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# Assessing multiple stressor effects to inform climate change management responses in three European catchments

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Bryan M. Spears<sup>1</sup>, Daniel Chapman<sup>1,2</sup>, Laurence Carvalho<sup>1</sup>, Katri Rankinen<sup>3</sup>,
 Konstantinos Stefanidis<sup>4,5</sup>, Stephen Ives<sup>1,6</sup>, Kristiina Vuorio<sup>3</sup>, Sebastian Birk<sup>7,8</sup>

- 8
- 9 1 UK Centre for Ecology & Hydrology, Penicuik, Midlothian, EH26 0QB, United Kingdom
- 10 2 Biological and Environmental Sciences, University of Stirling, Stirling, FK9 4LA, United Kingdom
- 11 3 Finnish Environment Institute, Latokartanonkaari 11, FI-00790 Helsinki, Finland
- 4 Hellenic Centre for Marine Research, Institute of Marine Biological Resources and Inland Waters,13 19013 Anavissos Attikis, Greece
- 14 5 Center for Hydrology and Informatics, National Technical University of Athens, 15780 Athina, Greece
- 15 6 Marine Scotland, Marine Laboratory, 375 Victoria Road, Aberdeen, AB11 9DH, United Kingdom
- 7 University of Duisburg-Essen, Faculty of Biology, Aquatic Ecology, Universitätsstraße 5, 45141
   Essen, 19 Germany 20

8 University of Duisburg-Essen, Centre for Water and Environmental Research, Universitätsstraße 5,
 45141 21 Essen, Germany

Author Contributions: All authors contributed to the original concept of this paper and to the development of the text.

**Corresponding author statement:** All correspondences should be sent to Bryan Spears, Centre for Ecology & Hydrology, Bush Estate, Penicuik, Midlothian, UK, EH260QB, email: <u>spear@ceh.ac.uk</u>; phone: +44 (0)131 445 8536

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#### 33 Abstract

Interactions between stressors, including climate change and nutrient enrichment, are 34 expected to be wide spread in firewater ecosystems, although the extent to which 35 these effects are locally moderated is not well understood. Our understanding of the 36 forms and frequency of occurrence of such interactions is limited; assessments using 37 field data have been constrained as a result of varying data forms and quality. To 38 address this, we demonstrate a statistical approach capable of assessing multiple 39 stressor interactions using contrasting data forms in three European catchments (Loch 40 Leven Catchment, UK: assessment of phytoplankton response in a single lake with 41 time series data, Pinios Catchment, Greece: macroinvertebrate response across 42 multiple rivers using spatio-temporal data; and Lepsämänjoki Catchment, Finland: 43 phytoplankton response across multiple rivers using spatio-temporal data). Statistical 44 models were developed to predict the relative and interaction effects of climate change 45 and nutrient enrichment sensitive indicators (stressors) on indicators of ecological 46 quality (ecological responses), within the framework of linear mixed effects models 47 (LMEs). In all catchments, indicators of nutrient enrichment were identified as the 48 primary stressor with climate change sensitive indicators causing secondary effects 49 (Loch Leven: additive, total phosphorus x precipitation; Pinios: additive, nitrate x 50 dissolved oxygen; Lepsämänjoki: synergistic, TP x summer water temperature), the 51 intensity of which varied between catchments and along the nutrient stressor gradient. 52 Simple, stressor change scenarios were constructed for each catchment and used, in 53 combination with mechanistic evidence support the models, to explore potential 54 55 management responses.

56

#### 57 **1. Introduction**

Fresh waters provide vital services to society including the provision of clean drinking 58 water, recreation and tourism, pollutant processing, biodiversity, food provision, and 59 energy (Reynard and Lanzanova, 2017). These services generally rely on good water 60 quality, underpinned by stable ecological processes, which are threatened globally by 61 multiple and potentially interacting stressors (Dodds et al., 2013), including nutrients, 62 hydrological modification, toxic chemicals, and non-native invasive species (Birk et al., 63 2019). Amongst these, climate change and nutrient stressors are expected to act 64 65 across large scales, although it has been proposed that management of nutrients may be achievable at local scales to relieve effects of both in fresh water ecosystems (Moss 66 et al., 2010). 67

It is clear that some stressors (e.g. nutrient enrichment) can be more easily 68 manipulated at the local scale than others (Friberg et al., 2016). For example, 69 catchment or ecosystem scale nutrient reduction has been demonstrated as an 70 effective approach in lake restoration (Jeppesen et al., 2005; Spears et al., 2016). In 71 contrast, other stressors acting on ecosystems but manifested from large-scale 72 drivers, such as climate change, may be impossible for catchment managers to 73 control. Stressors driven by anthropogenic activities operating at different scales can 74 also interact, for example, changes in temperature and flushing rate can alter 75 ecological responses to nutrient loading in rivers (Bowes et al., 2016) and lakes 76 (Carvalho et al., 2012). In fact, it is widely acknowledged that many mechanisms exist 77 through which the effects of climate change may be moderated in lakes and rivers 78 including geographically distinct projections in weather patterns, in the influence of 79 ecosystem morphology, and in the influence of other stressors, including nutrient 80 enrichment (Adrian et al., 2009). As a result, some authors have suggested that 81

disentangling these effects may be 'challenging, if not infeasible' (Benateau et al.,
2019).

