Supplementary Data

1 Implementation of the Crofton Method

The algorithm for fitting the Crofton Method (Crofton 1971) proceeds as follows. First, obtain a dataset with n hosts where each host has some parasite intensity 0 to p_{max}. Starting with the full dataset, guess a vector of pre-mortality parameters $(N_{p1}, \mu_{p1}, k_{p1})$ where N_{p1} is the total number of hosts before mortality, μ_{p1} is the parasite intensity before mortality, and k_{p1} is the parasite aggregation before mortality. Given these parameters, use a negative binomial distribution to calculate the predicted number of hosts with $0, 1, 2, \ldots p_{max}$ parasites. Compare the expected number of hosts with $0, 1, 2, \ldots p_{max}$ parasites to the observed number hosts with $0, 1, 2, \ldots p_{max}$ parasites and calculate the χ^2-squared statistic associated with the observed and predicted vectors. In reality, one often has to bin the parasite intensity data because all parasite intensities are not represented in the dataset. Continue to guess $(N_{p1}, \mu_{p1}, k_{p1})$ vectors until a set of parameters is found that minimizes the χ^2- squared statistic.

Second, choose a truncation value (t_2) such that $t_2 < p_{max}$. Truncate the data such that $\text{data}_{\text{truncated}} <= t_2$ and repeat the above iterative procedure to calculate another set of parameters $(N_{p2}, \mu_{p2}, k_{p2})$ that minimizes the χ^2-squared statistic on the truncated data. Choose a new truncated value $t_3 < t_2$ and repeat the first two steps. Continue to truncate the dataset until it only contains hosts with 0, 1, and 2 parasites (or 3 bins). Because the method attempts to estimate three parameters, at least 3 classes are needed for all 3 parameters to be identifiable (Royce and Rossignol 1990).

Once the iterative procedure has been completed, parasite-induced host mortality is traditionally identified by plotting the different truncation values t_i against the different values of N_{pi} and looking for a distinct "kink" in the resulting

plot. Once the "kink" as occurred, the values of N_{pi} will typically remain close to constant as t_i is decreased further. The "true" pre-mortality parameters N_{pt}, μ_{pt}, and k_{pt} are taken to be at the point where the "kink" occurs.

We provide an implementation and unit tests of the Crofton Method in Supplementary Data S3. Figure S10 visually shows that our implementation of the Crofton Method agrees with results previously published by Crofton (1971).

2 Implementation of the Adjei Method

The Adjei Method for estimating PIHM has two steps (Adjei et al. 1986). The first step is to estimate the parameters of the pre-mortality host-parasite distribution using the Crofton Method (see Supplementary Data S1). The three parameters estimated are the total number of hosts before mortality N_p, the mean number of parasites per host before mortality μ_p, and the aggregation of parasites before mortality given by the parameter k_p from a negative binomial distribution. When k_p is small, parasites are highly aggregated among hosts and when k_p is large parasites are more evenly distributed across hosts (Wilson et al. 2002). The implementation of the Crofton Method has been discussed at length elsewhere (e.g. Royce and Rossignol 1990; Lester 1984, and in Supplementary Data S1) and we provide a tested implementation of the method in Supplementary Data S3.

The second step of the Adjei Method is to make the assumption that infection, host mortality, and sampling occur in that order and are temporally separate (Adjei et al. 1986). Next, Adjei et al. assume that the host survival function follows the logistic form

$$h(\text{survival}; x, a, b) = \frac{\exp\left(a - b\log(x)\right)}{1 + \exp\left(a - b\log(x)\right)} \tag{1}$$

where x is the parasite intensity in a given host and a and b are the two parameters

of the logistic function. Generally, a larger a allows for hosts to tolerate larger parasite intensities before experiencing parasite-induced mortality and a larger b leads to a more rapid decline in the probability of host survival as parasite intensity increases. The value $\exp(a/b)$ is referred to as the LD_{50}. Individuals with loads higher than this will have a greater than 50% chance of death.

By taking the first and second derivatives of equation 1, one can easily find that the maximum rate of decline in host survival probability with increasing parasite intensity occurs at the LD_{50} and has a value of $b/4$. This is in many ways analogous to the parasite pathogenicity parameter α given in classic macroparasite models, which specifies the slope of the linear relationship between between host death rate and parasite intensity (Anderson and May 1978; Isham 1995). The parameter a is easily interpreted by holding b constant and looking at how a one unit change in a affects the log parasite intensity at which some percentage p of hosts experience mortality. Letting a_1 and a_2 be two different values of a and x_1 and x_2 be two different parasite intensities, a bit of rearranging of equation 1 gives.

$$\begin{aligned}
\log \frac{p}{1-p} &= a_1 - b \log x_1 \\
-\log \frac{p}{1-p} &= a_2 - b \log x_2 \\
0 &= a_1 - a_2 - b \log x_1 + b \log x_2 \\
a_2 - a_1 &= b(\log x_2 - \log x_1)
\end{aligned}$$

If $a_2 - a_1 = 1$, then the change in log parasite intensity at which p percentage of hosts survive is $1/b$.

To estimate the parameters in equation 1, the Adjei Method first calculates the expected number of hosts with a given parasite load x by using the equation $g(x; \mu_p, k_p) * N_p$, where $g(x; \mu_p, k_p)$ is the negative binomial pre-mortality

distribution. Second, the observed and predicted number of hosts with x parasites are paired as a single data point and the method then assumes that this data point follows a binomial distribution with the total number of "trials" equal to the predicted number of hosts and the total number of "successes" equal to the observed number of hosts. In some cases, the observed number of hosts is greater than the expected number of hosts and the Adjei Method alters the data so that the observed is equal to the predicted (Adjei et al. 1986). After this questionable manipulation, the (observed, predicted) pairs are fit to a standard Generalized Linear Model (McCullagh and Nelder 1989) with a binomial response variable and a logistic link function given by equation 1. This model provides estimates for parameters a, b and LD_{50}.

While not included in the original implementation of the Adjei Method, a χ^2 test with a degrees of freedom of 1 can be used to assess whether a GLM model that includes parasite intensity as a predictor of host survival probability is a "better" model than a GLM without this predictor. This allows the Adjei Method to determine whether PIHM is a significant factor in a host-parasite system.

The Adjei Method's most glaring deficiency is the need to alter the observed data in order to fit the model into the binomial GLM framework. A second more subtle problem with the Adjei Method is the potential need to bin data in order to predict greater than one host in a given parasite intensity class. For example, if the total number of hosts pre-mortality was 50, the mean number of parasites per host pre-mortality was 100 and the aggregation parameter was 1, applying the equation $g(x; \mu_p = 100, k_p = 1) * 50$ would result in less than 1 individual in all parasite intensities x. In other words, the Adjei Method cannot be applied to samples with either very high mean parasite loads, small sample sizes, or both without some sort of binning of the data. While this is not a flaw *per se*, it does add a certain level of subjectivity (i.e. which bins should you use?) to a method that already has serious potential issues. In this analysis, we always assume the

Adjei Method is not binning the data, though we provide code for applying the binning method in Supplementary Data S3.

3 Code and unit tests for estimating parasite-induced host mortality

Python code, unit tests, and a help file for the Crofton Method, the Adjei Method and the Likelihood Method can be found at `https://github.com/mqwilber/parasite_mortality`

References

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