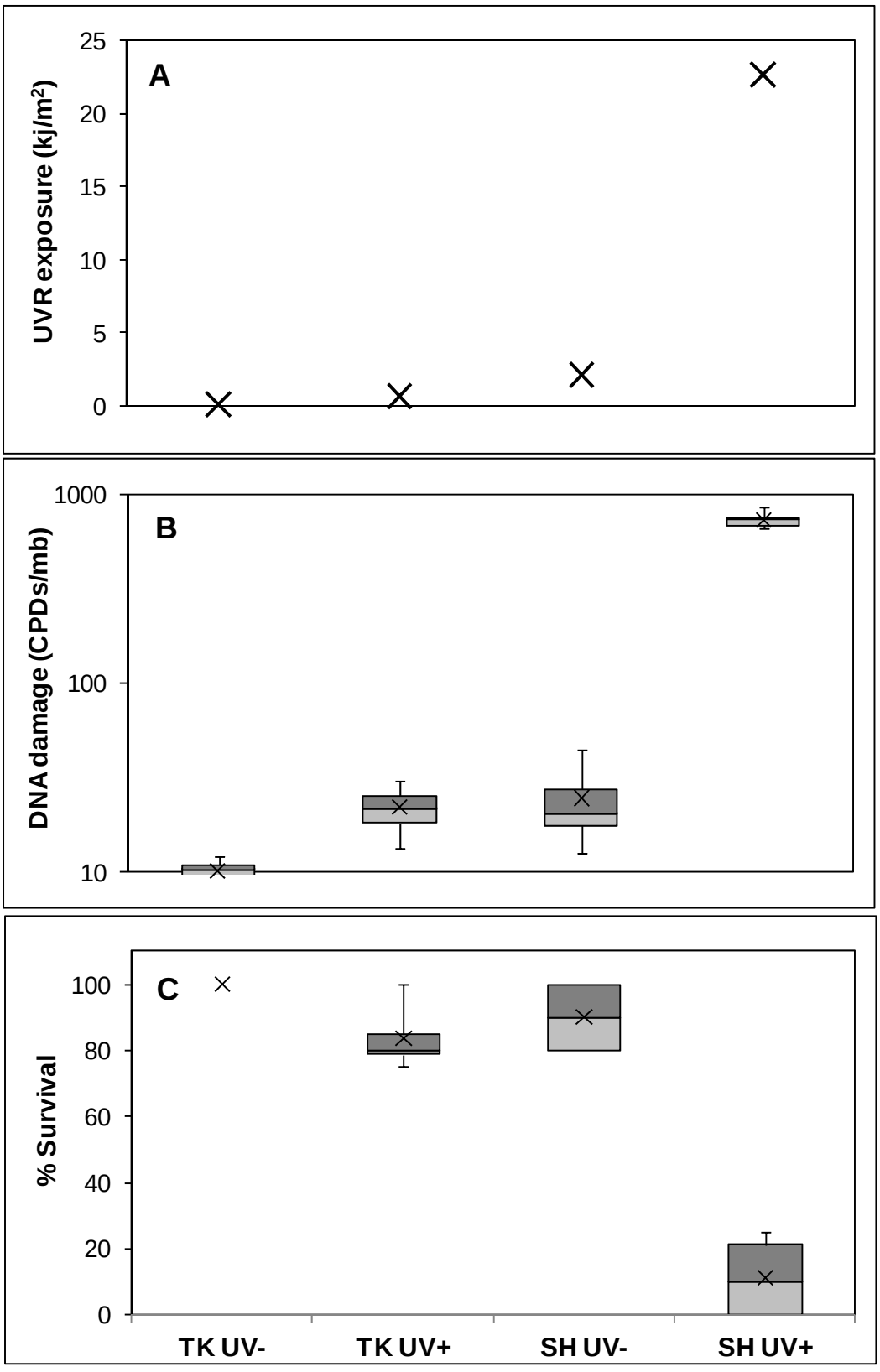
Figure 2. UVR exposure levels (A), DNA damage (B), and survival (C) of bluegill larvae in experimental microcosms. UVR exposure levels in the experimental microcosms were estimated from incident UVR measurements. DNA damage was measured in DNA dosimeters incubated in microcosms with larvae. Survival of bluegill larvae was measured after an 84 hour incubation in low (TK=Tahoe Keys) and high (SH=Sand Harbor) UVR sites when shielded (-) and unshielded (+) from incident UVR. Symbols are X for exposure estimates in panel A and mean values in panels B and C. Bars indicate maximum and minimum values within treatments. Boxes indicate the median and 25th and 75th percentiles.