Weyhenmeyer et al. (2007) provide empirical evidence on increasing rates of nitrate 84 depletion in European shallow lakes, suggesting that this phenomenon is driven by a 85 combination of decreased catchment and atmospheric nitrate loading as well as 86 87 increased denitrification related to warming between 1988 and 2003. Further, Moss et al. (2011) propose that such interactions will be wide spread, with local modifications 88 (e.g. in intensity of nutrient stress), resulting, generally, in exacerbation of 89 eutrophication effects including more severe and frequent algal blooms. It is, therefore, 90 important that such interactions be confirmed at a relevant scale of interest to support 91 the development of novel multiple-stressor management strategies. 92

93 We currently lack robust statistical frameworks to detect and predict the effects of multiple stressor mitigation options at catchment scales. Such a framework would 94 enable comparisons of frequency of occurrence and interaction forms across 95 ecosystems, scales, and data types (e.g. experimental, spatial, temporal, 96 spatiotemporal) common across routine monitoring programmes. Such a framework 97 would enable comparisons of frequency of occurrence and interaction forms across 98 99 ecosystems, scales, and data types (e.g. experimental, spatial, temporal, 100 spatiotemporal) common across routine monitoring programmes. One important consideration is that data forms and frequency vary significantly between ecosystems. 101 Feld et al. (2016) presented a synthesis of approaches for determining the presence 102 103 of interactions between multiple stressors in freshwater ecosystems in this context, including the use of generalised linear mixed effects modelling (GLMM) and Birk et al. 104 (2020) demonstrate that this approach is applicable to all forms of data to identify 105 interaction forms (Table 1) across scales in fresh waters. Here we extend this 106

approach to assess the probability of exceedance of water quality stressors when 107 applied to future stressor change scenarios (e.g. Figure 1). This approach addresses 108 two important statistical conditions necessary to underpin practical guidance to water 109 managers: (1) that models should be constructed to determine interactions, 110 specifically, and (2) that a standardized approach for model construction (i.e. stressor 111 selection) is necessary. These conditions minimize bias in the model construct to 112 113 improve representativeness and deliver the best 'model fit', statistically, whilst acknowledging the importance of scale and data guality as limiting factors in their 114 115 application.

We demonstrate this approach for three contrasting (i.e. data forms, frequency, and 116 scale) European catchments to test the hypothesis that significant interaction effects 117 would be detectable between nutrient and climate sensitive stressors on ecological 118 quality indicators, known locally to be sensitive to nutrients. The catchments were 119 120 selected to represent contrasting but realistic data forms: Loch Leven Catchment, UK: assessment of phytoplankton responses in a single lake with time series data; Pinios 121 Catchment, Greece: macroinvertebrate community response across multiple rivers 122 using spatial data; and Lepsämänjoki Catchment, Finland: phytoplankton response 123 across multiple rivers using spatio-temporal data. The resultant best fit paired stressor 124 GLMMs were applied to estimate the expected mean effect of stressor change on 125 ecological indicators relative to critical values. We discuss the strength of the 126 mechanistic evidence to support the model outputs and the implications of the 127 analyses with respect to informing local scale multiple stressor management 128 responses. 129

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#### 131 **2. Methods**

Our approach was to utilise local mechanistic understanding to select indicators 132 sensitive to climate change and nutrient enrichment as well as ecological quality 133 indicators being utilised to inform environmental management at the local scale. As 134 such, we do not produce comparable or balanced data sets with which to compare 135 136 statistically the model outputs between catchments and we acknowledge that other stressors may produce higher order interactions not captured here. We acknowledge 137 that data availability on such indicators will vary between sites and across scales (Birk 138 et al., 2020) and also that their biophysical behaviour will be moderated depending on 139 the ecosystem morphology and geographical locations (Adrian et al., 2009). As such, 140 the model outputs do not offer general explanation of wider scale ecological 141 responses. For each catchment, we used a 'dredge' analysis to produce a range of 142 models including combinations of pairwise stressor effects against ecological quality 143 indicators, the latter calculated as per requirements of site specific ecological quality 144 assessment procedures. From this analysis, we selected the best fit models using 145 model Akaike Information Criterion (AIC) values to explore catchment specific future 146 stressor change scenarios, informed by local climate change projections. The 147 indicators included from each site and the model selection criteria are described in 148 149 detail in the following sections.

# 150 **2.1 Study site description, data sources, and stressor change scenarios**

151 **2.1.1 Loch Leven** 

Loch Leven, a shallow lake in the UK, offers a time series from a single sample site with roughly fortnightly sampling frequency between 1967 and 2017. Data were obtained from the Loch Leven long term monitoring dataset (May & Spears, 2012) across multiple years (1968-2013) with 39 years of data used in the final analysis as

a result of missing values. In this study we consider the ecological response as 156 chlorophyll a concentration in the water column, a proxy for phytoplankton 157 concentration, as well as potential stressor indicators for nutrient enrichment (i.e. total 158 phosphorus (TP) concentrations) and climate change (i.e. water temperature and 159 precipitation) (Table 2). Water temperature, chlorophyll a, TP concentrations were 160 determined roughly fortnightly during the monitoring period and processed to provide 161 162 mean values as indicated in Table 2. Stressor data were averaged across growing season (May through September) and autumn/winter (October through April) with 163 164 chlorophyll a averaged annually to be more in line with WFD methodology (Poikane et al., 2010). Methods for the determination of the Loch Leven indicators are described 165 by Dudley et al. (2013), with the exception of precipitation data which were retrieved 166 from the British Atmospheric Data Centre and processed from daily values 167 representing local conditions as described by Carvalho et al. (2012). These indicators 168 have been shown previously to play an important role in ecological community 169 structure in Loch Leven (Ferguson et al., 2007). 170

Target values for chlorophyll *a* concentration 'good-moderate' boundary were selected from the site-specific targets defined by the EU Water Framework Directive (WFD) for annual mean concentrations at 11  $\mu$ g L<sup>-1</sup> (Carvalho et al., 2009). A review of target setting for Loch Leven and as conducted by the WFD generally is offered by Carvalho et al. (2012) and Hering et al. (2010), respectively.

