Rodent trapping to investigate rodent assemblage structure in Eastern Sierra Leone to understand the spatio-temporal hazard of Lassa fever spillover.

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In this chapter I report a longitudinal rodent trapping study at four study sites in a Lassa fever endemic region of Eastern Sierra Leone. I perform rodent trapping along socio-ecological gradients to understand the association of land use on rodent species assemblages and population structures. I will perform morphological and molecular speciation on sampled rodents to understand the spatial and temporal dynamics of rodent species at the selected study sites. Data obtained from this study will be used to inform species occupancy and distribution models in the Eastern province of Sierra Leone. These models can be used to identify regions of interest for further investigation of Lassa fever epidemiology.

# Abstract

*Lassa mammarenavirus*, the causative agent of Lassa fever is endemic to Eastern Sierra Leone. The principal reservoir species (*Mastomys natalensis*), is considered abundant in human dominated habitats, however, rodent species’ assemblages are not well described. To investigate the structure of these assemblages in this heterogeneous landscape we conducted three monthly rodent trapping surveys for two years. We describe these rodent assemblages and their structures through morphological and molecular speciation, age classification, reproductive status and habitat preference. We produce species distribution maps across the study region from presence and absence data.

We found that *Mastomys natalensis* displayed seasonal variation in abundance and habitat choice, with populations concentrated in areas of human habitation during the dry season. We found that other rodent species’ within these communities diversify into distinct habitat niches, and that during periods of increased competition for resources, generalist species were more abundant than specialist species. There was good concordance between morphological field based identification and subsequent molecular speciation. Species distribution maps identified areas of expected absence of our species of interest *Matsomys natalensis* and potential discontinuity between populations.

We identify a complex dynamic system within rodent species assemblages co-located with human communities in Eastern Sierra Leone. The hazard of *Lassa mammarenavirus* spillover will be driven by fluctuating populations of host species due to either rodent population numbers or increased viral transmission within these rodent population due. The produced species distribution maps go some way towards explaining the observed limited geographic radiation of outbreaks of Lassa fever in this region and the slow spread of Lassa fever. We anticipate that this data will help inform higher resolution models of rodent distributions across West Africa. This may be of interest when investigating the effect of increasing development and land use change on rodent assemblages and in particular the hazard of Lassa fever outbreaks. Finally, these data highlight the need for comprehensive studies of rodent species assemblages elsewhere in the Lassa fever endemic zone to understand the generalisability of these findings and for future production of hazard hotspot maps for zoonotic pathogen spillover risk.

# Introduction

* Description of Lassa fever geography and epidemiology
* Description of *Mastomys natalensis* distribution and dynamics associated with Lassa fever outbreaks
* Introduce complex species networks with competition and dynamic population numbers resulting in the ongoing establishment of generalist species
* Highlight that few studies have investigated the wider rodent/small mammal assemblages in this region
* Introduce longitudinal, high intensity trapping as a useful methodology to describe population abundance and diversity in the absence of a feasible mark-recapture-release approach
* Introduce the potential impact of land use and climate change on the increase of the Lassa fever endemic region based on changes in rodent populations and environmental suitability
* Highlight that the production of species distribution maps which incorporate interactions between different species may produce better estimates than those based on single species studies, particularly in these highly competitive situations

First, we aimed to identify which rodent species were active at our study sites, to measure their prevalence and to described the structure of these species assemblages. Second, we wanted to investigate the association between these species assemblages, their population structures and land use. Third, we aimed to investigate the potential alterations to these species assemblages due to projected climate and land use change, in particular how changes to *Mastomys natalensis* populations may modify the hazard of *Lassa mammarenavirus* spillover.

# Methods

Rodent trapping occurred at up to 7 sites in 4 villages in the Lassa fever endemic zone of the Eastern Province of Sierra Leone. The villages were enrolled based on accessibility to the sites during all seasons, discussions with the Lassa fever outreach team at Kenema Government Hospital and acceptability of the protocol to the village community. Lalehun (coordinates 8.1975 N, -11.0803 E), is a relatively large village (~ 1,000 inhabitants) located on the route between Panguma and Tonga, Lambayama (coordinates 7.8544 N, -11.1897 E) is a peri-urban community that is becoming enveloped within Kenema, Seilama (coordinates 8.1222 N, -11.1936 E) is a small village (~ 300 inhabitants) located to the South-West of Panguma and Baiama (coordinates 7.83708 N, -11.26845 E) is a small village (~300 inhabitants) to the West of Kenema located off the main road between Kenema and Bo.

