Efficient Sample Size Calculator

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OVERVIEW OF THE SOFTWARE

This software application ('Efficient Sample Size Calculator') is used to pinpoint the number of hosts that need to be tested without finding a positive disease case to have 95% probability that the true prevalence in the population is at or below 1% or 2%. Unlike traditional sample size calculators, this calculator considers the natural biological groupings of hosts and the correlation in disease status among them. Three sampling methods (i.e., the determination of which hosts are selected for testing) are available in the calculator: simple random sampling, high-harvest sampling, and two-stage cluster sampling.

We provide dozens of real-world examples that illustrate how the Efficient Sample Size Calculator can be used to plan health investigations when the goal is to rule out disease at the population scale. Examples use wild hosts, such as mammals, herpetofauna, avians, marine species and insects, each afflicted by various diseases, representing scores of combinations of population sizes, group sizes, within-group correlations (contact structures and transmission propensities), and diagnostic test sensitivities.

The Efficient Sample Size Calculator is useful in determining statistical significance while planning health investigations. The calculator can be used to explore desirable sampling schemes, compute a budget for a project, and suggest the scope of field coordination needed to meet target sampling goals. The calculator is also helpful in identifying whether a desired investigation is financially or logistically feasible. The statistical theory can be found in Booth et al. (2024) and Booth et al. (2025). The majority of the descriptions in this readme and code were adapted with permission from Hanley et al. (2024).

> *Note: Reproduction of this app requires working knowledge of R and RStudio, including the ability to reproduce an environment on a local machine from a lock file, if necessary.*

REQUIREMENTS TO USE THE CALCULATOR
Population
To engage with the Efficient Sample Size Calculator, you will need a total population size of 100 (or more) hosts and an estimate of the cluster size. The definition of a cluster is the natural clustering of individuals on the landscape, where a population can consist of more than one cluster (as in Booth et al. 2024) or a single cluster (as in Booth et al. 2025).

Correlation
At the time of this writing, there is no established mathematical way to compute correlation in disease status among members sharing a cluster (i.e., social group), but this is an active area of research by the authors. For now, use your best judgement to assign values for correlation ranging from zero (no transmission of disease among members sharing a cluster), to low (unlikely transmission among members of a cluster; correlation = 0.1) to high (highly likely transmission among members of a cluster; correlation = 0.9) or somewhere in between. When correlation is equal to 0, the structure of the clusters is irrelevant, so the Efficient Sample Size Calculator cannot be run. When correlation is equal to 1, all the hosts sharing a cluster will either be disease positive or disease negative and sampling a single animal from a cluster will provide all the disease information for that cluster. The Efficient Sample Size Calculator cannot be run under these conditions.

Diagnostic Sensitivity
Use of Live or Dead Hosts
Sometimes, two or more types of diagnostic tests can be used by investigators to confirm the presence of a pathogen or diagnosis of a disease. The Efficient Sample Size Calculator is invariant to the diagnostic test used as long as the sensitivity parameter is accurate for the pathogen and host species of interest. It makes no difference in the calculator if hosts are alive or dead when tested, which provides maximum flexibility to execute science despite real-world obstacles in sample procurement or testing. If more than one choice of tissues and diagnostic tests are available, you should use the tissue and test that has the highest sensitivity because that will result in fewer hosts needing to be tested (i.e., sample savings) over a test with lower sensitivity. Low sensitivity can negate savings arising from clustering, or worse, require more hosts to be tested than the number required when correlation is not considered. In all cases, users should enter the sensitivity of the test that they intend to use in the future investigation, even if it has low sensitivity.

Prevalence of Interest
The goal of the Efficient Sample Size Calculator is to determine sample size needed to declare a free-ranging population free from disease. Freedom from disease is not as well defined in applied wildlife health as it is in agricultural animal health. Sometimes, freedom from disease is defined in wildlife as a prevalence at or below the point of feasible detectability (with 1% prevalence a common usage). Other times in applied settings, wildlife managers may prefer to use fewer sampling resources to detect a prevalence at or below 2%. For populations affected with disease at greater levels than 1-2%, we recommend using Epitools (Sergeant 2018). In the future, we may add additional features to this calculator to ensure that correlation is considered for disease more common than 1% or 2% prevalence, or when the prevalence of interest needs to be more flexible for practitioners.