As a result of climate change, by 2050 the east of Scotland is expected to experience a 1-2°C rise in annual and summer average daily temperature, at the 10% probability level and assuming a medium emissions scenario (UKCP09 SRES A1B; Nakićenović et al., 2000). Under the same scenario, summer and winter precipitation is predicted to decrease by 20-30% and increase by 0-10%, respectively. More

frequent and intense rainfall events are also predicted. O'Reilly et al. (2015) report an
 observed warming rate of about 0.7°C per decade in Loch Leven surface waters.

#### 183 **Pinios Catchment, Greece**

The Pinios Catchment represents multiple river monitoring data across 76 river 184 monitoring sites collected during autumn (i.e. between September, October, 185 November) 2002. The ecological response indicator used was Average Score Per 186 Taxon (ASPT) calculated using macroinvertebrate taxon data from each site (Armitage 187 et al., 1983). Climate change sensitive indicators included water temperature, 188 discharge and dissolved oxygen concentration and indicators of nutrient enrichment 189 included PO<sub>4</sub>-P and NO<sub>3</sub>-N concentrations. Other authors have confirmed the 190 191 sensitivity of dissolved oxygen to climate change in fresh waters (Adrian et al., 2009; Benateau et al., 2019) where hypoxic events have been confirmed to coincide with 192 droughts and low-flow conditions in the Pinios catchment. Target values for ASPT 193 scores were set according to Lazaridou et al. (2016) and represent the 'good-194 moderate' boundary as defined by the EU WFD at 4.81. A description of the methods 195 used for these determinands in the Pinios Catchment are available from Panagopoulos 196 et al. (2014). 197

Climate change projections according to the most pessimistic scenario (RCP 8.5 rising emissions) predict an approximately 2 °C rise in mean annual air temperature and a 10% decrease in annual precipitation for the Pinios catchment by 2060 (Stefanidis et al., 2018). Assuming that projected increases in surface water temperatures are often 50 % to 70 % of the projected increases in air temperature (EEA, 2008), a 2 °C rise in mean annual air temperature could mean a 1.4 °C rise in water temperature. We

assume here that these changes will result in a decrease in dissolved oxygen
 concentration as discussed in detail by Stefanidis et al. (2018).

# 206 2.1.2 Lepsämänjoki Catchment, Finland

The Lepsämänjoki Catchment (214 km<sup>2</sup>) is a sub-basin of the Vantaanjoki River Basin in Southern Finland. It belongs to the Long-Term Ecosystem Research Network (http://www.lter-europe.net/lter-europe/about/organisation/facility-types/ltser-

210 platforms) and is one of the best studied catchments in the drainage basins of the Gulf of Finland and the Archipelago Sea in the Baltic Sea. In contrast to other large 211 catchments, these drainage basins consist of several small river catchments draining 212 directly to the sea. Thus, the data set for the Lepsämänjoki Catchment was 213 supplemented by measurements from the neighbouring catchments with similar soil 214 and agricultural production types. The resulting data set represents 10 river sample 215 sites which were sampled 7 to 25 times a year during the period 1985-2014. The 216 ecological response indicator chosen for this catchment was summer mean 217 chlorophyll a concentration. Climate change sensitive indicators included estimates of 218 219 modelled catchment run-off and water temperature and nutrient enrichment indicators included TP concentration. The methods used for each of these determinands are 220 221 described by Niemi et al. (2001). The target for summer mean chlorophyll a concentration was set at 14.5 µg L<sup>-1</sup> using expert judgement. 222

The mean annual precipitation in the area is 650 mm, and the mean annual temperature is +4°C. In winter, temperature drops below 0°C. Climate change projections, according to the most pessimistic scenario (RCP 8.5 rising emissions), predict an increase of approximately 2.5 °C in mean annual air temperature and less

than 10% increase in annual precipitation by 2060. The increase is most highlightedin spring and autumn.

#### 229 **2.2 Data processing and model construction**

# 230 2.2.1 Two-way interaction models

For each dataset (Table 2), statistical models were developed to predict the ecological quality indicators as a function of two main stressor effects and their interaction, within the framework of linear mixed effects models (LMEs). All response variables were modelled with Gaussian errors. The exact form of the model fixed and random effects varied depending on the dataset structure as described below. However, the full LME specification was,

237 
$$y = b_0 + b_1 x_1 + b_2 x_2 + b_3 x_1 x_2 + S + Y + \epsilon$$

in which y is the response variable,  $x_1$  and  $x_2$  are two stressor covariates, the b terms 238 are the model fixed effect coefficients and  $\epsilon$  is the residual error. For the multi-site and 239 240 multi-year study at Lepsämänjoki, it was necessary to include normally distributed random effects for site S (to account for repeated measures at the same site) and year 241 Y (to account for repeated measures over time). For simplicity, we only included these 242 random effects as random intercepts and not slopes. However, our approach could be 243 extended to specific applications where sufficient data can accommodate more 244 complex mixed effects models. No random effects were needed for the purely 245 temporal (Leven) or spatial (Pinios) studies. As such S and Y were dropped from their 246 model specification, simplifying the model to a standard linear model (LM). 247

All models were fitted in R by maximum likelihood using the stats or ImerTest packages
(Kuznetsova et al., 2017) for LM and LME models, respectively (R Core Team, 2020).
Prior to model fitting the response variables and covariates were transformed to

normal distributions using Box-Cox transformations, offset by a small value to ensure
all values of the variable were greater than 0. This ensured the models met
assumptions of normality of residuals, checked by examining model residual plots.

