

*Biological control agent selection under environmental change using functional responses, abundances and fecundities; the Relative Control Potential (RCP) metric*

Article

Accepted Version

Creative Commons: Attribution-Noncommercial-No Derivative Works 4.0

Cuthbert, R. N., Dick, J. T. A., Callaghan, A. and Dickey, J. W. E. (2018) Biological control agent selection under environmental change using functional responses, abundances and fecundities; the Relative Control Potential (RCP) metric. *Biological Control*, 121. pp. 50-57. ISSN 1049-9644 doi: <https://doi.org/10.1016/j.biocontrol.2018.02.008>  
Available at <http://centaur.reading.ac.uk/75929/>

It is advisable to refer to the publisher's version if you intend to cite from the work. See [Guidance on citing](#).

Published version at: file:///C:/Users/szscalam/Downloads/AS5936016837877761518537078957\_content\_1.pdf

To link to this article DOI: <http://dx.doi.org/10.1016/j.biocontrol.2018.02.008>

Publisher: Elsevier

All outputs in CentAUR are protected by Intellectual Property Rights law, including copyright law. Copyright and IPR is retained by the creators or other copyright holders. Terms and conditions for use of this material are defined in the [End User Agreement](#).

[www.reading.ac.uk/centaur](http://www.reading.ac.uk/centaur)

## **CentAUR**

Central Archive at the University of Reading

Reading's research outputs online

1 Biocontrol agent selection under environmental change using  
2 functional responses, abundances and fecundities; the Relative  
3 Control Potential (RCP) metric

4  
5 Ross N. Cuthbert<sup>1,2</sup>, Jaimie T.A. Dick<sup>1</sup>, Amanda Callaghan<sup>2</sup> and James W.E. Dickey<sup>1</sup>

6 <sup>1</sup>Institute for Global Food Security, School of Biological Sciences, Queen's University  
7 Belfast, Medical Biology Centre, Belfast, BT9 7BL, Northern Ireland

8 <sup>2</sup>Environmental and Evolutionary Biology, School of Biological Sciences, University of  
9 Reading, Harborne Building, Reading, RG6 6AS, England

10 Corresponding author: Ross N. Cuthbert (rcuthbert03@qub.ac.uk)

11

12 **Abstract**

13 1. We currently lack the capacity to rapidly and reliably predict the efficacy of biological  
14 control agents due to inadequate consistency in derivations of functional and numerical  
15 responses and potential effects of context-dependencies.

16 2. Here, we propose and apply a novel metric, Relative Control Potential (RCP), which  
17 combines the functional response (FR, *per capita* effect) with proxies for the numerical  
18 response (NR, agent population response) to compare agent efficacies, where  $RCP = FR \times$   
19 Abundance (or other proxies e.g. Fecundity). The RCP metric is a comparative ratio between  
20 potential biocontrol agents, where values  $> 1$  indicate higher relative control efficacy.

21 Further, RCP can compare the efficacy of agents under environmental contexts, such as

22 temperature change. We thus derived the RCP for two predatory cyclopoid copepods,  
23 *Macrocyclus albidus* and *Megacyclus viridis*, towards larvae of the mosquito *Culex pipiens*  
24 under temperatures representative of current and future climate.

25 3. Both copepods exhibited potentially population destabilising Type II FRs, with increasing  
26 temperatures inducing greater magnitude functional responses through increased attack rates  
27 and decreased handling times. Attack rates by *M. albidus* were higher than *M. viridis*, yet  
28 handling times and maximum feeding rates were similar between the species across all  
29 temperatures.

30 4. The inclusion of abundance data drives an elevated RCP of *M. albidus* and the integration  
31 of fecundity drives greater RCP of *M. albidus* at peak temperatures.  $Q_{10}$  values are indicative  
32 of increased feeding activity by both copepods synonymous with temperature increases,  
33 however relative feeding level increases of *M. viridis* slowed towards the peak temperature.  
34 We present RCP calculations and biplots that represent the comparative efficacies of the two  
35 biological control agents across temperatures.

36 5. *Synthesis and applications.* The Relative Control Potential (RCP) metric provides a tool  
37 for practitioners to better assess the potential efficacy of biocontrol agents before their  
38 integration into management approaches for pests, vectors and invasive species.

39 Keywords: Functional response; Numerical response; Relative Control Potential; *Culex*  
40 *pipiens*; Mosquito; *Macrocyclus albidus*; *Megacyclus viridis*; Copepod

41

## 42 **Introduction**

43 Biological control has been applied to manage pest and invasive species in a variety  
44 of ecological systems (O'Neil, 1990; Marten and Reid, 2007; Van Driesche and Bellows,

2011; Calvo et al. 2016). However, attempts to reveal agent efficacy through the coupling of functional and numerical responses (FRs, NRs) are limited in practice, reducing our predictive capacity for population-level effects (but see Heisswolf et al. 2009; Costa et al. 2017). Further, natural systems are characterised by a number of abiotic and biotic context-dependencies that can alter species interaction strengths, including structural complexity (Barrios-O'Neill et al. 2014b), temperature (Wasserman et al. 2016; South et al. 2017), dissolved oxygen (Laverly et al. 2015), parasitism (Bunke et al. 2015; Laverly et al. 2017b) and multiple/higher predators (Alexander et al. 2013; Barrios-O'Neill et al. 2014a). Thus, rapid and reliable FR and NR derivations under context-dependencies are critical for the future of biocontrol strategies.

Climate change, coupled with urbanisation, is stimulating an unprecedented change in the population dynamics and status of mosquito vectors and their transmission of disease (Townroe and Callaghan, 2014; Medlock and Leach, 2015; Siraj et al. 2017), with mosquito invasions increasing with the transportation of goods and humans (e.g. Yee, 2016; Medlock et al. 2017). The *Culex pipiens* (Linnaeus, 1758) complex is widespread globally and acts as the primary vectors of West Nile virus in the USA and continental Europe (Hubalek and Halouzka, 1999; Fonseca et al. 2004). Freshwater cyclopoid copepods exhibit marked potential for the biological control of mosquitoes (Marten and Reid, 2007; Baldacchino et al. 2017), and have been operationalised in large-scale field applications (Kay and Nam, 2005). However, we require rapid assessment of the relative biocontrol potential of such agents under changing climatic conditions. In this study, we therefore present and apply a new metric, based on FRs and NRs, to compare the efficacy of *Macrocyclops albidus* (Jurine, 1820) and *Megacyclops viridis* (Jurine, 1820) under current and predicted temperature regimes.

69 Functional responses (FRs) quantify consumption under differing resource densities,  
70 describing the key components of search, capture and handling time. Three broad FR types  
71 have been described: the linear Type I, hyperbolic Type II and sigmoidal Type III (Solomon,  
72 1949; Holling, 1959). FR form and magnitude are both powerful predictors of the impacts of  
73 consumers on resource populations across taxonomic and trophic groups (Dick et al. 2014;  
74 2017). However, as FRs only assess *per capita* impacts, incorporation of the Numerical  
75 Response (NR), that is the consumer population response, is also required to discern the Total  
76 Response (TR) of consumers, whereby:

$$77 \quad \quad \quad \text{TR} = \text{FR} \times \text{NR}$$

78 Eqn. 1.

79 In comparison to FRs, NRs are inherently more nebulous and difficult to quantify (Dick et al.  
80 2017). Thus, simple consumer abundance (AB) has recently been proposed as a proxy for the  
81 numerical response, giving the ‘Impact Potential’ (IP) metric, which has proved robust in  
82 predicting ecological impact in the context of invasion biology (Dick et al. 2017; Laverly et  
83 al. 2017a):

$$84 \quad \quad \quad \text{IP} = \text{FR} \times \text{AB}$$

85 Eqn. 2.

86 where FR is the maximum feeding rate (reciprocal of handling time) and AB is a measure of  
87 consumer field abundance. In the context of biocontrol, we can express this as ‘Control  
88 Potential’, CP:

$$89 \quad \quad \quad \text{CP} = \text{FR} \times \text{AB}$$

90 Eqn. 3.

91 where FR is the maximum feeding rate as above. However, in addition, we propose that  
 92 attack rate be used as a second measure of FR, as this parameter describes the slope of the FR  
 93 curve at low prey densities and high attack rates can thus be particularly destabilising to prey  
 94 populations. CP as an absolute measure is, however, rather meaningless, and needs a  
 95 comparator, such as where two or more biocontrol agents require assessment as to their  
 96 relative potential efficacies, hence ‘Relative Control Potential’ (RCP):

$$97 \quad \text{RCP} = \left( \frac{\text{FR agent A}}{\text{FR agent B}} \right) \times \left( \frac{\text{AB agent A}}{\text{AB agent B}} \right)$$

98 Eqn. 4.

99 Where RCP = 1, we predict no difference between biocontrol agents; for RCP < 1, we predict  
 100 agent A to have lesser efficacy than agent B; whereas when RCP > 1, agent A is predicted to  
 101 have greater efficacy than agent B. Further, increasing values above 1 indicate increasing  
 102 relative efficacy of agent A compared to agent B.