PREPARING THE CALCULATOR FOR USE ON YOUR MACHINE
Step 1. Create a working directory on your local machine.

> **1a.** Make a new folder on your computer and name it. This will be your working directory where associated files will be saved. This working directory is important because it will contain the environment necessary to run the application.

Step 2. Prepare the software application for use on your machine.

> **2a.** Download and save the contents into your working directory. The directory should contain:

- www (must unzip then remove the redundant www sub-folder, so the pictures exist within a single www folder)
- .Rprofile (RPROFILE File)
- renv.lock (LOCK File)
- app (.R File)
- App_Functions (.R File)

Step 3. If you do not have R Studio on your computer, you will need to download it now.

3a. Download R at https://www.r-project.org/. Follow the prompts and accept the defaults. R is free and open source.
- The specific R version used is 4.4.1.

3b. Download R Studio at https://posit.co/downloads/. Follow the prompts and accept the defaults.
- The specific R Studio version used is 2023.09.0 Build 375 'Cherry Hibiscus'.

Step 4. Open app.R in R Studio.

4a. Right click on app.R and select 'Open with,' then select R Studio.

Step 5. Ensure R Studio has the path to the materials in the working directory.

5a. In the top ribbon of R Studio, click 'Session,' 'Set Working Directory,' then 'Choose Directory.'

5b. Navigate to the folder that you initialized in **Step 1a** and click 'Open'.

Step 6. In the upper right-hand corner of R Studio, click 'Run App.'

Note: Depending on the configuration of existing packages on your local machine, you may be prompted to install one (or more) packages before the app can run. Follow the prompts to do so, then click 'Run App.'

If after installing the packages, the app still does not open, remove the # from the library(renv), renv::restore(), and renv::status commands (lines 38-40) and then run the code to troubleshoot which package versions on your machine might be out-of-sync with the lock file. The renv.LOCK file shows the exact environment used to create this code, and you will need to reproduce a compatible environment on your local machine before you can run this app. Refer to Ushey & Wickham (2024).

Step 7. Once the app opens, interact with the Efficient Sample Size Calculator.

Step 8. Close the Efficient Sample Size Calculator.

NAVIGATION THROUGH THE USER INTERFACE

The Efficient Sample Size Calculator opens to the Home tab, which contains buttons to begin the computation. The interface of the Efficient Sample Size Calculator is designed so the user progresses through the steps listed in the side menu using the navigational buttons located on each page. Navigation buttons to advance pages only appear when the user input entries are valid. Users can return to a previous step at any time using the '< Return' button. Once the user reaches Step 6, they can review their entries before clicking 'Launch Calculation.' A progress bar appears while the simulation is active and automatically vanishes once the simulation is complete. Simulation outputs will appear that reveal the number of hosts needed for testing without a positive case to have a 95% probability that the underlying prevalence of the disease in the population is at or below 1% or 2%.

Home Tab

The Home tab introduces the Efficient Sample Size Calculator and its funders. It also allows access to Step 1 of the calculator ("Begin Sample Size Calculation"). This tab is accessible at any time by clicking Home on the sidebar menu, or by navigating through each page using the back button.

Examples Tab

This tab contains sample size computations for over two dozen disease-host systems. Tutorials are for illustration and should not be used at face value without intense scrutiny from wildlife biologists (for the population parameters), epidemiologists (for the correlation parameter), and pathologists and/or diagnosticians (for the sensitivity parameter). Tutorials include:

Simple Random Sampling
- Aspergillosis in green heron (*Butorides virescens*)
- Aspergillosis in seagulls (*Larinae* spp.)
- Avian reovirus in American crow (*Corvus brachyrhynchos*)
- Canine distemper virus in skunks (*Mephitis mephitis*)
- Chytrid fungus in hellbenders (*Cryptobranchus alleganiensis*)
- Highly pathogenic avian influenza in penguins (*Spheniscidae* spp.)
- Mycoplasmal conjunctivitis in Eastern bluebirds (*Sialia sialis*)