For each dataset, a set of candidate stressor variables was identified as described 254 above (Table 2). To identify the best combinations of stressor variables to use we 255 256 conducted a 'dredge' analysis in which all possible model combinations with up to two main fixed effects and their interaction were fitted. For simplicity we did not consider 257 models with more fixed effects, though the analysis described below can 258 straightforwardly accommodate more complex models. Random effects were not 259 varied in model selection, as we considered them imposed by the data structure. For 260 the purposes of the analysis described below, the most parsimonious model was 261 selected for each catchment based on the lowest Akaike Information Criteria (AIC<sub>c</sub>); 262 although the output of the dredge analyses is provided displaying all model 263 264 combinations returned. We opted not to utilize model averaging approaches as they may obscure the detection of interactions. 265

#### 266 2.2.2 Risk of threshold exceedance

The probability of the response variables exceeding the site-specific threshold values were evaluated across both stressor gradients and visualized as a heat map, using the two strongest explanatory variables. Depending on the variable, exceedance can mean the response variable being below or above a threshold, but always indicates deterioration in ecological condition. When interpreting these heat maps, it should be noted that the direction of the independent and interaction effects patterns within the gradient of data, only, were used to construct the model.

The heat maps were constructed by calculating exceedance probabilities from the 274 model for a range of stressor combinations. For any values of the two stressors, the 275 model states that observed values of the response variable are normally distributed 276 with a mean of  $b_0 + b_1 x_1 + b_2 x_2 + b_3 x_1 x_2$  and variance of  $\sigma_s^2 + \sigma_y^2 + \sigma_{\epsilon}^2$ , where  $\sigma_s^2$  is 277 the site-level random effect variance,  $\sigma_Y^2$  is the year-level random effect variance and 278  $\sigma_{\epsilon}^2$  is the residual variance. Note that for the datasets for which LM was used, this 279 variance simplifies to  $\sigma_{\epsilon}^2$ . The cumulative distribution function of the Gaussian 280 distribution was used to calculate the probability that an observed response value 281 drawn from this distribution exceeded the chosen threshold. 282

As well as heat maps, stressor scenario plots were produced showing the effects on 283 ecological indicators relative to critical thresholds of predicted changes in climate 284 change sensitive stressor indicators in the context of nutrient stressor indicators. 285 Scenarios were selected to allow visualization around the climate change projections 286 outlined above for the stressors included in the model outputs for each catchment. The 287 assessment of climate change effects for the Pinios Catchment is based on the 288 assumption that oxygen concentration will decrease as a result of prolonged drought 289 periods and higher water temperatures (Stefanidis et al., 2018). The scenario ranges 290 were, for Loch Leven: 0.0; -0.5; -1.0; and -1.5 mmday<sup>-1</sup> change in growing season 291 mean precipitation (0-60% decrease relative to mean value across data); Pinios 292 Catchment: 0.0; -0.5; -1.5; and -2.5 mgL<sup>-1</sup> change in autumn dissolved oxygen 293 concentration (0-30% decrease); Lepsämänjoki Catchment: 0.0; +1.0; +2.0, and +3.0 294 °C change in mean growing season water temperature (0-17% increase). As above, 295 responses were quantified in terms of probability of threshold exceedance. 296

297 **3. Results** 

## 298 **3.1 Loch Leven Catchment**

According to AIC values, the most parsimonious LM model for Loch Leven (Supp 1a) indicated that annual mean chlorophyll *a* concentration was significantly related to winter mean TP and growing season mean precipitation with no significant interaction between them (Table 3). Although this was the best fitting model, there was also substantial support for a model based on growing season and winter TP ( $\Delta AIC_c =$ 0.420, Supp 1a).

The heat maps created with the best fit model are shown in Figure 2. For the best fit Loch Leven model, it is apparent that the highest effect and, therefore probability of exceeding the critical value, occurs when growing season mean precipitation is lowest and winter mean TP is highest.

309 We explored the effects of predicted changes in growing season mean precipitation, linked to climate change, in the context of the model produced for Loch Leven (Figure 310 3). It is apparent that the greatest relative effect of decreasing growing season mean 311 precipitation on the probability of exceeding critical values of annual mean chlorophyll 312 a concentration occurs at the lowest winter mean TP concentrations. The projected 313 decrease of up to 20% (annual mean precipitation) in this region would equate to about 314 - 0.5 mm d<sup>-1</sup>. Assuming this translates into a decrease during growing season mean 315 precipitation of the same value would result in an increased likelihood of failing the 316 critical value of about 10%; relative to no change; up to about 60 µg L<sup>-1</sup>, after which 317 the scenario lines converge. The growing season mean chlorophyll a concentration 318 during the monitoring period was 42.09  $\mu$ g L<sup>-1</sup> (Table 2). 319

#### 320 3.2 Pinios Catchment

321 The most parsimonious LM for ASPT in the Pinios Catchment (Supp 1b) supported 322 effects of nitrate and dissolved oxygen concentrations, without interactions. Nitrate

concentration varied negatively and dissolved oxygen positively with ASPT. The model comparison also provided support for alternative models based on nitrate alone ( $\Delta AIC_c$ = 0.331) and nitrate, pH and their interaction ( $\Delta AIC_c$  = 1.962).

