### Preregistration

# The effects of wolves on moose presence and abundance in south-central Sweden.

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# **Study Information**

**Title** The effects of wolves on moose presence and abundance in south-central Sweden.

#### Description

Predator-prey interactions often have strong effects on lower trophic levels in a given environment. For example, upon the re-establishment of wolves in a national park in North America, aspen trees have seen markedly increased recruitment (Ripple et al., 2014). In general, apex predator reintroduction is often considered to benefit ecosystems by limiting grazer populations and their consumption of lower trophic levels, and by providing meals for scavengers (Ripple & Beschta, 2004).

To date, most research on the effects of predator reintroduction has been conducted in natural areas minimally affected by human activities (Kuijper et al., 2016). However,

due to climate-driven relocation and controlled reintroduction programs, modern and future predator populations may become established in areas of strong anthropogenic influence (Kuijper et al., 2016). Consequently, when hoping to predict the effects of future predator reintroduction events, it is important to study areas with nearby human activity.

Recent studies in Sweden have found that wolf reintroduction does not impart biodiversity benefits to lower trophic levels, contrasting observations made in American National Parks. For example, Gicquel et al. (2020) reported that wolf reintroduction slightly increased moose browsing damage to Scots Pine trees in Sweden, as did Ausilio et al. (2021). By surveying moose pellets, Ausilio et al. (2021) also found that wolf presence and wolf territory establishment had slightly positive effects on moose presence and abundance. Other factors, including distance from roads (an indirect measure of human activities), had much stronger effects on moose populations than did wolves (Ausilio et al., 2021).

Future populations of predators will continue to establish themselves in areas of anthropogenic influence. It is important to critically investigate the effects that they may, or may not, have on prey populations and lower trophic levels in these areas. To increase certainty in their paradigm-questioning results, we will replicate Ausilio et al. (2021)'s research, conducting parallel analyses on the original study area over subsequent years, and asking whether or not wolves affect moose presence and abundance.

#### **Hypotheses** We hypothesize that:

- Wolf presence will moderately increase moose abundance, and probability of moose presence.
- 2) Time since wolf territory establishment will positively effect moose abundance, and probability of moose presence.

# Design Plan

# Study type and design

This study is a replication of original research by Ausilio et al., described in *Ecological Effects of Wolves on Anthropogenic Landscapes: The Potential for Trophic Cascades is Context-Dependent* (Ausilio et al., 2021).

Our hypotheses will be tested using a form of regression analysis. Data on 7 predictor variables and one response variable will be collected. Through modelling, we will predict the influence that the predictor variables have on both a) moose abundance, and b) probability of moose presence. Both outcomes will be informed by the response variable: moose pellet counts.

#### Data collection

The study will be conducted using data retrieved from various government and private sector institutions beginning in 2021 and ending in 2034, to replicate the 13-year original study by Ausilio et al. (2021). The study area will be the approximately  $100,000 \text{ km}^2$  range of south-central Sweden's breeding range of wolves, replicating the region assessed in the original study. The predominant trees in this landscape are Scots pine, Norway spruce, and birch. The average moose winter density in the study range is approximately 1.3 per km² (Zimmermann et al., 2014). Variables to be collected include: moose pellet counts, tree cover, roads, and wolf presence and time since wolf territory establishment. To replicate the original study as closely as possible, approximately 10,350 circular sample plots (area =  $38 \text{ m}^2$ ) will be surveyed, with the number sampled per year dependent on data availability.

#### Variables

Variable data will be collected within the defined study area in south-central Sweden. Data sources will be consistent with those used in the original study in order to maintain as close a replication study as possible. Seven variables will be used in our statistical analysis: 7 predictor variables and 1 response variable. Determining the effects that variables 7) and 8) below have on variable 1) will reveal the effects of wolves on the presence and abundance of moose. Exploring the effects of other predictor variables on variable 1) will potentially help identify other modulators of moose abundance, and probability of moose presence.

1) Moose pellet counts: Moose pellet counts will be used to gather presence and absence data on moose. Yearly surveys are conducted between the months of May and September throughout south-central Sweden (Fridman et al., 2014).

