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Modeling the potential effects of *Plasmodium* infection on the Galapagos penguin (*Spheniscus mendiculus*)

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Abstract

The recently discovered presence of a species of *Plasmodium* infecting the endangered Galapagos penguin (*Spheniscus mendiculus*) potentially threatens their long-term persistence. However, not much information is available on the transmission dynamics of *Plasmodium* in Galapagos or the impact of the parasite on infected penguins. The present work takes the model of the Galapagos penguin population devised by Vargas et al. (2007)—which did not include any impacts from disease—and adds a simple model of infection. Two variables—the probability of an individual becoming infected each year, and the increase in annual mortality caused by infection—define the dynamics of the disease component of the model; the stress from El Niño events could also affect infected individuals in different ways, and so three forms of stress-induced relapse are explored as well. The entirety of parameter space is explored for all three relapse scenarios. All the models show a high impact due to mortality from infection, and there are large parts of parameter space that have a 0% probability of persistence over the next 100 years. The probability of persistence decreases substantially if relapse events occur during all El Niño events, weak and strong. Increasing the breeding success of the population provides a modest benefit, but does not reverse the overall trend. In order to estimate the mortality that might be associated with *Plasmodium* infection, a comparison was made between census data from 1998–2009 and model predictions based on these same years. The models differed in their level of mortality from infection, and a range of plausible parameter values was determined from the best-fitting models; these ranged from 0–10% to 0–15%, depending on the type of relapse modeled. Even at these

relatively low levels of impact, *Plasmodium* infection still has the potential to drastically reduce the probability of persistence of the penguin population over the next 100 years.

Introduction

It has long been feared that avian malaria would find its way to the Galapagos Islands (Wikelski et al. 2004). The introduction of avian malaria and a suitable vector have been implicated in the extinction of several endemic bird species in Hawaii over the past century (Warner 1968; Atkinson et al. 1995). Like Hawaii, the Galapagos Islands are home to many small populations of endemic birds, long isolated from the mainland (Harris 1973). These factors make disease-induced extinctions more likely (Castro and Bolker 2005). Therefore, the discovery by Levin et al. (2009) of a *Plasmodium* species infecting the endangered Galapagos penguin (*Spheniscus mendiculus*) represents a serious threat—not just to this particular species, but to the avifauna of the Galapagos Islands as a whole.

The Galapagos penguin is endemic to the archipelago, with a small population of about 1800 individuals (F. H. Vargas, pers. comm.), down from an initial survey of 4000 individuals in 1970 (Vargas et al. 2005). Although the population experiences positive growth over the short term, the species has been experiencing a long-term decline over the past 40 years (Vargas et al. 2005). This is driven by the periodic occurrence of intense El Niño events. These events cause precipitous population declines, with over 50% reductions in the population, believed to be associated with reduced food availability (Vargas et al. 2006). In addition to reducing the size of an already small population, a stressful event such as El Niño could also worsen the effects of malaria infection

(Atkinson and van Riper 1991; Valkiunas 2005). Another cause for concern is that related species of *Spheniscus* penguins in captivity have shown high susceptibility to and mortality from *Plasmodium* infection (Stoskopf and Beier 1979; Cranfield et al. 1990; Fix et al. 1988). This vulnerability may be exaggerated in the Galapagos penguin population due to its low major histocompatibility complex (MHC) variation, which is an indicator of immune system strength against disease (Bollmer et al. 2007). The penguins' small population, their potential vulnerability to *Plasmodium* infection, and the periodic occurrence of devastating El Niño events all suggest that the Galapagos penguin could be severely threatened by the presence of malaria in Galapagos.

Unfortunately, little is known about the characteristics of malaria in Galapagos or how the penguins respond to it. Infected penguins were initially discovered by using PCR to amplify parasite DNA within samples of the host's blood. Using this technique, infected penguins have been found on the islands of Isabela, Fernandina, and Santiago, with a total prevalence of 5% (Levin et al. 2009). However, an enzyme-linked immunosorbent assay (ELISA), which is used to detect previous exposure to *Plasmodium*, was conducted on a subset of the same birds and found about 95% of the sample had been exposed (Palmer et al. in preparation). There is also reason to suspect that the Galapagos penguin is an unsuitable host for *Plasmodium*, due to the lack of observations, under the microscope, of the terminal stages in the life-cycle of the parasite—even in birds that have been recaptured and shown infection at multiple points in time (Parker et al. in prep.).

There are still many aspects of the situation that are currently unknown. Some of this information includes: How lethal is *Plasmodium* infection for the Galapagos

penguin? What mosquito species is the primary transmission vector, and what is the prevalence of infection in the vector? How exposed are the penguins to the vector? And are there other infected bird species that may act as reservoirs of infection? All of these parameters will be important in understanding how critical the situation in the Galapagos Islands is, and in determining how to manage the islands to minimize the risk of avian extinctions.

Investigating these factors will take time, and meaningful conservation decisions need to be made in the meantime to manage this vulnerable species. A useful tool in making these decisions is Population Viability Analysis (PVA), a technique used to predict the probability of extinction for a population by utilizing a stochastically driven computer simulation of future population growth (Possingham et al. 1993). It has been successfully used in predicting viability for some species (Brook et al. 2000; but see Ellner et al. 2002, Coulson et al. 2001). A previous PVA for the Galapagos penguin was conducted by Vargas et al. (2007) using the simulation program Vortex (Lacy et al. 2010). The presence of *Plasmodium* in the population was unknown at that time, so their model focused on how the pattern of El Niño events might influence the probability of persistence of the penguin population. Under the current frequency of El Niño events, they predicted a 70% probability of persistence over the next 100 years for the penguin population. They found that the less frequent but more damaging strong El Niño events have a greater impact on the penguin population than the more frequent but weak El Niño events. Another important factor is the adult mortality rate, with rates over 5% being especially damaging. It is unknown, though, how reliable these results are in the face of malaria's presence in Galapagos.

The present work extends the model of Vargas et al. (2007) to include a disease component in the analysis. We explore the possible consequences of malaria's introduction on the penguins' long-term probability of persistence and use the modeling framework to estimate a range of plausible parameter values for mortality from *Plasmodium* infection. Box 1 provides a list of terminology used in this paper.