The best fit model included a significant negative effect of nitrate and a significant positive effect of dissolved oxygen (Table 3). The heat maps created with the best fit model are shown (Figure 2) indicating that the probability of passing the critical value remains high at dissolved oxygen concentrations above about 8 mg L<sup>-1</sup> regardless of the NO<sub>3</sub>-N concentration and that at low NO<sub>3</sub>-N concentrations (i.e. near 0 mg L<sup>-1</sup>) the effect of DO is diminished.

The effects of predicted changes on the ASPT, linked with potential future changes in dissolved oxygen, assumed here to reflect future climate change effects (Stefanidis et al. 2018), are shown (Figure 3). A decrease in autumn mean dissolved oxygen concentration of up to 2.5 mg L<sup>-1</sup> would result in about 20% increased likelihood of failing the critical value for ASPT, relative to the no change scenario. This effect appears to be consistent above about 2 mg L<sup>-1</sup> NO<sub>3</sub>-N. The mean NO<sub>3</sub>-N concentration over the monitoring period was 2.21 mg L<sup>-1</sup>.

# 339 3.3 Lepsämänjoki Catchment

The dredge analysis of LME models for summer mean chlorophyll *a* concentration in the Lepsämänjoki Catchment (Supp 1c) indicated that the best fitting model had effects of mean summer TP, mean summer water temperature and their interaction. Four other model specifications had comparable support ( $\Delta AIC_c < 2$ ), and all of these included effects of summer water temperature and one other non-interacting stressor (Supp 1c).

The best fit model indicated that chlorophyll a concentration increased with increasing mean summer TP and water temperature and that these interacted synergistically (Table 3, Figure 2). The probability of exceedance of the critical value was low below about 16 °C regardless of summer mean water TP and the effect of temperature diminished below about 100  $\mu$ g L<sup>-1</sup> summer TP.

We explored the effects of predicted changes in summer mean water temperature, linked to climate change (Figure 3). An increase of 1 °C in the mean summer temperature would increase the probability of the critical threshold being exceeded by about 10% at the mean annual TP concentration of 120  $\mu$ g L<sup>-1</sup> (Table 2), relative to the no change scenario. An increase of 2 °C would increase the likelihood of failure by about 30% at the same TP concentration. The relative effects of the warming scenarios increase, generally, with TP concentrations.

#### 358 4. Discussion

The modelling approach reported provides a means of visualising and quantifying the 359 effects of paired stressors and their interactions on indicators of ecological responses. 360 We acknowledge that inclusion of a greater number of stressors would potentially 361 result in improved model fit and the discovery of higher order interactions. 362 Nevertheless, we have focussed our analysis to identify paired stressor interactions in 363 an attempt to provide relevant outputs for practical management considerations, which 364 365 require a level of simplification, focussing on nutrient enrichment and climate sensitive stressor indicators. We have demonstrated that the approach can be used on a range 366 of data types, including temporal, spatiotemporal and spatial data across single or 367 multiple ecosystems. There is potential for the approach to be used across all 368 ecosystem types and all data types, including experimental data (Birk et al. 2020). We 369

outline the specific models, evidence from the literature to support their underlying
 processes, and the relevance of the future stressor change analyses for future
 management in each case study in the following sections.

# 4.1 Effects of multiple and interacting stressors on each demonstration site

Our model indicated that summer precipitation and winter TP acted additively on 374 growing season chlorophyll a concentration in Loch Leven, and that TP was the 375 376 dominant stressor. The model results agree generally with other studies in which the drivers of water quality and chlorophyll a have been reported for Loch Leven. Carvalho 377 et al. (2012) associated a significant increase in spring Daphnia densities in recent 378 years to increases in water temperature that coincided with reductions in chlorophyll 379 a concentration and increases in water clarity in spring and early summer, indicating 380 higher order trophic interactions not explored here (Rigosi et al., 2014). At the same 381 time, high rainfall was associated with low chlorophyll a concentration, probably as a 382 result of increased flushing rate (Carvalho et al., 2012). However, May et al. (2017) 383 indicated that intense periods of rainfall also resulted in an increase of P load to Loch 384 Leven, although the ecological effects of this observation were not dominant in our 385 models. The winter TP concentration in Loch Leven is expected to reflect catchment 386 387 P loading to the lake, with summer conditions reflecting internal cycling of P between bed sediments and the overlying water column (Sharpley et al., 2013). Spears et al. 388 (2006) explored the potential for hydrological regulation of Loch Leven to increase the 389 flushing rate in summer months to relinquish P associated with internal loading which 390 is manifest within the lake as a summer peak in TP. The model presented here 391 suggests that this would also be a sensible option for controlling phytoplankton 392 biomass which is not unsurprising given the strong correlation between TP and 393 chlorophyll a concentration in this lake. The model demonstrates well the capacity for 394

P control to be used to, at least in part, reduce the impact of future drier summers although a reduction in the annual mean TP concentration even to 30  $\mu$ g L<sup>-1</sup>, would still carry a 50% risk of failure of the WFD chlorophyll *a* target if summer precipitation falls by only 1 mm day<sup>-1</sup>.