- Otarine herpesvirus-1 in California sea lions (*Zalophus californianus*)
- Glyptemys herpesvirus 2 in wood turtles (*Glyptemys insculpta*)
- Salamander chytridiomycosis in red spotted newt (*Notophthalmus viridescens*)
- *Trichinella spiralis* in wolverines (*Gulo gulo*)
- Tularemia in beavers (*Castor canadensis*)
- West Nile virus in bald eagles (*Haliaeetus leucocephalus*)

High-Harvest Sampling
- Adenovirus hemorrhagic disease in black-tailed deer (*Odocoileus hemionus columbianus*)
- Anthrax in pronghorn antelope (*Antilocapra americana*)
- Brucellosis in caribou (*Rangifer tarandus*)
- Campylobacteriosis in moose (*Alces alces*)
- Chronic wasting disease in white-tailed deer (*Odocoileus virginianus*)
- Oral necrobacillosis in reindeer (*Rangifer tarandus*)
- Pneumonia in big horn sheep (*Ovis canadensis*)
- Pneumonia in mule deer (*Odocoileus hemionus*)
- Treponeme-associated hoof disease in elk (*Cervus canadensis*)

Two-Stage Cluster Sampling
- Avian reovirus in American woodcock (*Scolopax minor*)
- Duck Viral Enteritis in mallards (*Anas platyrhynchos*)
- Erysipelothrix infection in bottlenose dolphins (*Tursiops truncatus*)
- Heartworm in wolves (*Canis lupus*)
- Highly pathogenic avian influenza in snow geese (*Anser caerulescens*)
- Lymphoproliferative disease virus in turkeys (*Meleagris gallopavo*)
- Mycoplasma in bison (*Bison bison*)
- Parvovirus in raccoons (*Procyon lotor*)
- Sarcoptic mange in foxes (*Vulpes vulpes*)
- Snake fungal disease in timber rattlesnakes (*Crotalus horridus*)
- *Staphylococcus aureus* infection in ants (*Lasius emarginatus*)
- White-nose syndrome in little brown bats (*Myotis lucifugus*)

This tab is accessible at any time by clicking Examples on the side bar menu.

Citations Tab
This tab contains the references underlying the theory, R programming, and examples.
This tab is accessible at any time by clicking Citations on the sidebar menu.

Questions Tab
This tab contains the contact details of the authors. This tab is accessible at any time by clicking Questions on the sidebar menu.

Step 1. Host Population

The Host Population tab is used to enter the population information (i.e., parameters) of the population of interest. This tab is only accessible using the navigation buttons on the main pages after the user begins the calculator. Once at Step 1, the user is not presented with a button to advance to Step 2 unless user-inputs population parameters are valid. Invalid population parameters include non-numeric inputs, or population or cluster sizes less than or equal to 0.

Total Population Size (user input required)
The user is asked to estimate the Total Population Size in the text box. The Total Population Size is defined as the total number of hosts in the entire population of interest. Appropriate entries for the Total Population Size include any whole positive number greater than 100 hosts.

> *Note: The minimum requirement of 100 hosts is related to the goal of investigating prevalence at or below 1%.*

Cluster Size (user input required)
The user is asked to enter the estimated Cluster Size into the text box. The Cluster Size is defined as the exact or approximate number of hosts that constitute each cluster (i.e., social group, family group, herd, or countless other group types). Appropriate entries for the Cluster Size include any whole positive number ranging from 1 host (in which case groups consist of singletons) to the number in the total population (in which case there is a single cluster in the population).

> *Note: Sample size estimates when the total population size is equal to the cluster size hinge on the prior on prevalence being Beta(1,9), which is hardcoded.*

Fixed or Variable Cluster Sizes (user input required)
The user is asked to select fixed or variable cluster sizes.

- Fixed (default). Clusters sizes contain the exact number of hosts specified by the user and are identical in size across the population.
- Variable Cluster Sizes. Cluster sizes are drawn from a Poisson distribution with the average cluster size as the central parameter.