Methods

The model was made in Vortex, version 9.99b (Lacy et al. 2010; Lacy 2000). Vortex is a stochastic, individual-based modeling program used for population viability analysis (PVA) (P. S. Miller and Lacy 2005). This type of modeling includes the effects of variable demography, environmental conditions, and rare catastrophes, instead of purely deterministic factors alone. The inclusion of these semi-random events means that a single run of the model gives only one possible outcome for the population, so each scenario (combination of parameter values) is run 1000 times to create a distribution of outcomes. Each run simulates the penguin population 100 years into the future, in one-year increments. The probability of population persistence can be calculated as the proportion of the 1000 runs that predict an intact population after 100 years, with an intact population being defined as having at least one member of each sex still alive. Vortex is highly customizable, allowing parameters in the model to be functions of other parameters or of user-created variables. The framework shared by all of our modeled scenarios has three general components: the demographic parameters of the penguin population, the occurrence of El Niño events, and the dynamics of malaria infection.

Penguin Demographics

The penguin demographic parameters are the same as used in the Vargas et al. (2007) paper's Current El Niño (CEN) model (parameters given in Table 1), with two exceptions. First, the initial population size is now set at 1800 individuals, in accordance with the penguin population census estimate from 2009 (H. Vargas, pers. comm.; see Vargas et al. 2005 for details on the census estimation technique). The sizes of the four island subpopulations are taken to be proportional to the population sizes reported in Table 2 of Vargas et al. (2007). Second, the mortality rates for each age class are now functions that take the infection status of an individual into account (see *Disease States* below, and Appendix A for details).

El Niño Events

As in Vargas et al. (2007), there are two types of El Niño events included in the model, strong and weak El Niño events (Vargas et al. 2006), and they are treated as 'catastrophes' in Vortex that occur randomly with a set probability. The frequency of occurrence for El Niño events is the same as the previous model (Table 1), but their duration and severity have been slightly altered. An oversight in the Vargas et al. (2007) model's implementation allowed both types of El Niño to occur in the same year, which has been fixed. Also, strong El Niño events are now modeled as two-year events, with differing severity for each year, to more closely match the dynamics of observed El Niño events. See Appendix A for details on these changes.

Disease States

Each individual penguin in the model can be in one of three disease states at any time: susceptible, acutely infected, or chronically infected. Susceptible individuals are not infected with *Plasmodium*, and so experience normal rates of mortality. Individuals become acutely infected for the first year after contracting malaria, and they experience increased mortality due to their infection. If an individual survives the first year of infection, they then become chronically infected; infection with *Plasmodium* can lead to persistent, long-term infections (Valkiunas 2005). Chronic infections are considered to be under control and so infected individuals in the model do not experience any increased mortality due to their infection, except under certain circumstances (see *Relapse Scenarios* below).

Variables of Infection

There are two variables that control the spread and severity of infection of malaria in the model. The probability of infection gives the probability each year that a susceptible individual will become infected. The pathogenicity variable is the increase in the probability of mortality that an individual experiences due to infection.

Relapse Scenarios

It is believed that individuals in high stress situations can become immunocompromised, leading to a worsening of symptoms from an existing infection (Atkinson and van Riper 1991; Valkiunas 2005). El Niño events have the potential to be stressful events for the penguins, as they are believed to be food-limited during these events due to changes in the Cromwell Current system leading to reduced fish numbers

(Vargas et al. 2006). However, it is unknown how infected penguins will respond to different El Niño conditions; they may suffer a recurrence of their symptoms (hereafter referred to as a relapse) during some events or be unaffected. So, three separate modeling scenarios of relapse have been considered: a scenario where no relapses occur, one where relapses occur during strong El Niño events, and one where relapses occur during all El Niño events, weak and strong. In all three scenarios, whenever a relapse-triggering event occurs, chronically infected individuals experience increased mortality due to their infection according to the pathogenicity for the current model (see *Exploration of Parameter Space* below). The model does not take into account the possibility of pathogenicity changing between the acute infection phase and subsequent relapses, due to either increased resistance in the host or increased susceptibility from the stressful El Niño conditions.

Exploration of Parameter Space

In order to assess the possible effect of malaria on the penguin population, the two variables that define the malaria dynamics—the probability of infection and pathogenicity—were varied over a range of possible values. Each variable could take on values from 5% to 100%, in 5% intervals, along with a baseline model that did not include any disease component; each unique combination of parameter values is a separate model of 1000 runs. In addition, this entire parameter space was individually analyzed for each of the three relapse scenarios described above. This resulted in the analysis of 1201 separate models (400 for each relapse scenario, plus the baseline), for a

total of 1,201,000 runs. For each model, the probability of population persistence over the next 100 years was recorded.

Model Assumptions

All models are built with assumptions, and it is important to be explicit about them, as much as possible. Some assumptions of the disease component of the model are:

- The probability of infection is the same for every individual, in every year, and on every island (within a set of 1000 runs for any model).
- The pathogenicity of infection is the same for the initial and subsequent episodes.
- Relapses only occur during El Niño years, if at all. This ignores other sources of stress that could lead to relapses, such as molting or reproduction (Richner et al. 1995).
- The dynamics of the vector(s) or possible reservoirs are not taken into account.

Effect of Increased Breeding Success on Probability of Persistence

In their previous modeling work in this system, Vargas et al. (2007) made several suggestions for management actions to increase the probability of persistence for the Galapagos penguin. One suggestion was to increase the percentage of females that successfully breed in a year. To determine how effective this strategy might be in the face of malaria's presence in the population, the analysis of 1201 models discussed above was repeated, but with the adult female breeding success increased by 10%, from 56.7% to 66.7%.

Estimation of Parameter Values

In order to assess the current threat posed by *Plasmodium*, as opposed to the range of possible outcomes, it is necessary to estimate values for the malaria variables, the probability of infection and pathogenicity. The ELISA performed by Palmer et al. (in preparation) found around 95% exposure of sampled penguins to *Plasmodium*. A non-systematic survey of the models' results suggests that the individual probability of infection is similar to the population's level of exposure, with exposure being a few percent lower than the probability of infection set for a particular scenario (unpublished data). The 95% exposure rate from the ELISA is therefore taken as an estimate of the probability of infection in the analysis on pathogenicity below.