For the Pinios catchment, nitrate and dissolved oxygen concentrations acted additively 399 400 on ASPT, and nitrate was the dominant stressor. These results are in agreement with the findings from previous studies (Stefanidis et al., 2016a; 2018) where ASPT was 401 associated with nutrients and dissolved oxygen reflecting the ability of ASPT to capture 402 changes in the abiotic environment related to nutrient and organic pollution (e.g. 403 anoxic conditions). The role of nutrients, mainly nitrogen, on the occurrence of benthic 404 invertebrate taxa has been documented in numerous studies performed elsewhere in 405 Europe and the rest of the world (Johnson and Hering 2009; Villeneuve et al. 2015). 406 For instance, several studies have reported important relationships between nitrogen 407 408 species (TN, NH<sub>4</sub>-N) and macroinvertebrate communities (Wang et al. 2007; Ashton et al. 2014; Stefanidis et al. 2016a), confirming that nitrogen is a key predictor of the 409 ASPT metric. However, these relationships indicate an indirect effect of eutrophication 410 on macroinvertebrate communities although direct toxic effects of nitrate on specific 411 invertebrates are possible given high enough concentrations or exposure time 412 (Camargo et al., 2005). Laboratory studies have shown that nitrate concentrations of 413 10 mg L<sup>-1</sup>, which is within the range observed in the study catchment, can have 414 adverse effects on sensitive aquatic animals (Camargo and Alonso 2006). 415 Furthermore, experimental studies have indicated that nutrient effects on stream 416 macroinvertebrates are indirect, affecting the food supply (e.g. periphyton) and thus 417 altering community composition (Elbrecht et al. 2016). In addition, under conditions of 418 nutrient surplus, excessive algal and microbial growth will lead to oxygen depletion 419

(Johnson and Hering 2009; Dahm et al. 2013). Lack of oxygen will have a direct effect 420 on aquatic animals, and a range of indicator species are especially sensitive to low 421 oxygen levels (e.g stonefly and mayfly taxa; Calapez et al. 2018). Since dissolved 422 oxygen concentration is inversely related to water temperature, warming is expected 423 to affect dissolved oxygen saturation (Cox and Whitehead 2009). Additionally, hypoxia 424 in running waters may occur not only because of organic pollution and eutrophication 425 426 but also due to drought and extreme low flow events, conditions that are becoming more common in Southern Europe (Gudmundsson et al. 2017; Panagopoulos et al. 427 428 2019). We acknowledge that such complexities will be difficult to resolve in our modelling approach, not least because of the potential for covariance between 429 stressors. In these circumstances, model outputs must be considered in the context 430 of comprehensive mechanistic understanding of the system of interest; and we use 431 our outputs, cautiously, to explore potential management implications below. We 432 highlight the need for targeted experimental studies to confirm cause and effect of 433 dominant stressors in such cases. 434

Prolonged periods of low flow or stagnation and high temperatures increase 435 productivity which in turn leads to the decomposition of the excessive organic material 436 and the depletion of oxygen levels (Marcarelli et al. 2010; Bernhardt et al. 2018). In 437 the Pinios Catchment, water overexploitation for irrigation combined with a dry climate 438 during summer maintains low river flows (Stefanidis et al. 2016b) while future climate 439 scenarios predict more frequent low flow and drought events (Stefanidis et al. 2018). 440 Thus, future climate change is expected to impact dissolved oxygen indirectly through 441 changes in the hydrologic regime and increased nutrient pollution but also directly due 442 to warming. These effects have been discussed by Stefanidis et al. (2016a; 2018) who 443 444 examined the impact of hydrologic alteration and nutrient enrichment on oxygen and

nitrate levels, confirming that these stressors are key predictors of benthic invertebrate 445 indices, including ASPT, in the Pinios Catchment. However, the form of the effect may 446 447 be expected to vary along the stressor gradient. For example, where dissolved oxygen concentrations are reduced to very low levels then greater rates of denitrification may 448 occur, as demonstrated for shallow European lakes by Weyhenmeyer et al. (2019), 449 potentially increasing stress of low dissolved oxygen on macroinvertebrates whilst 450 451 relieving stress through NO<sub>3</sub>-N. Nevertheless, no significant interaction term was returned in our model; perhaps indicating that important interactions may lurk outside 452 453 of our data range.

The model for the Lepsämänjoki Catchment showed that TP and water temperature 454 were the key factors controlling chlorophyll a concentration, indicating a significant 455 synergistic interaction effect (Rankinen et al. 2019). It is possible that light, here 456 measured as solar radiation, was not a limiting factor during the summer months due 457 458 to longer daylight hours at the higher latitudes (Lat 60 °N). Previous analyses have indicated that agricultural water protection measures have reduced nutrient loads by 459 3% to 43% compared to mid-1980s (Rankinen et al., 2016). In this context, reductions 460 in nutrient concentrations may partly be attributed to a positive effect of warming on 461 forest growth, as longer and warmer growing seasons have improved nutrient uptake 462 in vegetation (Henttonen et al., 2017). However, a longer growing season may 463 increase the need to mitigate P, because rising temperatures may increase yields and 464 thus add pressures to intensify agriculture. This in turn, would increase the nutrient 465 load to rivers in this area (Rankinen et al., 2013). 466