103 Furthermore, we propose the use of fecundity as a second proxy for NR, which  
 104 enables the incorporation into RCP of how quickly biocontrol agents can proliferate. Error  
 105 can also be incorporated into the RCP metric depending on data availability, using a  
 106 probability density function (pdf) to generate confidence intervals (CIs) and probabilities that  
 107 RCP > 1 or > 10 (see Dick et al. 2017). Moreover, as contexts such as temperature can have  
 108 profound impacts on consumer-resource interactions (Englund et al. 2011; Rall et al. 2012),  
 109 RCP can be integrated to compare the efficacy of each biocontrol agent across environmental  
 110 gradients. Here, we apply the RCP metric (Eqn. 4) to compare the biological control  
 111 potentials of the copepods *M. albidus* and *M. viridis*, towards the mosquito complex *C.*  
 112 *pipiens* over a temperature gradient reflective of current and future UK climate change  
 113 scenarios. We also apply the Q<sub>10</sub> coefficient to further illustrate feeding activity responses of  
 114 the two agents across temperature variations (Bennett, 1990).

115

116 **Materials and methods**117 *Animal collection and rearing*

118 *M. albidus* and *M. viridis* were collected at Glastry Clay Pit Ponds, Northern Ireland  
119 (54°29'18.5"N; 5°28'19.9"W) in January 2017 and kept in Queen's Marine Laboratory,  
120 Portaferry, N. Ireland, at 25 ± 2 °C under a 16:8 light:dark regime and 50 – 60% relative  
121 humidity. Cultures were initiated using ovigerous females, placed individually into 250 mL  
122 cups with dechlorinated tap water and fed *ad libitum* with *Chilomonas paramecium* and  
123 *Paramecium caudatum* to obtain nauplii. Starter cultures of these protozoans were available  
124 commercially (Sciento, Manchester, England) and cultured under the same laboratory  
125 conditions in 2 L glass beakers using autoclaved wheat seeds, with *C. paramecium* providing  
126 nourishment for nauplii and early copepodids and *P. caudatum* for late copepodids and  
127 adults. Adult copepods were identified by Maria Holyńska, Museum and Institute of Zoology,  
128 Warsaw, Poland. Copepods were mass-reared in 10 L tanks and fed *ad libitum* on the  
129 protozoan diet. At maturity, copepods were maintained at 12 ± 2 °C under a 12:12 light and  
130 dark regime and acclimatised for 7 days prior to experimentation in 5 L holding arenas of 22  
131 cm diameter fed *ad libitum* on the protozoan prey.

132 *Culex pipiens* were obtained from a laboratory colony established at the University of  
133 Reading, originating from field-collected mosquitoes at the The Pirbright Institute, Surrey.  
134 The colony was sustained under the same conditions as the copepods in 32.5 x 32.5 x 32.5 cm  
135 cages (Bugdorm, Watkins and Doncaster, Leominster, England) and fed three times per week  
136 with defibrinated horse blood (TCS Biosciences, Buckingham, England) using a Hemotek®  
137 blood-feeding system (Hemotek Ltd., Accrington, England) and additionally provided with  
138 cotton pads soaked in a 10% sucrose solution. Cages contained black cups filled with 200 mL



139 dechlorinated tap water for oviposition. Egg rafts were extracted three times per week and  
140 placed into larval bowls containing 3 L dechlorinated tap water, and fed *ad libitum* with  
141 ground guinea pig pellets (Pets at Home, Newtownabbey, Northern Ireland) until pupation.

#### 142 *Experimental procedure*

143 Non-ovigerous adult female *M. albidus* and *M. viridis* (1.6 – 1.8 mm and 2.0 – 2.3  
144 mm body length excluding caudal setae, respectively) were selected for experiments. We  
145 selected non-ovigerous females to standardise predators as cyclopoids are sexually dimorphic  
146 (Laybourn-Parry et al. 1988) and to eliminate cannibalism of hatching juveniles (Toscano et  
147 al. 2016). Prey used were recently hatched, first instar *C. pipiens* larvae (1.1 – 1.3 mm).  
148 Functional response experiments were undertaken in transparent polypropylene cups (42 mm  
149 dia.) containing 20 mL dechlorinated tap water from a continuously aerated source in a 12:12  
150 light and dark regime over 24 h at 12 °C, 16 °C and 20 °C (Clifton NEIB water baths),  
151 representing diurnal temperature shifts and reasonable current autumn/winter, spring/summer  
152 and future spring/summer temperatures in the UK, respectively (Hulme et al. 2002;  
153 Hammond and Pryce, 2007). Dissolved oxygen was monitored using a YSI model 550A  
154 meter to ensure levels remained above 80% saturation. Both predators and prey were  
155 acclimatised to the two elevated temperatures over a two hour period prior to experiments;  
156 temperatures were increased every 30 minutes by either 1 °C or 2 °C (i.e. to 16 °C or 20 °C).  
157 Following the acclimatisation period, we added single adult females of either *M. albidus* or  
158 *M. viridis* to containers with prey densities of 2, 4, 8, 15, 30 and 60 ( $n = 4$  per experimental  
159 group). Controls consisted of three replicates at each prey density and temperature in the  
160 absence of predators. Predators were individually starved for 24 h in containers of the same  
161 volume and diameter as the experimental arenas before being transferred to containers  
162 holding the corresponding prey density. Predators were removed from experimental arenas

163 after 24 hours, with the numbers of prey alive counted to derive the numbers killed in each  
 164 replicate.

165 *Data manipulation and statistical analyses*

166 Statistical analyses were undertaken in R v3.3.1. (R Core Team, 2015). Logistic  
 167 regression of proportion of prey killed as a function of prey density was used to infer FR  
 168 types; Type II FRs are characterised by a significant negative first-order term and Type III by  
 169 a significant negative second order term following a significant positive first order term. To  
 170 account for prey depletion, we fitted Rogers' random predator equation for conditions  
 171 without prey replacement (Trexler et al. 1988; Juliano, 2001):

$$172 \quad N_e = N_0(1 - \exp(a(N_e h - T)))$$

173 Eqn. 5.

174 where  $N_e$  is the number of prey eaten,  $N_0$  is the initial density of prey,  $a$  is the attack constant,  
 175  $h$  is the handling time and  $T$  is the total experimental period. The Lambert W function was  
 176 applied due to the implicit nature of the random predator equation (Bolker, 2008). Attack  
 177 rates and maximum feeding rates ' $1/h$ ' were non-parametrically bootstrapped ( $n = 30$ ) to  
 178 facilitate modelling of FR parameters with respect to 'predator' and 'temperature' factors and  
 179 their interactions. Bootstrapped parameters were analysed using generalised linear models  
 180 (GLMs) assuming a quasi-Poisson distribution.  $F$ -tests were used in a step-deletion process to  
 181 compare residual deviances between models (Crawley, 2007). We applied Tukey's HSD  
 182 method to infer specific pairwise differences using the 'multcomp' package in R (Hothorn et  
 183 al. 2008).

184 Benthic survey data for *M. albidus* and *M. viridis* (as *Acanthocyclops viridis*) derived  
 185 from Tinson and Laybourn-Parry (1986) were used to calculate RCP based on maximum  
 186 field abundances using pooled bootstrapped mean maximum feeding and attack rates across

187 all three temperatures. To calculate RCP using fecundity, we used results from Laybourn-  
 188 Parry et al. (1988) to discern the proportion of total consumed energy devoted to reproduction  
 189 across corresponding temperatures for the two copepods:

$$190 \quad \text{Fecundity} = \left( \frac{P_r}{C} \right) \times 100$$

191 Eqn. 6.

192 where  $P_r$  is the quantity of energy expended through the production of eggs and  $C$  is the total  
 193 energy consumed at a given temperature (Table 1). Reproductive energy proportions at 16 °C  
 194 were supplemented with those available for 15 °C.