- The data source for this variable is the Swedish National Forest Inventory.
- 2) Forest age stages: There are four forest age stages considered when moose pellet count surveys are conducted. These stages classified as clear-cut, young, thinned, and mature. The data source for this variable is the Swedish National Forest Inventory.
- 3) Pine proportion: Sample plots, the same as those used for moose pellet count surveys, will be surveyed to assess tree cover. The cover (m²) of lodgepole pine and Scots pine, as a proportion of all tree cover, will be generated as an index of food availability. The data source for this variable is the Swedish National Forest Inventory.
- 4) RAWO: rowan, aspen, willow, and oak: Tree cover consisting of tree species highly selected for by moose was used to generate an additional index of food availability. The sum of the cover (m²) of rowan, aspen, willow, and oak trees was converted to a categorical variable of presence or absence of RAWO species. This variable is denoted as 'RASE' in the analyses. The data source for this variable is the Swedish National Forest Inventory.
- 5) Distance to the nearest forest road: Distance (km) between each plot and the nearest forest road, as classified by the database, will be calculated as a proxy representing human influence on the landscape. The data source for this variable is the Swedish national road database from the Swedish Transport Administration.
- 6) Distance to the nearest main road: Distance (km) between each plot and the nearest main road, as classified by the database, will be calculated as a proxy representing human influence on the landscape. The data source for this variable is the Swedish national road database from the Swedish Transport Administration.
- 7) Wolf presence: Presence or absence of a wolf territory, as determined by monitoring obtained by snow tracking, DNA-samples, and GPS locations of collared wolves. A buffered radius will be used based on the average wolf territory size in Scandinavia (Liberg et al., 2011). The data source for this variable is the National wolf-monitoring system from County Administrative Boards.
- 8) Time since welf territory establishment: A continuous range from 1 year to 27 years since establishment, as noted by annually conducted surveys. The data

source for this variable is the National wolf-monitoring system from County Administrative Boards.

## Analysis Plan

#### Statistical models

To prepare our data analysis procedures, we simulated data based on the original dataset collected and published by Ausilio et al. (2021). In this simulated data, the response variable was over-dispersed and had a large number of zeros (90.68%). We anticipate that we will collect similar data, and thus we plan to use a zero inflated negative binomial and hurdle model to investigate the effect of wolves and roads on moose. Hurdle models are composed of two components - zero inflated and conditional. The zero-inflated component runs a generalized linear mixed model (GLMM) with a binomial distribution and logit link using moose pellet presence/absence as the response variable. The conditional component of hurdle modelling runs a GLMM with truncated negative binomial distribution and log link using moose pellets (n>=1) as the response variable. For both the zero-inflated and conditional components of the hurdle model, TAXAR (year) is set as a random effect. The zero-inflated component of the model included moose presence (pellet count - binary variable) as a function of three predictor variables: distance to main roads, distance to forest roads, and pine proportion. The conditional component of the model included moose abundance (pellet counts - continuous variable) as a function of eight predictor variables: distance to main roads, distance to forest roads, pine proportion, wolf presence, time since wolf establishment, RASE presence, forest stage, and an interaction term between forest stage and wolf presence.

To address the issue of colinearity between continuous predictor variables, a correlation matrix was calculated using Pearson's correlation coefficient 'r' >= 0.6 (Zuur et al., 2010). The correlation matrix showed there was very low to no correlation between predictor variables. To find the most parsimonious model, models were compared using small-sample-size corrected Akaike Information Criterion (AIC), deltaAIC, and AIC weights from the dredge function in the "MuMin" package (Barton, 2020). Model averaging was performed using all models with a deltaAIC < 2 to generate model averaged parameter estimates.

We validated these data analysis procedures using our simulated data (Figure 1).

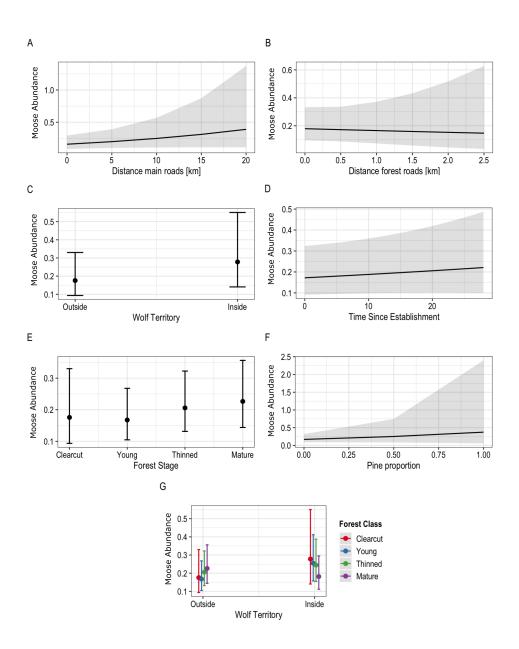


Figure 1: Marginal effects plot for predicted moose abundance in relation to (A) distance to main roads (km); (B) distance to forest roads (km); (C) wolf presence; (D) years since wolf territory establishment; (E) forest stage; F) pine proportion; and (G) the interaction between wolf presence and forest stage. Shaded regions indicate 95% confidence intervals.

# Exploratory analyses

We first investigated our simulated data by plotting the predictor variables of interest as a function of moose pellets to determine predictor variability and general trends within the data. We intend to conduct similar investigative, cursory analyses with our real data, once we collect it.

In addition to assessing the impacts of wolf presence and time since wolf territory establishment, we will determine if other predictor variables related to the forest environment and the level of human activity impact moose presence and abundance. Data for these predictor variables (variables 2-6 in the 'Variables' section above) are included in our models and their effects are included in our output (Figure 1).

We also included the interaction term between forest stage and wolf presence in the conditional component of our model to investigate the theory of habitat switching by moose in the presence of wolves.

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