According to the current Finnish agri-environmental programme, vegetation should be removed from buffer zones at least once during growing season to remove excess nutrients and to reduce dissolved P load (Aakkula et al. 2012). If the focus is also on

improving the ecological status of the river, our analysis suggests that other measures 470 around the river itself may also be necessary to achieve ecological quality targets 471 where nutrient enrichment is projected to increase. For example, the rise in water 472 temperature may be controlled through shading by allowing the growth of taller riparian 473 vegetation in buffer zones. It has been shown that shading by vegetation can decrease 474 water temperature by up to 3 °C (Garner et al. 2017; Loicq et al. 2018; Turunen et al. 475 476 2019). The benefits of riparian shading for maintaining low stream water temperatures have been documented by several studies (e.g. Kristensen et al 2015; Dugdale et al 477 478 2018), although the technical details regarding the implementation of riparian shading as a management measure are still vague (e.g. extent of cover, width of riparian strip, 479 etc.). Conversely, the reduction of water abstraction during the summer months could 480 act as a more feasible management option that may compensate for a decrease in 481 oxygen levels by ensuring higher flows and averting the risk of hypoxic events. Any 482 such measures require site scale assessment of effectiveness. 483

# 484 **4.2 Relevance for informing management of multiple stressors**

Our aim was to demonstrate a simple empirical modeling approach to allow the 485 detection of interactions between dominant paired (i.e. climate x nutrient) stressors in 486 487 three contrasting catchments. We confirm that in only one catchment was such an interaction returned. In the sections above, we have presented mechanistic 488 understanding to aid with interpretation of the model outputs and in some cases we 489 identify that interactions may be expected to occur outside of our data ranges, 490 complicating management responses. Nevertheless, our model outputs offer scope 491 for future assessment of climate change related management; especially where 492 nutrient reduction measures are viewed as being achievable at the local scale. 493 Indicators of nutrient enrichment and climate change stress were important predictors 494

of ecological quality in the models for all catchments. However, we stress the need to better understand the effects of projected climate change on relevant stressor indicators at the catchment scale (Adrian et al., 2009). Two important sources of uncertainty are relevant here. Firstly, the climate change projections, themselves, carry significant uncertainty. Secondly, the effects of projected changes in local or regional weather on stressors of ecological indicators can be difficult to constrain, as discussed for the Pinios Catchment above.

Our analysis suggests, generally, that the projected decrease in precipitation in the 502 Loch Leven catchment could be, at least partly, addressed by a reduction in winter TP 503 concentrations. The greatest increase in probability of exceeding critical thresholds for 504 Loch Leven occurred at lower nutrient levels. In contrast, in the Lepsämänjoki 505 Catchment the effect of an increase in summer water temperature was most prominent 506 at higher nutrient levels. So the potential for nutrient reduction to address climate 507 508 change effects appears to be greatest at lower nutrient levels in the Loch Leven Catchment but higher nutrient levels in the Lepsämänjoki Catchment. 509

Historically, management of water bodies has focussed on the control of single 510 stressors (Verdonschott et al., 2011) which are assumed to be dominant. This 511 approach is attractive in that it meets the practical needs of water managers, offering 512 a simple conceptual model; 'reduce the primary pressure and the ecosystem will 513 recover'. However, our results indicate that at the catchment scale secondary and 514 potentially interacting stressors may cause ecosystems to behave in a manner that is 515 516 unexpected when considering the single stressor management approach. Our models derived from compulsory monitoring programmes (e.g. Munné et al., 2015) feature 517 relatively poor relationships with a lot of noise remaining unexplained. Thus, this 518 approach should be considered to offer initial conceptual understanding of ecosystem 519

behaviour allowing managers to systematically go beyond the primary stressor
approach to consider adaptive responses to future climate change and nutrient
enrichment (Pullin and Knight, 2009; Ryder et al., 2010).

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Table 1. Overview of interaction types and indication from model outputs considering two potentiallyinteracting stressors.

Type of interaction	Characterisation
Synergistic	Model coefficients for both stressors and their interaction all have the same sign (i.e. all positive or all negative)
Antagonistic	Model coefficients for both stressors have the same sign, but their interaction has the opposite sign
Opposing	Model coefficients for both stressors differ, sign of the interaction term not important

Tables 2. Summary statistics for data included in the Loch Leven, Pinios, and Lepsämänjoki catchment
 analyses. Std. dev – standard deviation of the mean; Min – minimum of the range; Max – maximum of
 the range; N – number of computed values used in the analysis (e.g. one value per year for Loch Leven);
 EQI – ecological quality indicator; ASPT – average score per taxon value for macroinvertebrate
 community.