Figure 3. Relationship showing the increase in the frequency of CPDs with increasing UVR transparency (indicated here by the depth to which 1% of incident surface 320 nm irradiance penetrates). DNA damage values are estimated from a derived exposure vs. dosimeter value relationship and are standardized for depth and deployment duration for comparison to larval fish incubation experiments. Site 2 is the low UVR Tahoe Keys. Site 9 is high UVR Sand Harbor.



550

551 Figure 1.



552

553 Figure 2.

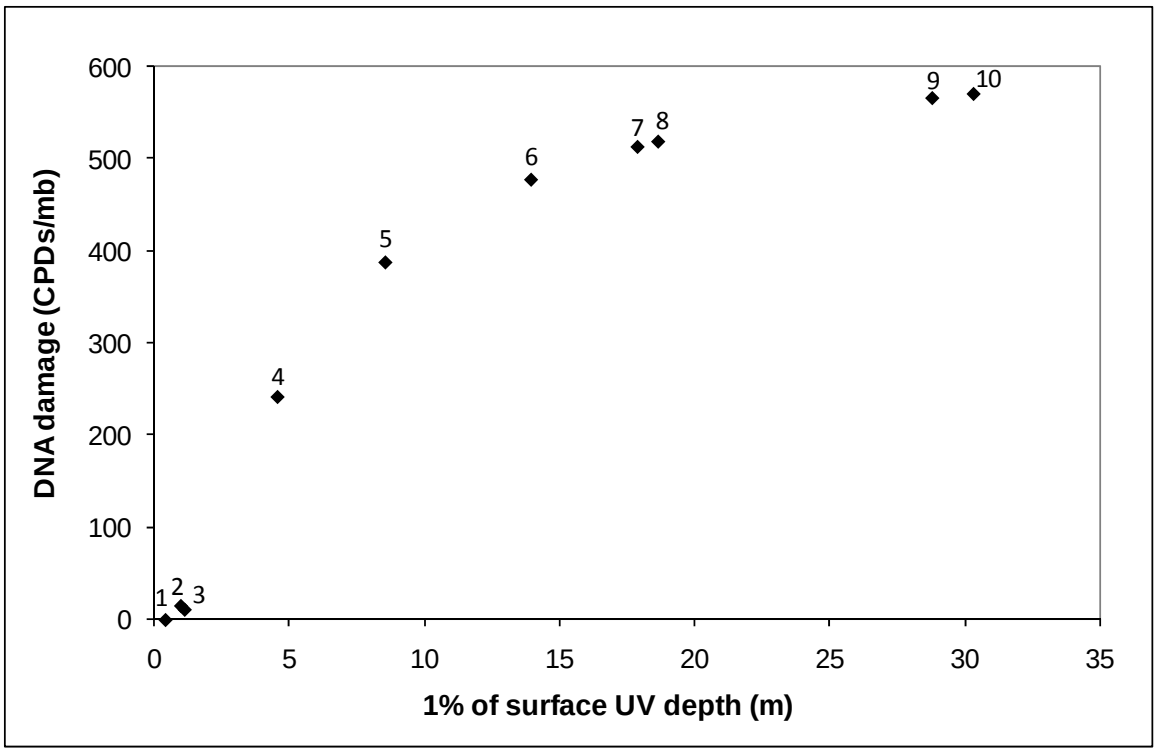

DNA damage (CPDs/mb)
600
500
400
300
200
100
0
1
2
3
4
5
6
7
8
9
10
0
5
10
15
20
25
30
35
1% of surface UV depth (m)

Figure 3.