195 We additionally calculated  $Q_{10}$  values to further quantify the effects of increased  
 196 temperature on feeding rates and compare how these varied between predatory cyclopoids:

$$197 \quad Q_{10} = \left( \frac{R_2}{R_1} \right)^{\left( \frac{10}{T_2 - T_1} \right)}$$

198 Eqn. 7.

199 where  $Q_{10}$  is a coefficient without units,  $R_1$  is the maximum feeding rate at temperature  $T_1$   
 200 and  $R_2$  is the maximum feeding rate at temperature  $T_2$ . The  $Q_{10}$  coefficient assesses how  
 201 temperature increases of 10 °C affect the rate of biological processes (Bennett 1990); values  
 202 of 1 – 1.5 are associated with a thermal plateau and values of 2 – 4 indicate substantive  
 203 increases in activity as temperature increases (Huey, 1982; Bennett 1990). We generated  
 204 ‘RCP biplots’ to present the RCP (see Laverty et al. 2017a) of the two predators using both  
 205 the abundance (AB) and fecundity (FE) proxies for the numerical response.

206

207 **Results**

208 Prey survival in control treatments exceeded 98.5% across all temperatures, and thus  
 209 experimental deaths were attributed to predation by copepods, which was also directly  
 210 observed. Type II FRs were found in all predator and temperature combinations, as indicated  
 211 by significantly negative first order terms (Table 2; Figure 1). Overall, attack rates (initial FR  
 212 slopes; see Figure 1) for *M. albidus* were significantly higher than for *M. viridis* ( $F_{1, 178} =$   
 213  $7.25, p < 0.01$ ) and increased significantly with temperature ( $F_{2, 176} = 74.41, p < 0.001$ ). There  
 214 were significant increases in attack rates between 12 °C and 16 °C ( $z = 5.61, p < 0.001$ ), and  
 215 12 °C and 20 °C ( $z = 6.75, p < 0.001$ ), but not between 16 °C and 20 °C ( $z = 1.20, p = \text{NS}$ ).  
 216 There was a significant ‘predator  $\times$  temperature’ interaction ( $F_{2, 174} = 3.09, p < 0.05$ ),  
 217 reflecting significantly greater attack rates by *M. albidus* only at the lowest temperature ( $z =$   
 218  $3.42, p < 0.01$ ; Table 2; Figure 1). Overall, maximum feeding rates (asymptotes of FR curves;  
 219 see Figure 1) did not differ significantly between the two predators ( $F_{1, 178} = 2.88, p = \text{NS}$ ),  
 220 and increased significantly with temperature ( $F_{2, 176} = 110.29, p < 0.001$ ; Figure 1). There  
 221 were significant increases in maximum feeding rates between all temperature levels (12 °C –  
 222 16 °C,  $z = 4.23, p < 0.001$ ; 16 °C – 20 °C,  $z = 4.79, p < 0.001$ ; 12 °C – 20 °C,  $z = 8.81, p <$   
 223  $0.001$ ). There was a significant ‘predator  $\times$  temperature’ interaction ( $F_{2, 174} = 3.46, p < 0.05$ ),  
 224 reflecting an insignificant difference between maximum feeding rates of *M. viridis* at 16 °C  
 225 and *M. albidus* at 20 °C ( $z = 2.48, p = \text{NS}$ ) compared to a significant difference between *M.*  
 226 *albidus* at 16 °C and *M. viridis* at 20 °C ( $z = 6.24, p < 0.001$ ).

227 The RCP calculations integrating field abundances with maximum feeding and attack  
 228 rates are presented in Table 3. These RCP scores exhibit high certainty and indicate strong  
 229 comparative efficacy of *M. albidus*. It is evident from the biplots in Figures 2a and 2b that *M.*  
 230 *albidus* populations display much higher densities than *M. viridis*, driving greater RCP using  
 231 both FR parameters. The fecundity results in Table 1 reveal a general increase in the  
 232 proportion of total consumed energy devoted to reproduction as temperature increases.

233 Anomalous to this is the response to warming of *M. viridis* at 20 °C, with fecundity here  
234 falling markedly. The ramifications of these fecundity variations for RCP are illustrated in  
235 Table 4 and Figures 2c and 2d, with relatively similar levels of efficacy for the two species  
236 illustrated at both 12 °C and 16 °C, followed by a substantial decrease in efficacy of *M.*  
237 *viridis* at 20 °C. The certainty of the RCP using fecundity strengthens at peak temperatures  
238 (Table 4). Under both NR proxies, differential efficacies were more pronounced using the  
239 attack rate parameter (Figure 2). The results for the  $Q_{10}$  coefficient for the two predators  
240 across the temperature gradient are illustrated in Table 5, and indicate that between 12 °C and  
241 20 °C both the feeding rates of *M. albidus* and *M. viridis* were highly responsive to  
242 temperature increases (2.25 and 2.95 respectively). There was a marked difference between  
243 the predators in the incremental drivers of this response, with *M. viridis* exhibiting a rapid  
244 increase between 12 °C and 16 °C (4.70) which slowed between 16 °C and 20 °C (1.85).  
245 Conversely, *M. albidus* was consistent in its feeding response to increased temperatures (12  
246 °C – 16 °C, 2.29; 16 °C – 20 °C, 2.22; Table 5).

247

## 248 **Discussion**

249 Biological control of pests, disease vectors and invasive species can be effective (e.g.  
250 Hajek, 2007; Nam et al. 2012; Veronesi et al. 2015), but efforts to predict the efficacy of  
251 natural enemies are limited when the functional response (FR) *per capita* effects are solely  
252 considered (Lester and Harmsen, 2002; Fernández-arhex and Corley, 2003). The  
253 complementary numerical response (NR) is, however, somewhat nebulous and difficult to  
254 derive, with proxies for the NR required to allow rapid assessment of the overall impact of a  
255 consumer (i.e. TR; Dick et al. 2017). Recent developments that combine functional and  
256 numerical responses (or their proxies) into a comparative metric (RIP; Dick et al. 2017) yield

257 high explanatory and predictive power for the impacts of invasive species, and hence have  
258 potential in the assessment of the efficacy of biocontrol agents. Further value in the  
259 application of such metrics surrounds the integration of context-dependencies associated with  
260 environmental change, which can strongly affect interactions between consumers and their  
261 resources (e.g. oxygen availability: Lavery et al. 2015; habitat complexity: Barrios-O'Neill  
262 et al. 2014b). Thus, we present the Relative Control Potential (RCP) metric that uses *per*  
263 *capita* and consumer population responses to compare efficacy among biocontrol agents and  
264 can allow predictions of changes in such efficacies under context-dependencies.

265         The risk of mosquito-borne disease at continental scales has reached unprecedented  
266 levels in recent decades (Medlock and Leach, 2015). Arboviruses such as Zika, West Nile,  
267 dengue and chikungunya present enormous public health concern, with disease dynamics  
268 shifting rapidly under environmental change (Benelli and Melhorn, 2016; Siraj et al. 2017).  
269 Agricultural systems will additionally be impacted (Chevalier et al. 2013). This risk  
270 necessitates the formation of techniques to assess and compare the potential efficacies of  
271 biological control agents. Here, temperature was shown to mediate changes to the FR  
272 parameters of *M. albidus* and *M. viridis*, driving higher magnitude FRs through increasing  
273 attack rates and decreasing handling times. The temperature-dependence of attack rates  
274 reported here contrasts to suggestions that this FR parameter is temperature-independent  
275 (Rall et al. 2012; Dell et al. 2014). We show that both predators exhibit high maximum  
276 feeding rates that exceed 30 of the West Nile virus vector *C. pipiens* per day at 20 °C.  
277 Critically, the Type II FRs found are indicative of a capacity to destabilise prey populations  
278 due to high proportional consumption at prey low densities (Long and Whitefleet-Smith,  
279 2013). *M. albidus* and *M. viridis* show strong similarities in their *per capita* consumption,  
280 although the attack rates of *M. albidus* were significantly greater overall, illustrated by

281 steeper gradients in the FR curves at low densities. As a result, *M. albidus* may be more  
282 effective in eliminating *C. pipiens* populations.

283         We demonstrate that integrating field abundances with RCP reveals far stronger  
284 control efficacies of *M. albidus* compared to *M. viridis*. The utility of abundance estimates  
285 lies in the projection of how many conspecifics may engage in the predator-prey (or other  
286 consumer-resource) interaction and it is thus a useful NR proxy. On the other hand,  
287 incorporating measures of fecundity estimates how rapidly biological control agents can  
288 reproduce when introduced. Using fecundity, we demonstrate temperature-dependencies of  
289 control efficacy. Stark fecundity variabilities are illustrated in the RCP biplots, with the  
290 reproductive devotion of *M. viridis* declining rapidly at 20 °C, whilst that of *M. albidus*  
291 continues to rise. The reduction in fecundity shown by *M. viridis* at 20 °C is concurrent with  
292 slowing foraging activity discerned through  $Q_{10}$  analysis, whereas *M. albidus* displays a  
293 consistent incremental increase with warming. Overall, *M. albidus* displays greater potential  
294 for the control of West Nile virus vector *C. pipiens* than *M. viridis*, particularly under climate  
295 change projections where our certainty for differential efficacy increases (e.g. Hulme et al.  
296 2002). This differential efficacy is more pronounced when the attack rate parameter is  
297 applied. An increased metabolic demand could enable *M. albidus* to sustain efficiency in the  
298 consumption of *C. pipiens*, which proliferate more rapidly at higher temperatures, particularly  
299 in domestic and peri-domestic habitats in urbanised environments (Townroe and Callaghan,  
300 2014).