Variable	Season	Mean	St dev	Median	Min	Max	Ν
Loch Leven Catchment, UK							
Total phosphorus (µg L <sup>-1</sup> )	Growing season	69.38	26.521	66	30.52	141.27	39
Total phosphorus (µg L <sup>-1</sup> )	Autumn/Winter	60.57	15.679	60.47	29.69	88.2	39
Precipitation (mm day <sup>-1</sup> )	Growing season	2.415	0.641	2.352	1.307	4.262	39
Precipitation (mm day <sup>-1</sup> )	Autumn/Winter	3.022	0.627	2.931	1.705	4.234	39
Water temperature (°C)	Growing season	15.05	0.966	15.25	12.1	17.36	39
Water temperature (°C)	Autumn/Winter	5.742	0.807	5.769	4.059	7.313	39
Chlorophyll <i>a</i> (µg L⁻¹) – EQI	Annual	42.09	19.461	37.31	18.25	93.22	39
Pinios catchment, Greece							
PO₄-P (µg L⁻¹)	Autumn	28.25	27.45	20.71	0.00	109.45	76
NO₃-N (mg L <sup>-1</sup> )	Autumn	2.21	2.91	1.64	0.00	17.17	76
Dissolved oxygen (mg L <sup>-1</sup> )	Autumn	8.16	1.64	8.15	3.71	10.93	76
Water Temperature (°C)	Autumn	16.85	2.79	17.65	8.30	20.80	76
рН	Autumn	8.00	0.46	7.97	6.27	8.64	76
Discharge (m <sup>3</sup> s <sup>-1</sup> )	Autumn	3.79	2.84	2.95	0.00	8.54	76
ASPT – EQI	Autumn	4.74	1.33	4.69	1.67	7.44	76
Lepsämänjoki Catchment, Fin	land						
Total phosphorus (µg L⁻¹)	Growing season	120.65	48.07	113.45	41.5	349.33	177
Catchment run-off (mm day <sup>-1</sup> )	Growing season	4.17	3.03	3.26	0.48	17.38	177
Water temperature (°C)	Growing season	17.92	1.70	17.94	13.77	22.15	177
Chlorophyll a (µg L⁻¹) – EQI	Growing season	18.61	19.67	12.63	1.4	142	177

Table 3. Summary of fixed effects from models for each study system, explaining different ecological
 responses (Leven, Chlorophyll *a*; Pinios, ASPT; Lepsämänjoki, Chlorophyll *a*). For each system, the
 optimal combination of two fixed effects from Table 2 and their interaction were selected based on
 AIC values. Also given are the adjusted R<sup>2</sup> calculated based on the likelihood-ratio tests against the
 intercept-only model.

	Estimate	se	t	Р
Loch Leven Catchment, UK (R <sup>2</sup> <sub>adj</sub> = 0.616)				
Intercept	0.000	0.107	0.000	>0.999
Winter mean total phosphorous	0.610	0.117	5.200	<0.001
Growing season mean precipitation	-0.276	0.117	-2.355	0.024
Pinios Catchment, Greece (R <sup>2</sup> adj = 0.352)				
Intercept	0.000	0.095	0.000	>0.999
Nitrate concentration	-0.370	0.151	-2.449	0.017
Dissolved oxygen concentration	0.239	0.151	1.582	0.112
Lepsämänjoki Catchment, Finland (R <sup>2</sup> adj = 0.301)				
Intercept	-0.011	0.134	-0.083	0.935
Summer mean total phosphorous	0.075	0.079	0.948	0.346
Summer mean water temperature	0.415	0.079	5.223	<0.001
Interaction (synergistic)	0.140	0.066	2.110	0.036

#### 753 Figure Legends

Figure 1. General analytical framework for approach and description of assessment of risk factors
 including expected responses in relation to critical threshold and the probability that the critical
 threshold will be exceeded for a given stressor combination.







761 Figure 2. Contour plots (a) and (b), Loch Leven, show the effects of winter total phosphorus (TP) concentration and growing season mean precipitation on the expected response in annual mean 762 chlorophyll a concentration (left hand panel) and the probability of exceeding the critical value (right 763 hand panel; the critical value, back line, is the WFD good/moderate target of 11 µg L<sup>-1</sup> annual mean 764 chlorophyll a concentration). Contour plots (c) and (d), Pinios Catchment, showing the effects of 765 766 nitrate concentration and dissolved oxygen concentration on the expected response in ASPT (left hand 767 panel) and the probability of exceeding the critical value (right hand panel; the critical value, the black 768 line, is the WFD good/moderate target of 4.81). Contour plots (e) and (f), Lepsämänjoki Catchment, 769 show the effects of summer mean total phosphorus (TP) concentration and summer mean water 770 temperature on the expected response annual mean chlorophyll a concentration (left hand panel) and 771 the probability of exceeding the critical value (right hand panel; the critical value, the black line, is the 772 WFD good/moderate target of 14.5  $\mu$ g L<sup>-1</sup> summer mean chlorophyll *a* concentration).





774 Figure 3. Climate change scenario assessments for Loch Leven (a), the Pinios Catchment (b) and the 775 Lepsämänjoki Catchment (c). Evidence to support each scenario is provided in the methods section; 776 they are considered realistic for each catchment. The assessment for Loch Leven assumes four levels 777 of precipitation change and resultant effects on probability of exceeding the critical value for chlorophyll *a* concentration (i.e. the WFD good/moderate target of 11 µg L<sup>-1</sup> annual mean chlorophyll 778 a concentration) relative to the winter mean total phosphorus (TP) concentration. The Pinios 779 780 Catchment assessment assumes four levels of DO change and resultant effects on probability of 781 exceeding the critical value of ASPT (i.e. the WFD good/moderate target of 4.81) relative to the 782 nitrogen concentration. The Lepsämänjoki Catchment assessment assumes four levels of temperature change and resultant effects on the probability of exceeding the critical value (i.e. the WFD 783 good/moderate target of 14.5  $\mu$ g L<sup>-1</sup> summer mean chlorophyll *a* concentration) of chlorophyll *a* 784 concentration relative to the summer mean total phosphorus (TP) concentration. All scenario levels 785 786 are shown in the graph legends above each panel.