> *Note: After the user enters the population parameters, two adjustments are automatically made by the Efficient Sample Size Calculator, in sequence.*
> - *First automatic adjustment: The total number of clusters in the population is automatically adjusted to be equal to the largest number of complete clusters in the user-specified population.*

- o *Second automatic adjustment: The user-entered population size is automatically adjusted to the new total population size, which is the closest total population that constitutes complete clusters.*

Note: The Efficient Sample Size Calculator may automatically make two additional adjustments, if necessary.
  - o *The Calculator does not allow randomly generated cluster sizes of 0. Anytime the random number generator produces a cluster size of 0, the software automatically updates the offending cluster to size 1.*
  - o *The Calculator automatically scales the total populations size to the nearest value above 100 in which all clusters (fixed or variable) are complete.*

Note: There may be a delay in the reactivity of this tab if the user-entered population size is large and cluster size is small. Please be patient and wait until the graph loads to proceed.

Step 2. Sampling Method

The Sampling Method tab is used to enter the sampling method used to select hosts from a population for disease testing. This tab is only accessible using the navigation buttons on the main pages after the user begins the calculator. Once at Step 2, the user is not presented with a button to advance to Step 3 unless user-inputs regarding the sampling configuration are valid. Invalid configurations include entries where desired sample sizes exceed available hosts to sample.

Sampling Method (user input required)
The user is asked to Select a Sampling Method:

- Simple Random Sampling (default): Every host from the entire population has an equal chance of being sampled, regardless of the cluster to which it belongs.

- High-Harvest Sampling: Hosts occupying certain clusters ('high-harvest clusters') are more likely to be sampled than hosts occupying remaining clusters ('non-high harvest clusters'). For example, a subset of clusters of game species are more accessible to hunters than others. If wildlife agencies rely on hunter-harvested animals for opportunistic disease sampling, then that subset of clusters will be sampled more frequently than others.

- Two-Stage Cluster Sampling: Clusters themselves are chosen at random to be sampled, then hosts within the selected clusters are randomly sampled. This scheme means that unselected clusters (and the hosts within them) are not sampled at all.

  Conditional Input When High-Harvest Sampling is Selected
  Proportion of High-Harvest Clusters (conditionally required input)

The proportion of clusters in the population to be sampled with higher intensity.

Proportion of High-Harvest Samples (conditionally required input)
The proportion of total samples to be taken from high-harvest clusters.

Conditional Input When Two-Stage Cluster Sampling is Selected
Proportion of Clusters to be Sampled (conditionally required input)
The proportion of clusters in the population to be randomly selected. All hosts tested in the population are themselves randomly sampled from the selected clusters.

Note: There may be a delay in the reactivity of this tab if the user-entered population size is large, cluster size is small, or both. Please be patient and wait for the graphic to load.

Step 3. Correlation

The Correlation tab is used to enter prior knowledge regarding the contact structure within social groups (i.e., clusters) and the transmissibility of the disease of interest. This tab is only accessible using the navigation buttons on the main pages after the user begins the calculator. Once at Step 3, the user is not presented with a button to advance to Step 4 unless user-inputs are valid. Invalid entries include correlations equal to 0 or 1.

Average Correlation (user input required)
The user is asked to Set the Average within Cluster Correlation. Average within Cluster Correlation is defined as the correlation in disease status between hosts in a cluster, as inferred through contact rates and transmission probabilities. A correlation of 0 means that disease status is independent between hosts sharing a cluster. When correlation is 0, disease transmission among hosts is just as likely to occur between clusters as within clusters. When correlation is between 0 and 1, disease transmission is likely to occur between hosts sharing a cluster. A correlation of 1 indicates that all hosts in the same cluster have identical disease status (which can be disease-positive or disease-negative). In this calculator, a correlation of 0.01 is the lowest possible correlation which should be used whenever clusters consist of only one individual (i.e., no social grouping within the population), or when correlation in disease status is low.

Note: Priors in Booth et al. (2024) include correlation and prevalence. However, for simplicity in this app, we hard coded the prior on prevalence to be Beta(1,9).

Step 4. Diagnostic Details

The Diagnostic Details tab is used to enter the sensitivity of the method used to conduct disease testing. This tab is only accessible using the navigation buttons on the main pages after the user begins the calculator. Once at Step 4, the user is not presented with a button to advance to Step 5 unless user-inputs are valid. An invalid entry occurs when sensitivity is equal to 0.