301         In this study, temperature had a profound effect on predator-prey interactions.  
302 Temperature independence of ambush predators such as the benthic copepods examined here  
303 has been described (Awasthi et al. 2012; Novich et al. 2014), with strong dependencies  
304 driven, rather, in respect to prey foraging responses to temperature change. These interactions  
305 make predator-prey systems highly specific to both the species (Englund et al. 2011) and

306 environment (Broitman et al. 2011), with optimal foraging patterns of ectothermic prey  
307 potentially peaking at intermediate temperatures (Englund et al. 2011; Kalinoski and DeLong  
308 2016). In this study, feeding rates of candidate biocontrol agents peaked at the highest  
309 temperature, however the rate of increase slowed between 16 °C and 20 °C, as compared to  
310 between 12 °C and 16 °C; this trend was particularly marked for *M. viridis*. Temperature  
311 additionally has a substantial influence on the development of the focal prey, *C. pipiens*,  
312 driving significant reductions in development times under conditions of warming (Loetti et  
313 al. 2011; Ruybal et al. 2016), and necessitating increased foraging intensity. Yet, increases in  
314 *C. pipiens* mortality due to drivers outside of predation are also evident as temperatures rise  
315 (Ruybal et al. 2016). *M. albidus* has proved particularly effective against the invasive  
316 arbovirus vector *A. albopictus* following field trials (Marten, 1990; Veronesi et al. 2015).  
317 Previous research has suggested that copepods are more efficient consumers of *Aedes* spp.  
318 than *Culex* spp. as a result of morphological variations between the genera (Marten and Reid,  
319 2007). However, laboratory trials have shown similar levels of overall predation by *M.*  
320 *albidus* towards both *A. albopictus* and *C. pipiens* (Veronesi et al. 2015), as well as by  
321 *Mesocyclops annulatus* (Micieli et al. 2002). Further field trials are required to elucidate  
322 whether their efficiencies towards *C. pipiens* translate empirically, particularly as it is the  
323 major West Nile virus vector in the USA and Europe (Hubalek and Halouzka, 1999; Fonseca  
324 et al. 2004) and part of one of the most widespread mosquito complexes in the world  
325 (Harbach, 2012).

326         This is the first study to develop and apply the RCP metric to biological control agent  
327 selection. We demonstrate that the integration of abundance and fecundity estimations can  
328 provide a means to differentiate between biocontrol agents that display similar *per capita*  
329 efficacies across temperature gradients. Adaptations of this metric have been applied  
330 successfully hitherto in the context of invasion biology to explain and predict the impact of



331 invasive species (Dick et al. 2017; Lavery et al. 2017), yet similar fundamental principles  
332 enable its application to the selection of biocontrol agents. Overall, temperature increases will  
333 induce greater *per capita* predation pressure by predatory copepods towards *C. pipiens* and  
334 likely other mosquito species. Copepod applications to waterbodies can form an integral part  
335 of mosquito control efforts (Baldacchino et al. 2015), with large-scale field trials having  
336 proved successful (e.g. Kay and Nam, 2005), particularly given their ability to thrive in  
337 ranging natural and artificial waterbodies (Marten and Reid, 2007). Importantly, copepods  
338 can be augmented synergistically using existing control methods, such as the use of bacterial  
339 *Bacillus thuringiensis var. israelensis* (BTI; Kosiyachinda et al. 2003). Nanoparticles have  
340 additionally been founded to heighten predation (Murugan et al. 2015). The straightforward  
341 derivation of the RCP metric, and its visual representation in biplots, will allow comparisons  
342 of biological control agents across many ecological systems, and could increase cost-  
343 effectiveness of natural enemies in the long-term. Further proxies for numerical response,  
344 such as biocontrol agent longevity or biomass, can be integrated into the RCP metric as per  
345 the requirements of the assessed system or biocontrol approach, increasing the robustness and  
346 flexibility of the method. Moreover, the additional integration of a qualifier to account for  
347 target organism responses under matched environmental change scenarios could bolster the  
348 power of the RCP metric in reliably selecting biocontrol agents, and this is worth further  
349 consideration in future research.

350

### 351 **Authors' contributions**

352 Relative Control Potential (RCP) concept and framework: RNC, JTAD and JWED. Design  
353 and execution of experimental trials: RNC and JTAD. Fitting functional response models and

354 undertaking RCP calculations: RNC. First draft of the manuscript: RNC. Manuscript  
355 revisions: all authors.

356

### 357 **Acknowledgements**

358 *Culex pipiens* eggs were originally developed and supplied by The Pirbright Institute under  
359 BBSRC project code: BBS/E/I/00001701. This study was funded through, and forms part of,  
360 a PhD studentship provided by the Department for the Economy, Northern Ireland.

361

### 362 **Data accessibility**

363 Underlying functional response data will be made available on the Dryad Digital Repository.

364

### 365 **References**

366 Alexander, M.E., Dick, J.T.A., O'Connor, N.E., 2013. Trait-mediated indirect interactions in  
367 a marine intertidal system as quantified by functional responses. *Oikos* 122(11), 1521–  
368 1531. <http://dx.doi.org/10.1111/j.1600-0706.2013.00472.x>.

369 Awasthi, A.K., Wu, C-W., Tsai, K-H., King, C-C., Hwang, J-S., 2012. How Does the  
370 Ambush Predatory Copepod *Megacyclops formosanus* (Harada, 1931) Capture  
371 Mosquito Larvae of *Aedes aegypti*? *Zool. Stud.* 51(7), 927–936.

372 Baldacchino, F., Caputo, B., Chandre, F., Drago, A., della Torre, A., Montarsi, F., Rizzoli,  
373 A., 2015. Control methods against invasive *Aedes* mosquitoes in Europe: A review. *Pest*  
374 *Manage. Sci.* 71(11), 1471–1485. <http://dx.doi.org/10.1002/ps.4044>.

375 Baldacchino, F. Bruno, M.C., Visentin, P., Blondel, K., Arnoldi, D., Hauffe, H.C., Rizzoli,

- 376 A., 2017. Predation efficiency of copepods against the new invasive mosquito species  
377 *Aedes koreicus* (Diptera: Culicidae) in Italy. *Eur. Zool. J.* 84(1), 43–48.  
378 <http://dx.doi.org/10.1080/11250003.2016.1271028>.
- 379 Barrios-O'Neill, D., Dick, J.T.A., Emmerson, M.C., Ricciardi, A., MacIsaac, H.J., Alexander,  
380 M.E., Bovy, H.C., 2014a. Fortune favours the bold: A higher predator reduces the  
381 impact of a native but not an invasive intermediate predator. *J. Anim. Ecol.* 83(3), 693–  
382 701. <http://dx.doi.org/10.1111/1365-2656.12155>.
- 383 Barrios-O'Neill, D., Dick, J.T.A., Emmerson, M.C., Ricciardi, A., MacIsaac, H. 2014b.  
384 Predator-free space, functional responses and biological invasions. *Funct. Ecol.* 29(3),  
385 377–384. <http://dx.doi.org/10.1111/1365-2435.12347>.
- 386 Benelli, G., Melhorn, H., 2016. Declining malaria, rising of dengue and Zika virus: insights  
387 for mosquito vector control. *Parasitol. Res.* 115(5), 1747-1754.  
388 <http://dx.doi.org/10.1007/s00436-016-4971-z>.
- 389 Bennett, A.F., 1990. Thermal dependence of locomotor capacity. *Am. J. Physiol.* 259(2),  
390 253–258.
- 391 Bolker, B.M., 2008. *Ecological Models and Data in R*. Princeton University Press, USA.
- 392 Broitman, B.R., Szathmary, P.L., Mislán, K.A.S., Blanchette, C.A., Helmuth, B., 2009.  
393 Predator-prey interactions under climate change: the importance of habitat vs body  
394 temperature. *Oikos* 118(2): 219–224. [http://dx.doi.org/10.1111/j.1600-](http://dx.doi.org/10.1111/j.1600-0706.2008.17075.x)  
395 [0706.2008.17075.x](http://dx.doi.org/10.1111/j.1600-0706.2008.17075.x).
- 396 Bunke, M., Alexander, M.E., Dick, J.T., Hatcher, M.J., Paterson, R., Dunn, A.M., 2015.  
397 Eaten alive: cannibalism is enhanced by parasites. *Open Sci.* 2, 140369.  
398 <http://dx.doi.org/10.1098/rsos.140369>.