Within each village up to 7 study sites were chosen to provide coverage of the different observed land classifications of Eastern Sierra Leone. Study sites were geolocated for repeated trapping and alterations to land use at the study site were recorded at each visit. Within each study site up to 49 individual Sherman traps (**size and reference**) were baited with a locally produced mixture of oats, palm oil and dried fish for 4 consecutive nights. Each morning the traps were checked and closed for the day prior to rebaiting during the evening. Traps identified to contain rodents were brought to the biopsy site. Rodents were sedated and euthanised using cervical dislocation prior to obtaining morphological measurements and samples of blood and tissue.

Morphological speciation in the field was performed using a dichotomous key to identify rodents to species or genus (**will place in supplementary**). Molecular speciation was performed on formalin fixed tissue samples (**insert further details**). Rodents were sexed based on external and internal genitalia. Age estimation was performed through both, description of the rodents reproductive status and weighing of dried eye lenses.

## Rodent prevalence and species assemblage structure

* Adequacy of trapping effort at each village site was assessed using species accumulation curves
* Presence of an individual rodent species was detected through a successful trapping event within a study site
* Pseudo-absence of an individual was defined as no detection after 3,000 (**or some other value**) consecutive trap-nights at a study site within a study village
* We describe species assemblages at multiple geographic scales. First, all species identified within a single study-site. Second, all species identified within a village (i.e. an area in when rodents would be expected *a priori* to be able to diffuse across). Thirdy, all species identified within a single habitat type across multiple study sites and villages.
* The structure of a species assemblage includes description of the proportion of male and female individuals and their age classification (e.g. juvenile, adult).

## Rodent species assemblages and land use

* Relative abundance of a species in different land use types will be reported - consistent trapping effort will support this estimate in the absence of mark-recapture-release studies which are not suitable due to concerns about Lassa infection of rodents at the study sites.
* The change in these assemblage by season will be explored and associated with remote sensed environmental data such as NDVI, ground temperature, isothermality and precipitation
* Species distribution maps will be produced for each identified species with sufficient data using Bayesian additive regression trees
* These distributions will be examined to investigate the potential role of competition between species at study sites where absence or abundance is less than would be expected based on these distribution maps (i.e. mus or rattus displacing mastomys from suitable habitats)

## Impact of climate or land use change on rodent distributions

* Using projected land use change/climate change what impact will this have on selected rodent species(i.e. rattus, mus and mastomys)

# Results

## Summary of trapping effort and rodent descriptives (**potentially methods**)

Trapping visits to villages occurred at three monthly intervals for at least two years. One village (Bambawo) was removed from the study due to concerns raised by the study team over equipment security.