The most reliable method for determining the pathogenicity of a strain of parasite on its host is to conduct experimental infections of parasite-free individuals (Valkiunas 2005). In the absence of such data, we have used our modeling framework to estimate a range of plausible parameter values. By running several scenarios that differ only in the value of the pathogenicity parameter, and comparing their output to actual data, the scenarios that best fit the data provide the best estimates for the pathogenicity.

The data used to assess the scenarios' fit were the penguin population census data from 1998–2009 (Vargas et al. 2005; H. Vargas pers. comm.), specifically the growth rate from one year to the next, calculated as $r=N(t+1)/N(t)$. Comparing the population size each year would be inappropriate, because the size of the population in one year is dependent on the size in previous years. The scenarios were run for 11 years, instead of 100 years as in the main model. These 11 years represented those following the last strong El Niño event; choosing these years avoids complicating the analysis with the

possible interactions between infection, pathogenicity, and strong El Niño events, while still allowing enough data points for comparisons to be made. The years corresponding to 2006, 2008, and 2009 were modeled as weak El Niño years, determined using sea-surface temperature data from the Charles Darwin Foundation Climate Database and the definition of 'weak El Niño' given in Vargas (2006). The demographic parameters of the model were kept the same, except that the starting population size was set to 780, the population estimate from 1998 (Vargas et al. 2005).

To assess the fit of each scenario, the difference between the population growth rate of the model, averaged over all 1000 runs, and the census data was found for each year, and the differences were then averaged across all years. The scenarios included in this analysis had pathogenicity values from 5–30% in 5% intervals, along with a model that did not include any disease component. The decision to stop at 30% was arbitrary. For each value of pathogenicity, scenarios were run from 80–100% probability of infection in 5% intervals; however, for each level of pathogenicity, the results were very similar for all levels of the probability of infection, and so they were averaged together. This whole analysis was repeated for the scenario without relapses and the scenario with relapses during all El Niño events.

Although only one parameter value will give the best fitting model to the data for each relapse scenario, other values may not be significantly worse fits to the data. Using normal statistical tests, such as two-sample *t*-tests, to determine which models were significantly different would not be appropriate; the standard error in the fit of the models could be arbitrarily increased or decreased by changing the number of model runs. Instead, we took the 1000 runs of the best-fitting model and calculated the fit of each

individual run to the census data. The average fit of the other scenarios can then be compared to this statistical distribution, to determine which scenarios are significantly worse fits to the data. The Bonferroni-corrected significance level used for this comparison was $\alpha = 0.05/12 = 0.0042$, to correct for multiple comparisons across both relapse scenarios.

Results

Figure 1 shows the probability of persistence, after 100 years, across parameter space for all three scenarios of relapse. In all three scenarios, increasing pathogenicity leads to a steep decline in the probability of persistence, while increasing the individual probability of infection causes a less severe decline. Increasing both parameters causes a rapid decrease. There are large portions of the parameter space that show a 0% probability of persistence after 100 years. The scenarios without relapses and with relapses during strong El Niño events have similar shapes across parameter space, while the scenario with relapses for all El Niño events predicts a smaller probability of persistence for all parameter values. Figure 2 shows that increasing the proportion of adult females successfully breeding by 10% leads to an increase in the probability of persistence, and in a similar fashion for all three relapse scenarios. When the value of at least one of the disease parameters is kept low, there is a modest increase in the probability of persistence. When both parameters increase, though, the benefits of increasing the breeding success quickly decline, and there are still large areas of parameter space that have a 0% probability of persistence.

The scenario that best fit the census growth rates was the scenario that did not include any effect of *Plasmodium* infection (Figure 3a). The no-disease scenario closely predicted the average trend in the actual growth rate, but only if the strong recovery of the population in 1999, immediately after the previous strong El Niño, is not included (Figure 3b). When the scenarios with malaria are compared to the distribution of the best-fit model, the following parameter values fall within the rejection region: 20% pathogenicity when relapses do not occur in the model, and 15% pathogenicity when relapses occur during all El Niño events. This means that pathogenicity values up to 15% and 10% respectively are not significantly different from the best-fitting scenario. Figure 4 shows the probability of persistence predictions for the next 25, 50, and 100 years for the plausible values of pathogenicity.

Discussion

Our modeling work provides the first estimate for the pathogenicity of *Plasmodium* infection in the Galapagos penguin. Under both the relapse scenarios considered, the estimated levels of mortality associated with infection were relatively low. This is consistent with what information is available on *Plasmodium*'s presence in Galapagos. The Galapagos penguin population has continued to grow since 2003 (Vargas et al. 2005; F. H. Vargas pers. comm.), despite near-ubiquitous exposure to *Plasmodium* by the penguins (Palmer et al. in preparation). Over this period of time, ten individuals have been found and recaptured that were PCR-positive at both times (Levin et al. 2009; unpublished data). Five of these individuals have survived for three or more years. This all suggests that at least some of the penguins are suffering only minimal effects from

326 malarial infections. However, the recaptures and estimates of malaria exposure were all
327 done starting in 2003, several years after the latest strong El Niño event. It is possible that
328 infection is relatively benign under most conditions (as our results suggest), but that
329 pathogenicity is high during strong El Niño events. Our models do not investigate this, as
330 they assume that the pathogenicity of infection is the same for the initial, acute infection
331 and for subsequent relapses.

332 It is possible, though, that malaria has played a role in previous penguin
333 population crashes. A reanalysis of G. D. Miller et al.'s (2001) penguin blood samples,
334 taken in 1996, found one infected penguin out of 109 retested (Parker, pers. comm.),
335 showing that *Plasmodium* was present in the Galapagos penguin population during the
336 1997–1998 strong El Niño. We do not know when *Plasmodium* arrived in the Galapagos
337 Islands, leaving open the possibility that it was also present during the 1982–1983 strong
338 El Niño as well, but undetected. The heavy mortality observed during strong El Niño
339 events may, in part, be due to the presence of infected penguins that cannot cope with the
340 stressful conditions. However, with only two recent strong El Niño events to consider,
341 and no information on pathogenicity or prevalence during these events, it is difficult to
342 draw any conclusions on the matter. Observational studies of penguins during the next
343 strong El Niño event, undertaken alongside continued disease monitoring, will shed light
344 on this issue.