- 399 Calvo, F.J., Torres-Ruiz., A., Velázquez-González, J.C., Rodríguez-Leyva, E., Lomeli-Flores,  
400 J.R., 2016. Evaluation of *Dicyphus hesperus* for biological control of sweet potato  
401 whitefly and potato psyllid on greenhouse tomato. *Biocontrol* 61(4), 415-424.  
402 <http://dx.doi.org/10.1007/s10526-016-9719-2>.
- 403 Chevalier, V., 2013. Relevance of Rift Valley fever to public health in the European Union.  
404 *Clin. Microbiol. Infect.* 19(8), 705-708. <http://dx.doi.org/10.1111/1469-0691.12163>.
- 405 Costa, J.F., Matos, C.H.C., de Oliveira, C.R.F., da Silva, T.G.F., Lima Neto, I.F.A. 2017.  
406 Functional and numerical responses of *Stethorus tridens* Gordon (Coleoptera:  
407 Coccinellidae) preying on *Tetranychus bastosi* Tuttle, Baker & Sales (Acari:  
408 Tetranychidae) on physic nut (*Jatropha curcas*). *Biol. Control* 111, 1-5.  
409 <https://doi.org/10.1016/j.biocontrol.2017.04.015>.
- 410 Crawley, M.J., 2007. *The R book*. John Wiley & Sons, Chichester.  
411 <http://dx.doi.org/10.1002/9780470515075>.
- 412 Dell, A.I., Pawar, S., Savage, V.M., 2014. Temperature dependence of trophic interactions  
413 are driven by asymmetry of species responses and foraging strategy. *J Anim Ecol* 83(1),  
414 70–84. <http://dx.doi.org/10.1111/1365-2656.12081>.
- 415 Dick, J.T.A., Alexander, M.E., Jeschke, J.M., Ricciardi, A., MacIsaac, H.J., Robinson, T.B.,  
416 Kumschick, S., Weyl, O.L.F., Dunn, A.M., Hatcher, M.J., Paterson, R.A., Farnsworth,  
417 K.D., Richardson, D.M., 2014. Advancing impact prediction and hypothesis testing in  
418 invasion ecology using a comparative functional response approach. *Biol. Invasions*  
419 16(4), 735–753. <http://dx.doi.org/10.1007/s10530-013-0550-8>.
- 420 Dick, J.T.A., Lavery, C., Lennon, J.J., Barrios-O'Neill., D., Mensink, P.J., Britton, R.,  
421 Médoc, V., Boets, P., Alexander, M.E., Taylor, N.G., Dunn, A.M., Hatcher, M.J.,  
422 Rosewarne, P.J., Crookes, S., MacIsaac, H.J., Xu, M., Ricciardi, A., Wasserman, R.J.,

- 423 Ellender, B.R., Weyl, O.L.F., Lucy, F.E., Banks, P.B., Dodd, J.A., MacNeil, C., Penk,  
424 M.R., Aldridge, D.C., Caffrey, J.M., 2017. Invader Relative Impact Potential: a new  
425 metric to understand and predict the ecological impacts of existing, emerging and future  
426 invasive alien species. *J. Appl. Ecol.* 54(4), 1259–1267. [http://dx.doi.org/10.1111/1365-](http://dx.doi.org/10.1111/1365-2664.12849)  
427 [2664.12849](http://dx.doi.org/10.1111/1365-2664.12849).
- 428 Englund, G., Ohlund, G., Hein, C.L., Diehl, S., 2011. Temperature dependence of the  
429 functional response. *Ecol. Lett.* 14(9), 914–921. [http://dx.doi.org/10.1111/j.1461-](http://dx.doi.org/10.1111/j.1461-0248.2011.01661.x)  
430 [0248.2011.01661.x](http://dx.doi.org/10.1111/j.1461-0248.2011.01661.x).
- 431 Fernández-arhex, V., Corley, J.C., 2003. The Functional Response of Parasitoids and its  
432 Implications for Biological Control. *Biocontrol Sci. Technol.* 13(4), 403–413.  
433 <http://dx.doi.org/10.1080/0958315031000104523>.
- 434 Fonseca, D.M., Keyghobadi, N., Malcolm, C.A., Mehmet, C., Schaffner, F., Mogi, M.,  
435 Fleischer, R.C. and Wilkerson, R.C., 2004. Emerging vectors in the *Culex pipiens*  
436 complex. *Sci.* 303(5663), 1535–1538. <http://dx.doi.org/10.1126/science.1094247>.
- 437 Hajek, A., 2007. Introduction of a fungus into North America for control of gypsy moth, in:  
438 Vincent, C., Goettel, M.S., Lazarovits, G. (Eds.) *Biological control: a global perspective*.  
439 CAB International, Wallingford, pp. 53–62.  
440 <http://dx.doi.org/10.1079/9781845932657.0000>.
- 441 Hammond, D., Pryce, A.R., 2007. Climate change impacts and water temperature.  
442 Environment Agency Science Report SC060017/SR. Bristol.
- 443 Harbach, R.E., 2012. *Culex pipiens*: Species versus species complex - taxonomic history and  
444 perspective. *J. Am. Mosq. Control Assoc.* 28(4), 10–23. [http://dx.doi.org/10.2987/8756-](http://dx.doi.org/10.2987/8756-971X-28.4.10)  
445 [971X-28.4.10](http://dx.doi.org/10.2987/8756-971X-28.4.10).

- 446 Heisswolf, A., Klemola, N. Ammunét, T., Klemola, T. 2009. Responses of generalist  
447 invertebrate predators to pupal densities of autumnal and winter moths under field  
448 conditions. *Ecol. Entomol.* 34(6), 709–717. [http://dx.doi.org/10.1111/j.1365-  
449 2311.2009.01121.x](http://dx.doi.org/10.1111/j.1365-2311.2009.01121.x).
- 450 Holling, C.S., 1959. Some characteristics of simple types of predation and parasitism. *Can.*  
451 *Entomol.* 91(7), 385–398. <http://dx.doi.org/10.4039/Ent91385-7>.
- 452 Hothorn, T., Bretz, F., Westfall, P., 2008. Simultaneous inference in general parametric  
453 models. *Biometrical J.* 50(3), 346–363. <http://dx.doi.org/10.1002/bimj.200810425>.
- 454 Hubalek, Z., Halouzka, J., 1999. West Nile fever - a remerging mosquito-borne viral disease  
455 in Europe. *Emerg. Infect. Dis.* 5(5), 643–650. <http://dx.doi.org/10.3201/eid0505.990505>.
- 456 Huey, R.B., 1982. Temperature, physiology, and the ecology of reptiles, in: Gans, C., Pough,  
457 F.H. (Eds.), *Biology of the Reptilia*. Academic Press, New York, pp. 25–91.  
458 <http://dx.doi.org/10.1093/icb/icq095>
- 459 Hulme, M., Jenkins, G.J., Lu, X., Turnpenny, J.R., Mitchell, T.D., Jones, R.G., Lowe, J.,  
460 Murphy, J.M., Hassell, D., Boorman, P., McDonald, R., Hill, S., 2002. Climate change  
461 scenarios for the United Kingdom. The UKCIP Scientific Report. Tyndall Centre for  
462 Climate Change Research, School of Environmental Sciences, University of East  
463 Anglia, Norwich.
- 464 Juliano, S.A., 2001. Non-linear curve fitting: predation and functional response curves, in:  
465 Scheiner, S.M., Gurevitch, J. (Eds.), *Design and Analysis of Ecological Experiments*.  
466 Oxford University Press, Oxford, pp. 178–196. [http://dx.doi.org/  
467 10.4319/lo.1995.40.8.1533](http://dx.doi.org/10.4319/lo.1995.40.8.1533).
- 468 Kalinoski, R.M. and DeLong, J.P., 2016. Beyond body mass: how prey traits improve

