Diagnostic guidelines to confirm Day 0

Modified from Sponheim A, Alvarez J, Fano E, Rovira A, McDowell E, Toohill E, Dalquist L, Pieters M. A diagnostic approach to confirm *Mycoplasma hyopneumoniae* "Day zero" for pathogen eradication. Prev Vet Med 2023;221:106057.
Compiled by Sponheim, Fano, and Pieters, 2023.

Number of individual pigs to sample and cut-point based on the **mean or 95% lower confidence limit (LCL) sensitivity** of experimentally infected pigs when using deep tracheal secretions early during *M. hyopneumoniae* infection. For a lowest risk option, choose the number of individual pigs to sample and cut-point based on **95% LCL diagnostic sensitivity.** Assumes 100% diagnostic specificity, 95% population sensitivity and specificity, and population size of 1,000. The cut-point is the number of PCR negative individual samples to define a herd as "not properly exposed": if negatives < c herd was successfully exposed, if negatives ≥ c exposure was not successful.

	Number of individuals to sample			
	Cut-point			
	Design prevalence (% negative)			
Diagnostic Sensitivity	**≤1**	**≤5**	**≤10**	**≤15**
Mean[1]	*	335	104	56
		28	11	7
95% LCL[2]	*	*	315	148
			79	40

[1] Ninety-four percent unit sensitivity at 28 dpi (Sponheim et al., 2020)
[2] Seventy-nine percent unit sensitivity at 28 dpi (Sponheim et al., 2020)
* Target herd sensitivity and specificity cannot be achieved for a given design prevalence and diagnostic sensitivity

Looking for a different scenario? Use the link to calculate number of individuals to sample and cut-point: Epitools - FreeCalc: Calculate sample size for freedom t ... (ausvet.com.au)

Inputs:

Population size = select population size of interest

Test sensitivity = diagnostic sensitivity, since we are interested in detecting negative gilts (vs. positive), enter test or diagnostic specificity here (ie: 1 = 100%).

Test specificity = diagnostic specificity, since we are interested in detecting negative gilts (vs. positive), enter test or diagnostic sensitivity here. For deep tracheal secretions use **94% for mean** or **79% for 95% LCL** (ie: for lowest risk use LCL of 0.79 for 79%).

Design prevalence = prevalence estimate (% **negative**, ie: 0.10 for 10%)

Desired type I error = 1 – minimum population/herd specificity (ie: 0.05 for 5%)

Desired type II error = 1 – minimum population/herd sensitivity (ie: 0.05 for 5%)

Select modified hypergeometric exact for small populations or simple binomial for large populations. Results are presented as the number of individuals to sample and corresponding cut-point number of **negatives** acceptable to achieve the specified type I and type II errors for the given population, design prevalence and test performance.

References:

Cameron, A.R., Baldock, F.C., 1998. A new probability formula for surveys to substantiate freedom from disease. Prev. Vet. Med. 34, 1-17.

Sergeant, ESG, 2018. Epitools epidemiological calculators. Ausvet Pty Ltd. Available at: http://epitools.ausvet.com.au.

Sponheim, A., Alvarez, J., Fano, E., Rovira, A., McDowell, E., Toohill, E., Dalquist, L., Pieters, M. 2023. A diagnostic approach to confirm *Mycoplasma hyopneumoniae* “Day zero” for pathogen eradication. Prev. Vet. Med. 221, 106057. https://doi.org/10.1016/j.prevetmed.2023.106057.

Sponheim A, Alvarez J, Fano E, Schmaling, E., Dee, S., Hanson, D., Wetzell, T., Pieters, M. Comparison of the sensitivity of laryngeal swabs and deep tracheal catheters for detection of *Mycoplasma hyopneumoniae* in experimentally and naturally infected pigs early and late after infection. Vet Microbiol 2020; 241:108500. https://doi.org/10.1016/j.vetmic.2019.108500.