345 It appears that the type of relapse that may occur during El Niño events, and even
346 the pathogenicity of infection, does not have an appreciable effect on the probability of
347 persistence over the short term (Figure 4). It is only when considering the population's
348 persistence over 50 or 100 years that differences in these parameters lead to different

predictions for the population's fate. The presence of *Plasmodium* in the Galapagos penguin population represents a serious, long-term threat.

Looking at the whole of parameter space, and not just at the range of most plausible values, allows us to consider the effect of reducing one variable or the other on the 100-year probability of persistence. For instance, would conservation effort be more effectively spent on reducing disease transmission or pathogenicity? Somewhat counter-intuitively, given the near ubiquity of exposure to *Plasmodium*, each percent decrease in pathogenicity has a greater impact than a similar decrease in the individual probability of infection. However, the effectiveness of an intervention also depends on how cost effective it is; the expense and logistical difficulty of protecting already infected penguins from the effects of their disease may be prohibitive compared to a campaign to greatly reduce—or eliminate—transmission to susceptible individuals.

Conclusions

Under normal conditions during non-strong El Niño years, pathogenicity from malaria for the Galapagos penguin is likely low; even so, over the long term even low levels of mortality from disease can lead to a high likelihood of extinction if exposure is high. Increasing the breeding success of the population can increase the probability of persistence for a time, but does not alter the overall potential effect of malaria on population persistence. We have very little information on how infected penguins react to strong El Niño events, which could greatly affect the accuracy of our predictions. Future modeling projects in this system will also require more details on the roles of malaria vectors and reservoir species, factors which the present work did not account for.

Despite the gaps in our knowledge, our work highlights the flexible and modular nature of population modeling research. A population viability analysis (PVA) will be accurate only if it contains reliable information on all of the relevant threats to a population's persistence; this will be possible only for the most well studied systems, if at all. However, models can be updated as new threats are discovered, reparameterized as new data are gathered, and rerun when novel analyses are devised. There are benefits to modeling, despite its limitations and biases. In order to maximize these benefits, we recommend that modelers design their PVAs to facilitate future modification. How the model is programmed and implemented will have an effect on this, and extensive documentation of what the model is doing, and why, is essential. This allows other researchers to replicate or expand upon models already developed.

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Tables and Figures

Box 1: Terminology used in this paper

Scenario – a single scenario refers to the specific combination of parameters used for a particular simulation. A scenario may also refer to a set of models that use the same structure for relapses.

Modeling framework – the equations used to define how the simulation runs.

Year – one increment of time in the model; the current population size, births, deaths, catastrophes, etc. are used to calculate the new population size after that year. Each scenario runs for 100 years.

Run – an independent, 100-year calculation of the model; the probability of persistence is calculated from 1000 runs of the model.

Stochastic events – events that occur semi-randomly; they do not have the exact same value in each run of the model.

Probability of persistence – the proportion of runs that end with an intact population (defined as one or more individuals of each sex remaining alive).

Probability of infection – the probability of a susceptible individual becoming infected each year.

Susceptible – an individual in the model that is not infected with *Plasmodium*.

Acute infection – an individual in the model that is infected with *Plasmodium* and is currently experiencing symptoms of disease (increased mortality).

Chronic infection – an individual in the model that is infected with *Plasmodium*, but is otherwise not affected.

Pathogenicity – the probability of an individual dying in a year when acutely infected or suffering a relapse, in addition to (added to) their mortality rate without disease.

Relapse – a reoccurrence of symptoms (i.e., pathogenicity) in a chronically infected individual, triggered in the model by El Niño events.

Strong El Niño – a two-year event that reduces the penguins' survival and reproductive success.

Weak El Niño – a one-year event that reduces only the penguins' reproductive success.

Table 1: Basic Parameters of the Model. Adapted from Vargas et al. (2007)

Parameter / variable	Basic Model

Number of iterations	1000
Number of years	100
Extinction definition	One sex remains
Number of populations	4
Inbreeding depression	No
Correlation of demographic rates among subpopulations	0.9
Concordance of variation in reproduction and survival	Yes
Breeding system	Long-term monogamy
Number of types of catastrophes	2
Dispersing age range (youngest-oldest)	(1-1)
Dispersing sex(es)	Both
Percent survival of dispersers	80
Dispersal rates	*
Age of first offspring for females	3
Age of first offspring for males	3
Maximum age of reproduction	20
Maximum number of broods per year	1
Sex ratio at birth (% male)	50%
Annual reproductive rates	
% adult females breeding	56.7
Annual variation in % breeding	SD = 13
% females producing 1 progeny	33.5
% females producing 2 progeny	46.4
% females producing 3 progeny	12.4
% females producing 4 progeny	7.7
Mortality rates (same for both sexes)	
% mortality between ages 0 and 1	67
Annual variation in % 0-1 mortality	SD = 10
% mortality between ages 1 and 2	25
Annual variaiton in % 1-2 mortality	SD = 5
% mortality between ages 2 and 3	5
Annual variation in % 2-3 mortality	SD = 3
% mortality after age 3	5
Annual variation in % 3+ mortality	SD = 3
Catastrophe 1: Strong El Niño	
Frequency	5%
Multiplicative impacts on reproduction, survival	†

Catastrophe 2: Weak El Niño	
Frequency	20% ‡
Multiplicative impacts on reproduction, survival	0.8•, 1.0
% males in breeding pool	100%
Initially at stable age distribution?	Yes
Initial population size	1800**
Carrying capacity (K)	4200
SD in K due to environmental variation (EV)	420
Harvest	No
Supplementation	No

*See Vargas et al. (2007)

†See Appendices A and B

•The multiplicative impact on reproduction was incorrectly reported as 0.2 in Vargas et al. (2007) (R. Lacy, pers. comm.)