- 469 predictions of functional response parameters. *Oecol.* 180(2), 543–550.  
470 <http://dx.doi.org/10.1007/s00442-015-3487-z>.
- 471 Kay, B.H. and V.S. Nam. 2005. New strategy against *Aedes aegypti* in Vietnam. *Lancet*  
472 365(9459): 613-617. [http://dx.doi.org/10.1016/S0140-6736\(05\)17913-6](http://dx.doi.org/10.1016/S0140-6736(05)17913-6).
- 473 Kosiyachinda, P., Bhumiratana, A., Kittayapong, P., 2003. Enhancement of the efficacy of a  
474 combination of *Mesocyclops aspericornis* and *Bacillus thuringiensis var. israelensis* by  
475 community-based products in controlling *Aedes aegypti* larvae in Thailand. *Amer. J.*  
476 *Trop. Med. Hyg.* 69(2), 206–212.
- 477 Lavery C., Dick, J.T.A., Alexander, M.E., Lucy, F.E., 2015. Differential ecological impacts  
478 of invader and native predatory freshwater amphipods under environmental change are  
479 revealed by comparative functional responses. *Biol Invasions* 17(6), 1761–1770.  
480 <http://dx.doi.org/10.1007/s10530-014-0832-9>.
- 481 Lavery, C., Green, K.D., Dick, J.T.A., Barrios-O'Neill, D., Mensink, P.J., Médoc, V.,  
482 Spataro, T., Caffrey, J.M., Lucy, F.E., Boets, P., Britton, J.R., Pegg, J., Callagher, C.,  
483 2017a. Assessing the ecological impacts of invasive species based on their functional  
484 responses and abundances. *Biol. Invasions* 19(5), 1653–1665.  
485 <http://dx.doi.org/10.1007/s10530-017-1378-4>.
- 486 Lavery, C., Brenner, D., McIlwaine, C., Lennon, J.J., Dick, J.T.A., Lucy, F.E., Christian,  
487 K.A., 2017b. Temperature rise and parasitic infection interact to increase the impact of  
488 an invasive species. *Int. J. Parasitol.* 47(5), 291–296.  
489 <http://dx.doi.org/10.1016/j.ijpara.2016.12.004>.
- 490 Laybourn-Parry, J., Abdullahi, B.A., Tinson, S.V., 1988. Temperature-dependent energy  
491 partitioning in the benthic copepods *Acanthocyclops viridis* and *Macrocyclus albidus*.  
492 *Can. J. Zool.* 66(12), 2709–2713. <https://doi.org/10.1139/z88-397>.

- 493 Lester, P.J., Harmsen, R., 2002. Functional and numerical responses do not always indicate  
494 the most effective predator for biological control: An analysis of two predators in a two-  
495 prey system. *J. Appl. Ecol.* 39(3), 455–468. [http://dx.doi.org/10.1046-](http://dx.doi.org/10.1046/j.1365-2664.2002.00733.x)  
496 [2664.2002.00733.x](http://dx.doi.org/10.1046/j.1365-2664.2002.00733.x).
- 497 Loetti, V., Schweigmann, N., Burrionia, N., 2011. Development rates, larval survivorship  
498 and wing length of *Culex pipiens* (Diptera: Culicidae) at constant temperatures. *J. Nat.*  
499 *Hist.* 45(36-36), 2207–2217. <http://dx.doi.org/10.1080/00222933.2011.590946>.
- 500 Long, W.C., Whitefleet-Smith, L., 2013. Cannibalism in red king crab: habitat, ontogeny, and  
501 the predator functional response. *J. Exp. Mar. Biol. Ecol.* 449, 142–148.  
502 <http://dx.doi.org/10.1016/j.jembe.2013.09.004>.
- 503 Marten, G.G., 1990. Elimination of *Aedes albopictus* from tire piles by introducing  
504 *Macrocyclus albidus* (Copepoda, Cyclopidae). *J. Am. Mosq. Control Ass.* 6(4), 689–  
505 693.
- 506 Marten, G.G., Reid, J.W., 2007. Cyclopoid copepods. *J. Am. Mosq. Control Ass.* 23(2), 65–  
507 92. [http://dx.doi.org/10.2987/8756-971X\(2007\)23%5B65:CC%5D2.0.CO;2](http://dx.doi.org/10.2987/8756-971X(2007)23%5B65:CC%5D2.0.CO;2).
- 508 Medlock, J.M., Leach, S.A., 2015. Effect of climate change on vector-borne disease risk in  
509 the UK. *Lancet Infect. Dis.* 15(6), 721–730. [http://dx.doi.org/10.1016/S1473-](http://dx.doi.org/10.1016/S1473-3099(15)70091-5)  
510 [3099\(15\)70091-5](http://dx.doi.org/10.1016/S1473-3099(15)70091-5).
- 511 Medlock, J.M., Vaux, A.G.C., Cull, B., Schöffner, F., Gillingham, E., Pfluger, V., Leach, S.,  
512 2017. Detection of the invasive mosquito species *Aedes albopictus* in southern England.  
513 *Lancet Infect. Dis.* 17(2), 140. [http://dx.doi.org/10.1016/S1473-3099\(17\)30024-5](http://dx.doi.org/10.1016/S1473-3099(17)30024-5).
- 514 Micieli, M.V., Marti, G. and García, J.J., 2002. Laboratory evaluation of *Mesocyclops*  
515 *annulatus* (Wierzejski, 1892) (Copepoda: Cyclopidea) as a predator of container-



- 516 breeding mosquitoes in Argentina. Mem. Inst. Oswaldo Cruz. 97(6), 835–838.  
517 <http://dx.doi.org/10.1590/S0074-02762002000600014>.
- 518 Murugan, K., Benelli, G., Panneerselvam, C., Subramaniam, J., Jeyalalitha, T., Dinesh, D.,  
519 Nicoletti, M., Hwang, J.S., Suresh, U., Madhiyazhagan, P., 2015. *Cymbopogon citratus*-  
520 synthesized gold nanoparticles boost the predation efficiency of copepod *Mesocyclops*  
521 *aspericornis* against malaria and dengue mosquitoes. Exp Parasitol 153, 129–138.  
522 <http://dx.doi.org/10.1016/j.exppara.2015.03.017>.
- 523 Nam, V.S., Yen, N.T., Duc, H.M., Tu, T.C., Thang, V.T., Le, N.H., Le Loan, L., Huong,  
524 V.T.Q., Khanh, L.H.K., Trang, H.T.T., Lam, L.Z.Y., Kutcher, S.C., Aaskov, J.G.,  
525 Jeffery, J.A.L., Ryan, P.A., Kay, B.H., 2012. Community-based control of *Aedes aegypti*  
526 by using *Mesocyclops* in Southern Vietnam. Am. J. Trop. Med. Hyg. 86(5), 850–859.  
527 <http://dx.doi.org/10.4269/ajtmh.2012.11-0466>.
- 528 Novich, R.A., Erickson, E.K., Kalinoski, R.M., DeLong, J.P., 2014. The temperature  
529 independence of interaction strength in a sit-and-wait predator. Ecosphere 5(10), 1–19.  
530 <http://dx.doi.org/10.1890/ES14-00216.1>.
- 531 O’Neil, R.J., 1990. Functional response of arthropod predators and its role in the biological  
532 control of insect pests in agricultural systems, in: Dunn, P.E., Baker, R.R. (Eds.) New  
533 directions in biological control: alternatives for suppressing agricultural pests and  
534 diseases. Alan R. Liss, Inc., New York, pp. 83–96.
- 535 R Core Team, 2015. R: A language and environment for statistical computing. R Foundation  
536 for Statistical Computing, Vienna.
- 537 Rall, B.C., Brose, U., Hartvig, M., Kalinkat, G., Schwarzmüller, F., Vucic-Pestic, O.,  
538 Petchey, O.L., 2012. Universal temperature and body-mass scaling of feeding rates.  
539 Philos. Trans. R. Soc. Lond., Series B Biol. Sci. 367(1605), 2923–2934.