Sensitivity of Diagnostic Test (user input required)
The user is asked to Set the Sensitivity of the Diagnostic Test. Sensitivity is defined as the ability for the test to detect a true positive. A sensitivity of 0.01 means that the test correctly identifies the disease 1% of the time, while a sensitivity of 1 means the test correctly identifies the disease 100% of the time.

Step 5. Simulation Details

The Simulation Details tab is used to enter the details of the simulation. This tab is only accessible using the navigation buttons on the main pages after the user begins the calculator.

Number of Simulations (user input required)
The user is asked to Enter the Number of Simulations. Greater values reduce errors in estimates but increase computation time.

> *Note: Most examples are initialized with 1,000 simulations to minimize runtime, but at least 10,000 simulations are recommended for real investigations. Runs with 1,000 simulations take about 5 minutes to complete while runs with 10,000 simulations can take an hour or more to complete.*

Step 6. Compute Sample Size

The Compute Sample Size tab contains a summary of parameters, buttons to return to prior pages to adjust inputs, the button used to launch the simulation, and (after the simulation completes its run) the results from the simulation. This tab is only accessible using the navigation buttons on the main pages after the user begins the calculator. The content on this tab will disappear once 'Launch Calculation' is clicked, and simulation outputs will appear in its place after the simulations are complete.

> *Note: Depending on the configuration of user inputs, computational runtime can be as fast as a minute or as long as one or more hours. Please be patient.*

Output Plot
A plot depicting an estimate of the sample size needed to have high probability that prevalence is at or below 1% (orange) or 2% (dark red).

> *Note: The estimate of sample size (i.e., point estimate) arises from the Monte Carlo simulation, which means estimates vary depending on the run and number of simulations used.*

The output plot further includes error bars depicting the approximate standard error of the estimated sample size ($se(\hat{n})$). Standard errors are approximated by the Calculator using the equation:

$$se(\hat{n}) \approx \frac{1}{\frac{(\hat{p}_+ - \hat{p}_-)}{(\hat{n}_+ - \hat{n}_-)}} \sqrt{\frac{\alpha*(1-\alpha*)}{N_{sims}}}, \tag{1}$$

where $\hat{p}_-$ and $\hat{p}_+$ represent the probabilities on either side of the simulated probability ($\alpha*$), $\hat{n}_-$ and $\hat{n}_+$ correspond to the estimated sample sizes at those locations (respectively), and N_{sims} represents the number of simulations used. The distance between $\hat{p}_-$ and $\alpha*$, and $\alpha*$ and $\hat{p}_+$ is assumed to be 0.01. Generally, the user can reduce the standard error by increasing the N_{sims} used in the simulation.

Download Button
A button to download the parameters and outputs from the calculator as a .csv file.

Reset Button
A button to restart the calculator at the Home page.

NOTES
Models described in Booth et al. (2024, 2025) and Smith et al. (in review) both leverage a beta binomial framework to infer sample sizes needed to declare with statistical assurances that a free-ranging wildlife population is disease free. However, Booth et al. (2024, 2025) assume that hosts naturally cluster into social groups that share disease more readily among group members, while Smith et al. (in review) assume that groupings represent the set of hosts that share a common likelihood of being selected for sampling. Accordingly, sample sizes for declaring freedom from disease among the two approaches may not agree. Appropriate planning hinges on the selection of the modeling approach that best aligns with the assumptions of the proposed investigation.

TECHNICAL DETAILS
The app.R script calls the environment from file and drives the user interface. The App_Functions.R script contains the functions underlying the simulations. Please see the renv LOCK file for the R version and list of package versions used in the environment.

FINANCIAL SUPPORT

LICENSE

This Software is shared under a MIT License.

SUGGESTED CITATION FOR RE-USE

Hanley, B.J., Booth, J.G., Hodel, F.H., Thomspon, N.E., Reeder, A.A., Bloodgood, J.C.G., Dion, J.P., Van de Berg, S., Gonzalez-Crespo, C., Huang, Y., Wang, J., Miller, L.A., Hollingshead, N.A., Peaslee, J.L., & Schuler, K.L. 2025. Efficient Sample Size Calculator [Software]. Cornell University Library eCommons. DOI: 10.7298/smzr-1a70.

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