**The population size of each subpopulation used the same 'mean percent of population' reported in Table 2 of Vargas et al. (2007)

‡A weak El Niño won't occur in the same year as a strong El Niño; see Appendix B

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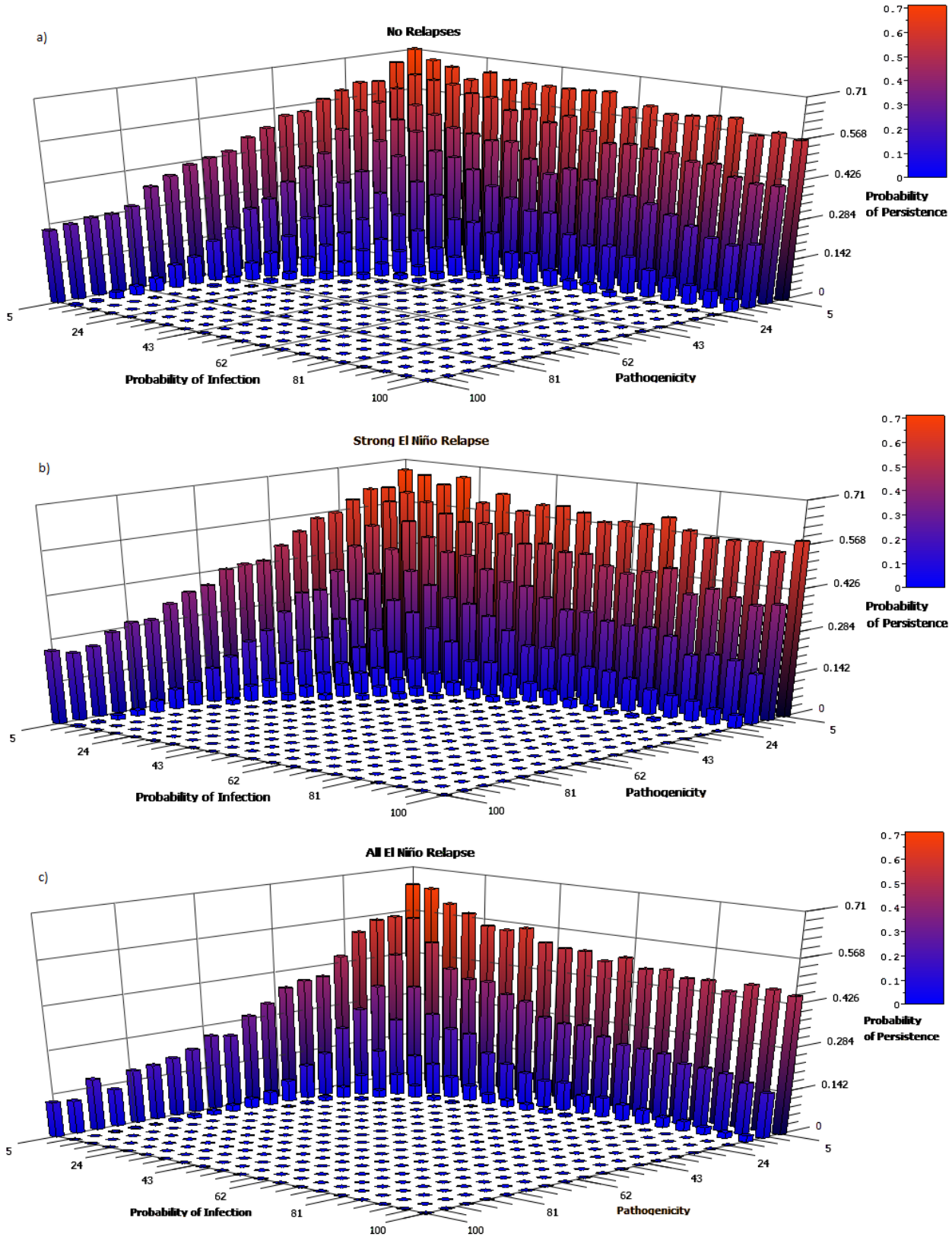
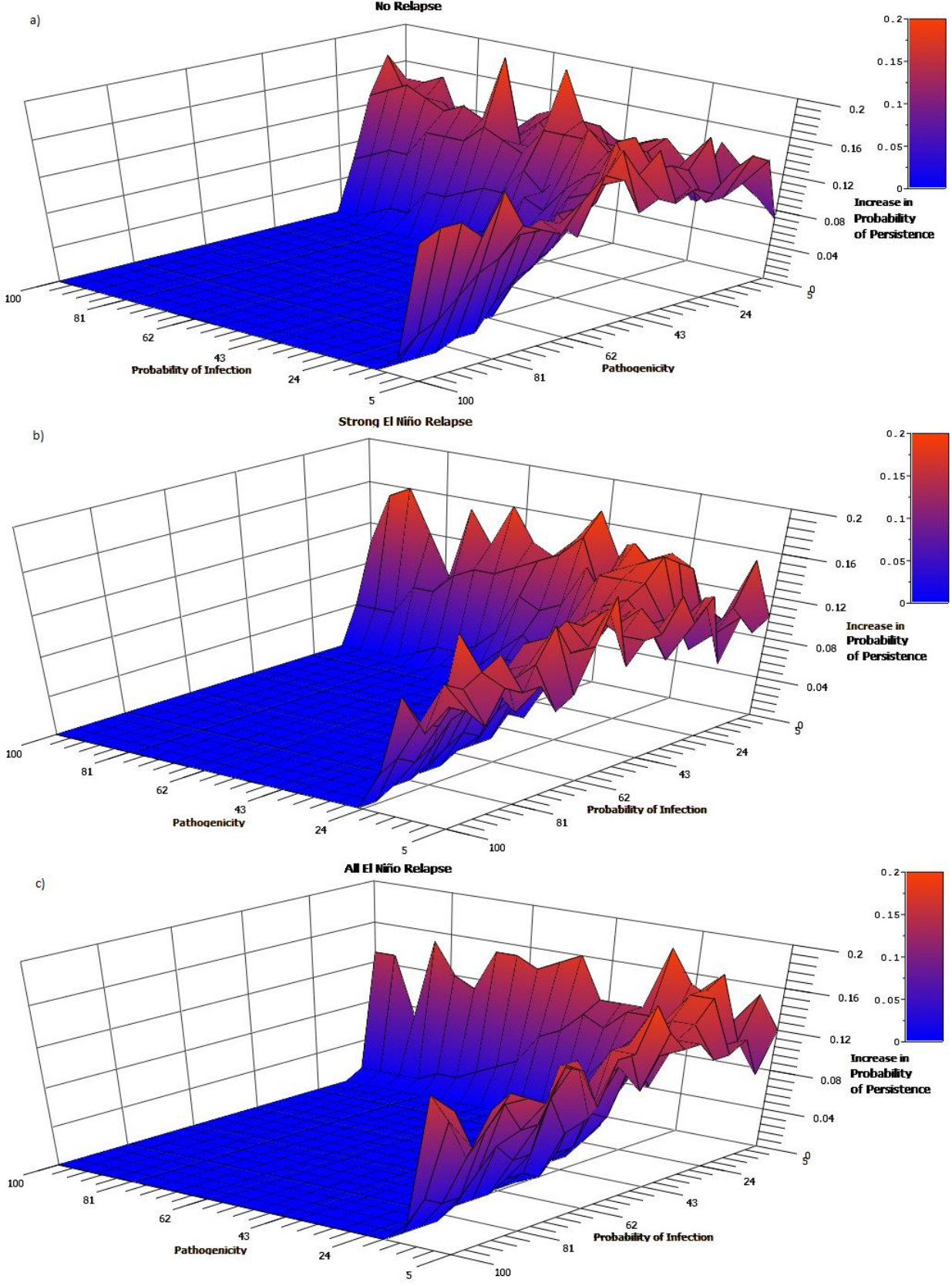