- 540 <http://dx.doi.org/10.1098/rstb.2012.0242>.
- 541 Ruybal, J.E., Kramer, L.D., Kilpatrick, A.M., 2016. Geographic variation in the response of  
542 *Culex pipiens* life history traits to temperature. *Parasit. Vectors* 9(116), 1–9.  
543 <https://doi.org/10.1186/s13071-016-1402-z>.
- 544 Siraj, A.S., Oidtmann, R.J., Huber, J.H., Kraemer, M.U.G., Brady, O.J., Johansson, M.A.,  
545 Perkins, T.A., 2017. Temperature modulates dengue virus epidemic growth rates  
546 through its effects on reproduction numbers and generation intervals. *PLOS Negl.*  
547 *Trop. Dis.* 11(7), e0005797. <https://doi.org/10.1371/journal.pntd.0005797>.
- 548 Solomon, M.E., 1949. The natural control of animal populations. *J. Anim. Ecol.* 18, 1–35.
- 549 South, J., Dick, J.T.A., McCard, M., Barrios-O'Neill, D., Anton, A., 2017. Predicting  
550 predatory impact of juvenile invasive lionfish (*Pterois volitans*) on a crustacean prey  
551 using functional response analysis: effects of temperature, habitat complexity and light  
552 regimes. *Environ. Biol. Fish.* <http://dx.doi.org/10.1007/s10641-017-0633-y>.
- 553 Tinson, S., Laybourn-Parry, J. 1986. The distribution and abundance of benthic cyclopoid  
554 copepods in Esthwaite Water, Cumbria. *Hydrobiol.* 131(3), 225–234.  
555 <http://dx.doi.org/10.1007/BF00008858>.
- 556 Toscano, B.J., Rombado, B.R., Rudolf, V.H.W., 2016. Deadly competition and life-saving  
557 predation: the potential for alternative stable states in a stage-structured predator–prey  
558 system. *Proc. R. Soc. Biol.*, pii, 20161546. <http://dx.doi.org/10.1098/rspb.2016.1546>.
- 559 Townroe, S., Callaghan, A., 2014. British container breeding mosquitoes: The impact of  
560 urbanisation and climate change on community composition and phenology. *PLoS*  
561 *ONE*, 9(4), e95325. <https://doi.org/10.1371/journal.pone.0095325>.
- 562 Trexler, J.C., McCulloch, C.E., Travis, J., 1988. How can the functional response best be

- 563 determined? *Oecologia* 76(2), 206–214. <https://doi.org/10.1007/BF00379954>.
- 564 Van Driesche, R., Bellows, T.S., 2011. *Biological control*. Springer, Berlin.
- 565 <https://doi.org/10.1007/978-1-4613-1157-7>.
- 566 Veronesi, R., Carrieri, M., Maccagnani, B., Maini, S., Bellini, R. 2015. *Macrocyclus albidus*  
567 (Copepoda: cyclopidae) for the Biocontrol of *Aedes albopictus* and *Culex pipiens* in  
568 Italy. *J. Am. Mosq. Control Assoc.* 31(1), 32–43. <https://doi.org/10.2987/13-6381.1>.
- 569 Wasserman, R.J., Alexander, M.E., Weyl, O.J.F., Barrios-O'Neill, D., Froneman, P.W., Dalu,  
570 T., 2016. Emergent effects of structural complexity and temperature on predator-prey  
571 interactions. *Ecosphere*, 7(2), e01239. <http://dx.doi.org/10.1002/ecs2.1239>.
- 572 Yee, D.A. 2016. Thirty years of *Aedes albopictus* (Diptera: Culicidae) in America: An  
573 introduction to current perspectives and future challenges. *J Med Entomol.* 53(5), 989-  
574 991. <https://doi.org/10.1093/jme/tjw063>.
- 575
- 576
- 577
- 578
- 579
- 580
- 581
- 582
- 583

584 **Tables**

585 Table 1. Fecundity (%) calculations, entailing proportion of total consumed energy ( $C$ )  
 586 devoted to production of eggs ( $P_r$ ) in adult female *M. albidus* and *M. viridis* across 12 °C, 16  
 587 °C and 20 °C. Data relating to energetics adapted from Laybourn-Parry et al. (1988,  $n = 5$ ,  
 588 Eqn. 6).

Species	Temperature (°C)	$C$ (mJ)	$P_r$ (mJ)	Fecundity (%)
<i>M. albidus</i>	12	31210	2355	7.55
	16	26150	2907	11.12
	20	29150	3691	12.66
<i>M. viridis</i>	12	34433	2851	8.28
	16	25311	3020	11.93
	20	24960	1671	6.70

589

590

591

592

593

594

595 Table 2. Results of logistic regression to denote functional response type across all predator  
 596 and temperature treatments, alongside the starting attack rate ( $a$ ) and handling time ( $h$ )  
 597 parameter estimates generated using the Rogers' random predator equation (Eqn. 5).

Species	Temperature (°C)	First order term	$p$	$a$	$p$	$h$	$p$
<i>M. albidus</i>	12	-0.024	< 0.001	1.276	< 0.001	0.065	< 0.001
<i>M. albidus</i>	16	-0.031	< 0.001	1.917	< 0.001	0.044	< 0.001
<i>M. albidus</i>	20	-0.037	< 0.001	2.373	< 0.001	0.035	< 0.001
<i>M. viridis</i>	12	-0.025	< 0.001	0.945	< 0.001	0.071	< 0.001
<i>M. viridis</i>	16	-0.030	< 0.001	1.896	< 0.001	0.039	< 0.001
<i>M. viridis</i>	20	-0.039	< 0.001	2.186	< 0.001	0.032	< 0.001

598

599

600

601

602

603

604

605

606 Table 3: Mean Relative Control Potential (RCP) using abundances for *M. albidus* and *M.*  
 607 *viridis*, alongside uncertainties reflected through 60% confidence intervals (CI) and  
 608 probability that the RCP output exceeds 1 using maximum feeding and attack rates. Asterisks  
 609 denote significant levels of certainty that the RCP score is greater than 1 (\* > 95%, \*\* > 99%,  
 610 \*\*\* > 99.9%).

Comparators (agent A, agent B)	FR parameter (mean $\pm$ SD)	Abundance (mean ind. m <sup>-2</sup> $\pm$ SD)	RCP	CI $p_{\text{RCP} > 1}$ (%)
<i>M. albidus</i> , <i>M.</i> <i>viridis</i>	1/h: 22.80 ( $\pm$ 8.37), 24.41 ( $\pm$ 10.55)	6727 ( $\pm$ 1018.23), 562 ( $\pm$ 288.50)	16.77	6.79 – 23.78 99.97***
<i>M. albidus</i> , <i>M.</i> <i>viridis</i>	$\alpha$ : 1.98 ( $\pm$ 0.72), 1.77 ( $\pm$ 0.67)	6727 ( $\pm$ 1018.23), 562 ( $\pm$ 288.50)	19.34	8.18 – 27.35 99.99***

611

612

613

614

615

616

617

618

619

620

621 Table 4: Mean Relative Control Potential (RCP) using fecundities (%) across temperature  
 622 change for *M. albidus* and *M. viridis* alongside uncertainties reflected through 60%  
 623 confidence intervals (CI) and probability that the RCP output exceeds 1 using maximum  
 624 feeding and attack rates. Asterisks denote significant levels of certainty that the RCP score is  
 625 greater than 1 (\* > 95%, \*\* > 99%, \*\*\* > 99.9%).

Comparators (agent A, agent B)	Temperature (°C)	FR parameter (mean ± SD)	Fecundity (%)	RCP	CI $p_{\text{RCP} > 1}$ (%)
<i>M. albidus</i> , <i>Me viridis</i>	12	1/h: 15.88 (± 4.36), 13.99 (± 2.67)	7.55, 8.28	1.07	0.77 – 1.34 51.93
<i>M. albidus</i> , <i>Me viridis</i>	16	1/h: 22.12 (± 7.64), 25.98 (± 9.68)	11.12, 11.93	0.90	0.53 – 1.21 32.58
<i>M. albidus</i> , <i>M. viridis</i>	20	1/h: 30.42 (± 5.24), 33.25 (± 6.73)	12.66, 6.70	1.80	1.39 – 2.17 98.21*
<i>M. albidus</i> , <i>Me viridis</i>	12	$\alpha$ : 1.43 (± 0.52), 1.05 (± 0.34)	7.55, 8.28	1.37	0.82 – 1.83 66.71
<i>M. albidus</i> , <i>Me viridis</i>	16	$\alpha$ : 2.17 (± 0.51), 2.02 (± 0.39)	11.12, 11.93	1.04	0.77 – 1.28 49.04
<i>M. albidus</i> , <i>M. viridis</i>	20	$\alpha$ : 2.34 (± 0.75), 2.24 (± 0.54)	12.66, 6.70	2.09	1.39 – 2.69 95.34*

626

627

628 Table 5:  $Q_{10}$  coefficient (Eqn. 7) values associated with mean maximum feeding rates for both  
 629 predators between temperature gradients.

Species	Temperature (°C)	Mean max. feeding rate (1/h)	$Q_{10}$ value
<i>M. albidus</i>	12 – 20	15.88 – 30.42	2.25
<i>M. albidus</i>	12 – 16	15.88 - 22.12	2.29
<i>M. albidus</i>	16 – 20	22.12 – 30.42	2.22
<i>M. viridis</i>	12 – 20	13.99 – 33.25	2.95
<i>M. viridis</i>	12 – 16	13.99 – 25.98	4.70
<i>M. viridis</i>	16 – 20	25.98 – 33.25	1.85

630

631

632

633

634

635

636

637

638

639

640

641

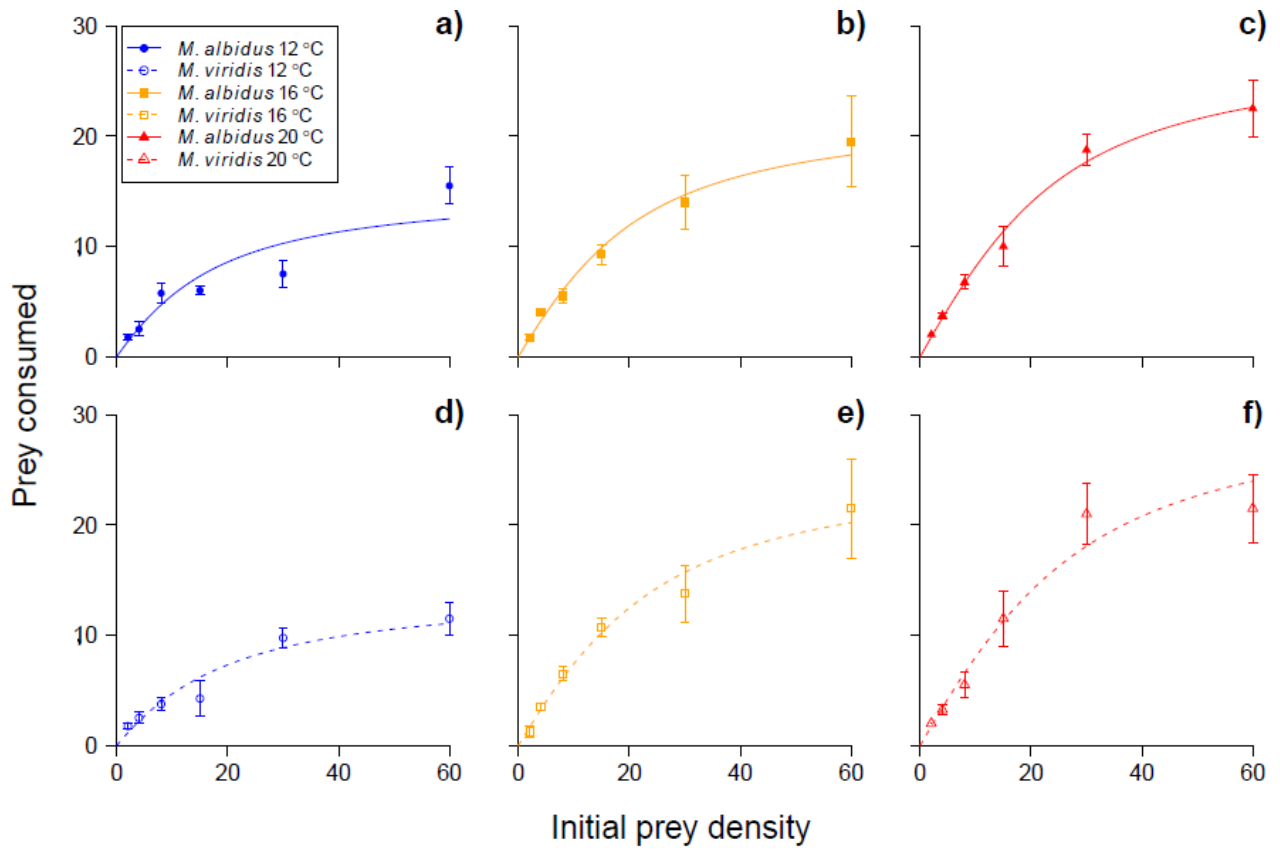
642

643



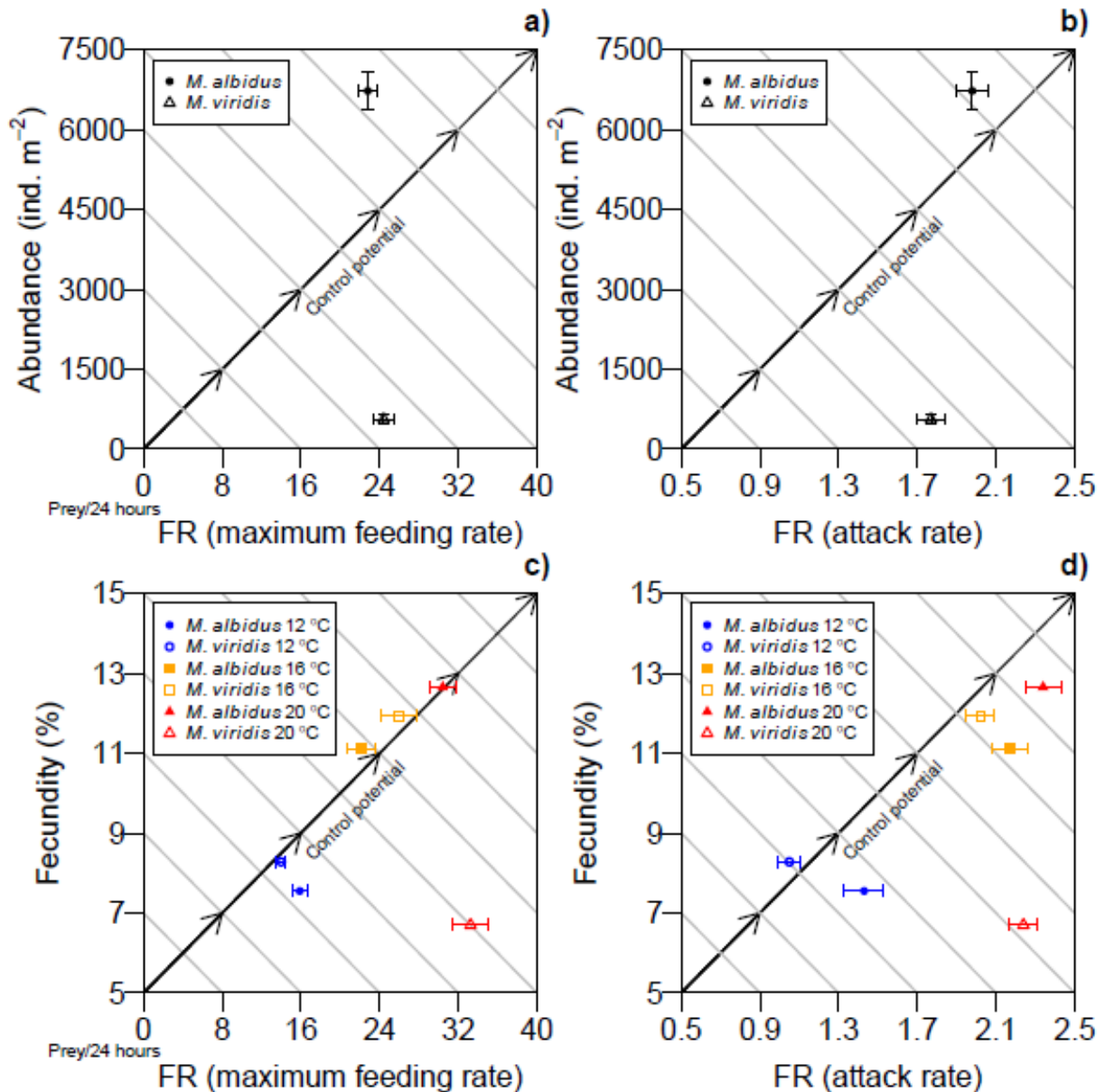
644 **Figures**

645



646

647 Figure 1. Functional responses of *M. albidus* (a, b, c) and *M. viridis* (d, e, f) towards first648 instar *C. pipiens* larvae at 12 °C (a, d), 16 °C (b, e) and 20 °C (c, f) over the 24 hour649 experimental period. Means are  $\pm$  SE at each prey density ( $n = 4$ ).



650

651 Figure 2. RCP biplots comparing *M. albidus* and *M. viridis* using abundance estimates (a, b;  $n$ 

652 = 8) and fecundity calculations (c, d; Table 1), with FR parameters of maximum feeding (a,

653 c) and attack rate (b, d). FR parameters in abundance biplots are pooled bootstrapped

654 estimates across all temperatures ( $n = 90$ ); those in fecundity plots are temperature-specific655 estimates ( $n = 30$ ). Increasing CP is read from bottom left to top right. Abundance and FR656 parameter means are  $\pm$  SE.

657