a)
No Relapses
b)
Strong El Niño Relapse
c)
All El Niño Relapse
Probability of Infection
Pathogenicity
Probability of Persistence
5
24
43
62
81
100
0
0.142
0.284
0.426
0.568
0.71

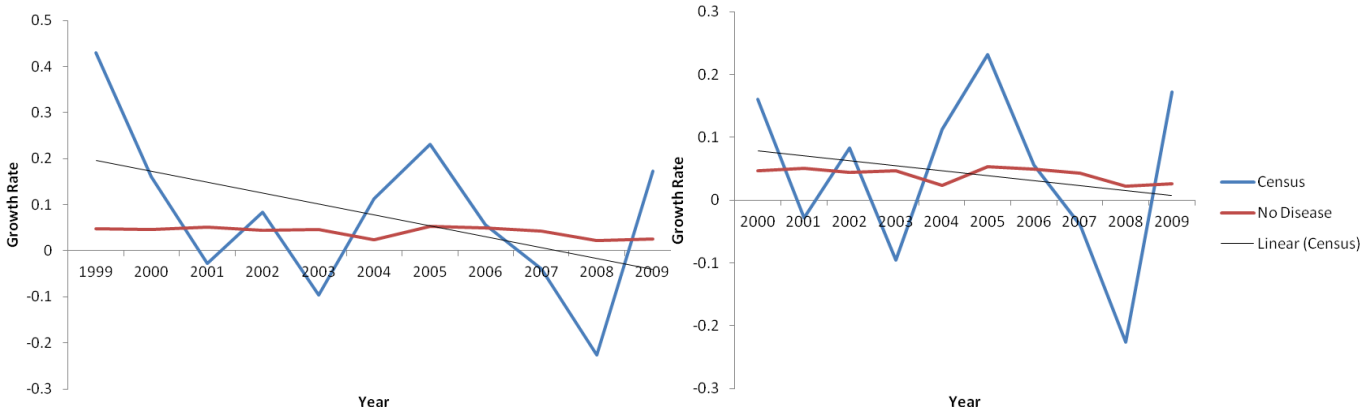
547
548

549 Figure 1: Mean probability of persistence for the next 100 years under the (a) scenario
550 without relapses, (b) scenario with relapses during strong El Niño events, and (c) scenario
551 with relapses during all El Niño events. Each graph shows the probability of persistence
552 for each combination of parameter values (probability of infection and pathogenicity),
553 starting at 5% and increasing in 5% intervals. The baseline model used by Vargas et al.
554 (2007) gave a probability of persistence of 70%.



555
556
557 Figure 2: The improvement over the base model due to increasing the proportion of adult
558 females successfully breeding by 10%, for (a) the scenario without relapses, (b) the
559 scenario with relapses during strong El Niño events, and (c) the scenario with relapses
560 during all El Niño events.

561



562
563

564 Figure 3: Yearly growth rates (in blue) and a linear regression (solid line) for the penguin
565 census data, and the average predicted growth rates over 1000 runs for the best-fitting
566 model, with no disease component (in red). The predictions of the no-disease model are
567 similar to the linear trend of the census data, but only if the high growth in 1999 is not
568 included.

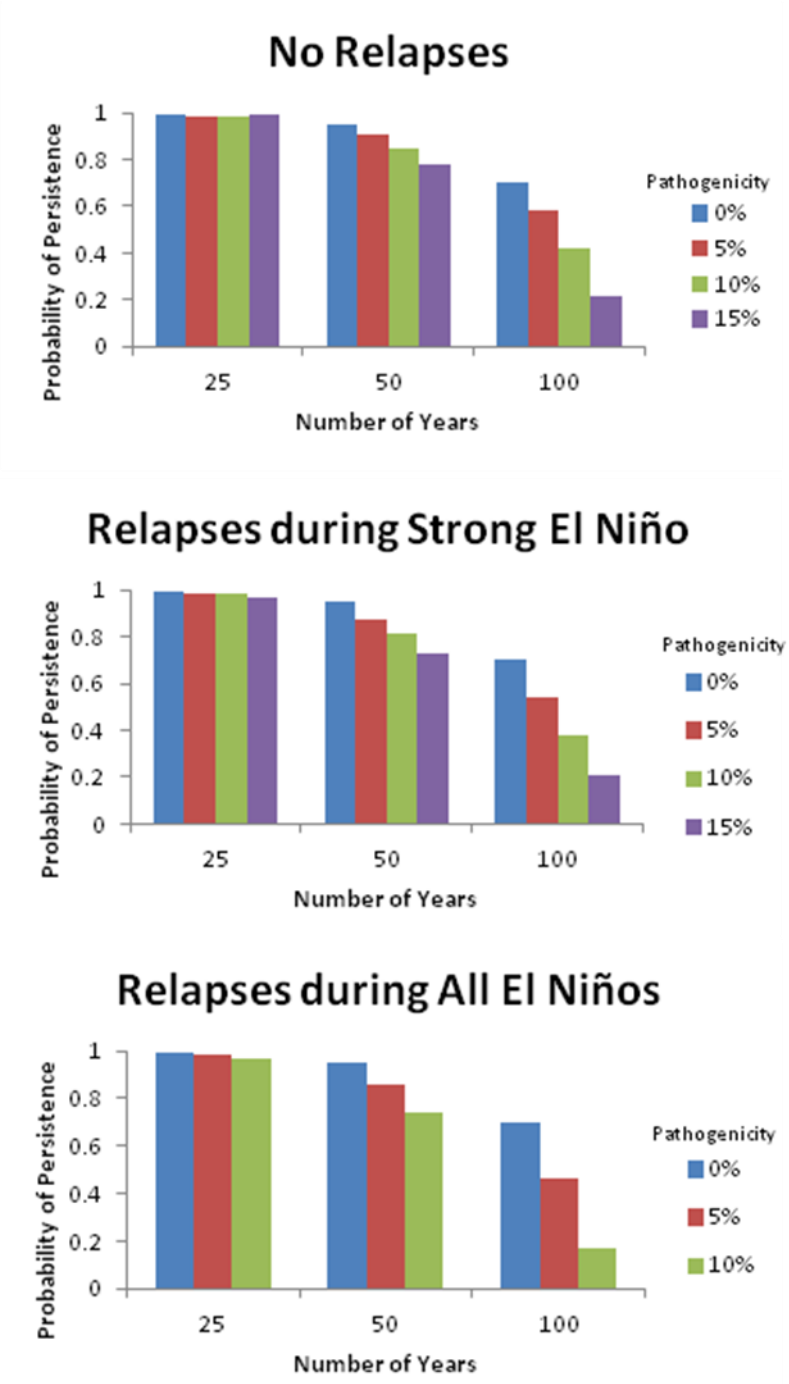


Figure 4: Probability of persistence over the next 25, 50, and 100 years for the range of plausible parameter values for pathogenicity for the scenarios (a) without relapses (0–15% pathogenicity), (b) with relapses during strong El Niño events (0–15%), and (c) with relapses during all El Niño events (0–10%). For (b), the plausible mortality values given are the same as for (a), as they have the same relapse schedule for the years tested here. All scenarios with malaria included had the probability of infection set at 95%. The data for 100 years are the same as reported in Figure 1.

Appendices

Appendix A: Duration and Severity of Weak and Strong El Niño Events

In Vortex, catastrophes (such as the two types of El Niño events) are handled as rare, one-year events that can alter the survival probability and/or reproduction of individuals in the population. While Vargas et al. (2007) used constants for these values, the present model treats them as functions to achieve two ends: to prevent overlapping of El Niño events (eqtn. 15 in Appendix B), and to alter the duration and severity of strong El Niños to more closely match observations (eqtns. 3–4, 6, 12–14). Vortex treats each catastrophe as an independent event, and so, using the default settings, a weak and a strong El Niño event could occur simultaneously in the model. This would happen with a probability of (0.05 probability of strong El Niño * 0.2 probability of weak El Niño = 0.01) per year. Having both types of events occurring together could affect the frequency of relapse events in the different model scenarios.

Two strong El Niño events have been observed over the past 47 years, in 1982–1983 and 1997–1998, which lasted for 18 and 17 months respectively (Vargas 2006). The intensity of these events followed a bell-shaped curve—they started off as a weak El Niño, intensified into a strong El Niño for about a year, then tapered off into another weak El Niño. This contrasts with the representation of strong El Niño events in the Vargas et al. (2007) model, as one-year events with a single level of severity. The present modeling work treats strong El Niños as two-year events (eqns. 6, 12). Each time a strong El Niño event occurs, one year is randomly chosen (using eqns. 3–4) to be the year with the higher severity, while the other year takes into account the effect of the months of weaker El Niño activity (eqns. 13–14). The stronger year has the same impact on the population as the strong El Niño event used in Vargas et al. (2006)—reproduction is reduced to 1% of its normal rate, and survival is reduced to 30% of its normal rate. The weaker year has no effect on survival and reduces reproduction to 90% its normal rate. A full weak El Niño event, unassociated with a strong El Niño, reduces reproduction in the model to 80%; the weak activity before and after the main El Niño effect will in most cases likely overlap with only part of the breeding season, and so the effect in the model has been halved to account for this.

Appendix B: Equations Used in Vortex to Define the Model

These equations are presented in the same form that they were input into Vortex. In Version 9.99b, there are three types of user-created variables: Global State (GS) Variables, Population State (PS) Variables, and Individual State (IS) Parameters. Each of these types of variables requires different inputs or functions to determine their behavior. GS variables require a function that specifies its value for the first year of the simulation (the initial function), and how the value of the variable changes from one year to the next (the transition function). GS variables operate at the level of the metapopulation. PS variables only use a transition function, and they operate independently for each subpopulation. IS variables are assigned to every individual in the population; in addition

to the initial and transition functions, IS variables also have a function that determines their value for new-born individuals. For the mortality rates and catastrophe frequency/severity, these equations are functions of the original variables of the model.

Global State Variables

1) GS1: Probability of infection – Initial function is a proportion between 0 and 1.0. Transition: =GS1.

> This variable represents the probability of an individual becoming infected in a single year. This variable is included for ease of data entry; the variable PS1 (eqn. 5) is what is actually used for transmission in the equations.

2) GS2: Pathogenicity – Initial function is a whole number between 0 and 100. Transition: =GS2.

> In the model, an infected individual has an increased probability of dying in some years. This variable gives the amount of that increase; for example a mature individual normally has a 5% probability of dying in a year, but an infected individual has a (5+GS2)% probability of dying. The total probability of an individual dying is capped at 99% (see the Mortality Rates section, eqns. 9–11).

Global state variables 3–5 are used for book-keeping, but not as part of the model itself.

3) GS6 – Initial and Transition: =SRAND((R*100)+Y)

4) GS7 – Initial and Transition: =SRAND((R*100)+(Y-1))

> A strong El Niño event in this model has two strengths, one for each of its years—a strong effect [the same as the strong El Niño event in the Vargas et al. (2007) model] and a weak effect [corresponding to the build-up and settle-down time surrounding a strong El Niño event; see Appendix A]. These functions are used by the Catastrophe functions (eqns. 13 and 14) to determine the order in which these two effects occur. Note that GS7 returns the same result as GS6 from the previous year.

Population State Variables

5) PS1: Probability of infection =GS1

> This variable represents the probability of an individual becoming infected in a single year. It is the same for each subpopulation, and equals the value given in eqn. 1.

6) PS2: =(CAT(1)<1)*(PS2<1)

This function is what causes a strong El Niño event to take two years instead of one (see eqn. 12). During the first year of the strong El Niño, PS2=1, then it reverts to PS2=0 after the second year.

Population state parameters 3–5 are used for bookkeeping, similar to the global state parameters 3–5.

Individual State Parameters

7) IS1: Chronic Infections – Initial: =(RAND<0.90) Birth: =0. Transition: =IS2

In this model, an individual is assumed to retain their infection for life. The initial, acute infection period causes increased mortality, while the chronic infection is considered to be under control (except when relapses are allowed during El Niño events; see eqns. 9–11 below). The acute stage lasts for only the first year of an individual's infection in this model. At the beginning of each run (at Year 0), 90% of the population is chronically infected, in accordance with the *Plasmodium* exposure found by Palmer et al. (in preparation). See also eqn. 8 below.

8) IS2: Acute Infections – Initial: =((RAND<0.50)OR(IS1=0))
Birth: =(RAND<PS1) Transition: =IS2+((IS2<1)*(RAND<PS1))

At the beginning of each run of the simulation, 5% of the population is given to be acutely infected (arbitrarily set as the PCR-detected level of infection found in Levin et al. (2009)). This, along with eqn. 7 above, gives 95% of the population as being exposed at the start of each run. Because 90% of the population has already been assigned to be chronically infected, half of the remaining 10% of the population is set as chronically infected. In every year following the first, PS1% (eqn. 5) of the population becomes infected. The name of this variable is a misnomer, though, because it stays non-zero after the acute period is over. The actual effect of infection on the model is handled by the Mortality functions.

An individual's infection status is determined by their values for IS1 and IS2 together (eqns. 7 and 8). If (IS1=0) and (IS2=0), then they are uninfected. If (IS1=0) and (IS2=1), they are acutely infected. If (IS1=1) and (IS2=1), then they are chronically infected.

Mortality Rates

Base mortality for each age class:

0–1 years old: 67%;
1–2 years old: 25%;
2–3 years old: 5%;
3+ years old: 5%

Mortality functions for the no-relapse model:

9) =67+((MIN(GS2:32))*((IS1=0)*(IS2=1)))

=25+((MIN(GS2:74))*((IS1=0)*(IS2=1)))

=5+((MIN(GS2:94))*((IS1=0)*(IS2=1)))

When an individual is uninfected, they experience an X% chance of mortality each year, according to their age class (with X = 67, 25, or 5). For an individual to experience increased mortality (GS2, capped at 99%), they must be infected (IS2=1, eqn. 8). Additionally, the infection must not yet have become chronic (IS1=0, eqn. 7). This means that, under this model, individuals only experience increased mortality from disease the year that they become infected, not anytime afterward.

Mortality functions for the all El Niño relapse model:

10) =67+((MIN(GS2:32))*(IS2=1)*((IS1=0)OR((CAT(1)=0)OR(CAT(2)=0)))

=25+((MIN(GS2:74))*(IS2=1)*((IS1=0)OR((CAT(1)=0)OR(CAT(2)=0)))

=5+((MIN(GS2:94))*(IS2=1)*((IS1=0)OR((CAT(1)=0)OR(CAT(2)=0)))

As in eqn. 9, individuals will experience heightened mortality when (IS1=0, eqn. 7) and (IS2=1, eqn. 8); that is, they are acutely infected. However, in this model only (IS2=1) is strictly necessary; mortality for these individuals will also be increased during a strong El Niño year (CAT(1)=0) or a weak El Niño year (CAT(2)=0). This translates into chronically infected individuals experiencing increased mortality during all El Niño events.

Mortality functions for the strong El Niño relapse model:

11)

=67+((MIN(GS2:32))*(IS2=1)*((IS1=0)OR(((PS2=1)*(GS6<0.5))OR((CAT(1)=0)*(PS2=0)*(1-(GS7<0.5))))))

=25+((MIN(GS2:74))*(IS2=1)*((IS1=0)OR(((PS2=1)*(GS6<0.5))OR((CAT(1)=0)*(PS2=0)*(1-(GS7<0.5))))))

=5+((MIN(GS2:94))*(IS2=1)*((IS1=0)OR(((PS2=1)*(GS6<0.5))OR((CAT(1)=0)*(PS2=0)*(1-(GS7<0.5))))))

This model includes relapses, but only during the stronger year of a strong El Niño event. Which of the two years is stronger is randomly chosen, determined in part by the variables GS6 (eqn. 3) and GS7 (eqn. 4). If (GS6<0.5), then the first

year of the Strong El Niño (as given by PS2=1, eqn. 6) will be the stronger year. If (GS6>0.5) [or equivalently, (1-(GS7<0.5))], then the second year of the Strong El Niño [given by (CAT(1)=0)*(PS2=0)] will be the stronger year. Again, this is used to determine when a relapse will occur for chronically infected individuals.

Catastrophe Functions

12) Strong El Niño Frequency: =5+(100*(PS2!=0))

Strong El Niño events begin with a probability of 5% each year. At the beginning of the second year, PS2 (eqn. 6) equals 1, causing the strong El Niño event to continue for that second year, at which point PS2 returns to 0.

13) Strong El Niño Reproduction Severity: =((PS2=1)*(((GS6<0.5)*0.01)+((1-(GS6<0.5))*0.9)))+((PS2=0)*(((1-(GS7<0.5))*0.01)+((GS7<0.5)*0.9)))

During the first year of a strong El Niño (PS2=1, eqn. 6), the El Niño severity will be strong (if GS6<0.5, eqn. 3) or weak (if GS6>0.5). In the second year (PS2=0), the opposite effect will occur (because GS7, eqn. 4, returns the same number as last year's GS6). The value of 0.9, from the terms ((1-GS6<0.5)*0.9) and ((GS7<0.5)*0.9), represents the effect of the weaker year during a strong El Niño event (see Appendix A).

14) Strong El Niño Survival Severity: =((PS2=1)*(((GS6<0.5)*0.3)+((1-(GS6<0.5))*1.0)))+((PS2=0)*(((1-(GS7<0.5))*0.3)+((GS7<0.5)*1.0)))

The effects of this function are similar to the above, but affecting survival instead of reproduction.

15) Weak El Niño Frequency: =20-(100*(CAT(1)=0))

Weak El Niños occur with 20% probability each year, except in years when a strong El Niño is already occurring.