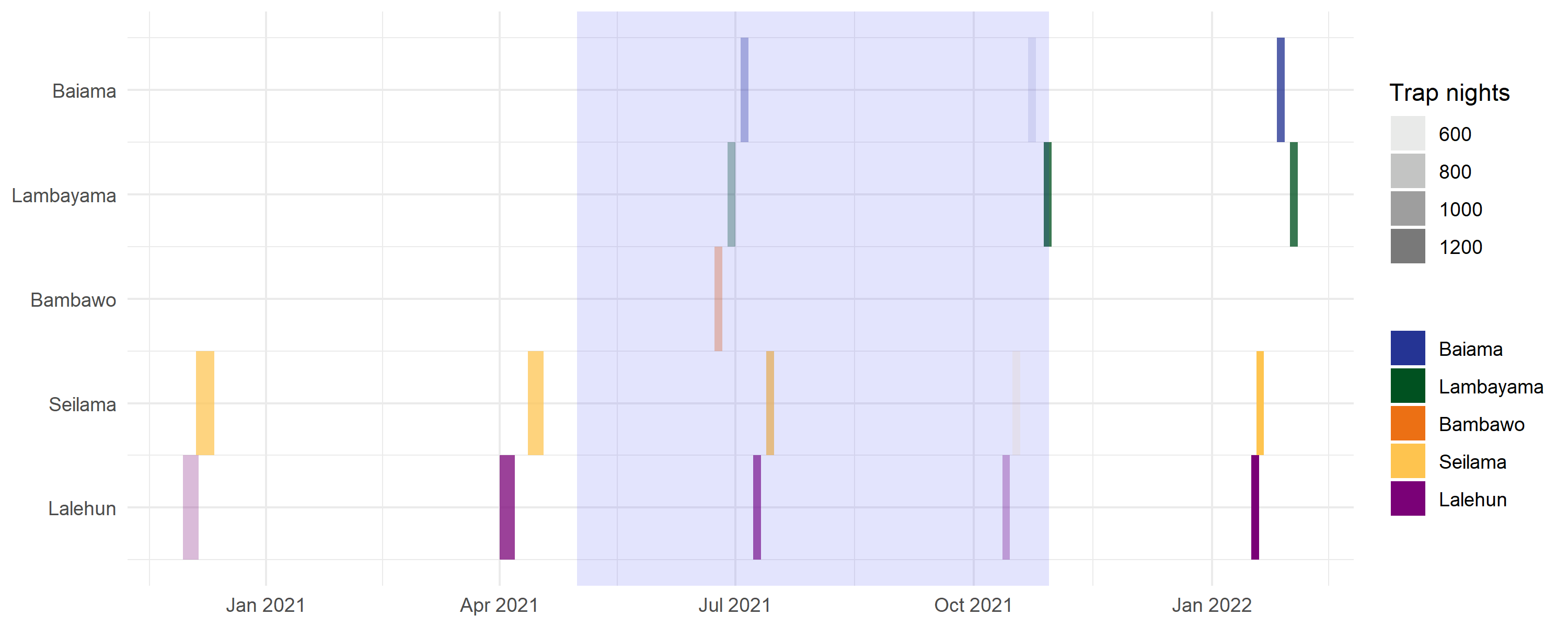


Figure 1. Timeline of trapping activity at village sites. Intensity of the colour relates to the number of trap nights obtained during the trapping visit. The blue shaded area represents the rainy season in Eastern Sierra Leone (monthly precipitation >300mm)

## Rodent presence and species assemblage structure

* Description of presence/pseudo-absence of species across study site and habitat type by season
* Describe the assemblages of different species across study site and habitat type by season

## Rodent species assemblages and land use

* Model seasonal change in a) relative abundance of all rodents and b) number of species by seasonal indicators and proportions of different land use types as covariates
* Species distribution maps will be produced
* Difference between observed and expected will be investigated by comparing to observations of other species within these distributions

## Impact of climate or land use change on rodent distributions

* Apply these species distribution models to expected future scenarios

# Discussion

# Conclusion

Five villages have been enrolled, one will not be carried forward due to concerns from the local team. The village are:

* Bambawo (coordinates 8.009 N, -11.1303 E)
  + Visit 1 - 2021-06-23 to 2021-06-27
  + Stopped after a single visit
* Lalehun (coordinates 8.1975 N, -11.0803 E)
  + Pilot/Visit 1 - 2020-11-30 to 2020-12-03
  + Visit 2 - 2021-04-01 to 2021-04-04
  + Visit 3 - 2021-07-08 to 2021-07-12
  + Visit 4 - 2021-10-12 to 2021-10-15
  + Visit 5 - 2022-01-16 to 2022-01-19
* Lambayama (coordinates 7.8544 N, -11.1897 E)
  + Visit 1 - 2021-06-28 to 2021-07-02
  + Visit 2 - 2021-10-28 to 2021-10-31
  + Visit 3 - 2022
* Seilama (coordinates 8.1222 N, -11.1936 E)
  + Pilot/Visit 1 - 2020-12-05 to 2020-12-09
  + Visit 2 - 2021-04-12 to 2021-04-15
  + Visit 3 - 2021-07-13 to 2021-07-17
  + Visit 4 - 2021-10-16 to 2021-10-19
  + Visit 5 - 2022-01-18 to 2022-01-21
* Baiama (coordinates 7.83708 N, -11.26845 E)
  + Visit 1 - 2021-07-03 to 2021-07-07
  + Visit 2 - 2021-10-22 to 2021-10-25
  + Visit 3 - 2022

Rodents are trapped at up to 6 distinct trap sites per village with up to 49 traps per site. Data was previously collected on this group of [data collection forms](https://drive.google.com/drive/folders/1yE_JAThq-DM9y5yvtSPMgM2ezMcZ_zyO?usp=sharing). Since June 2021 all data has been collected on digitised versions using electronic pads/phones in the field through the [ODK](https://opendatakit.lshtm.ac.uk/lshtm-odk-servers/) platform all digital forms are encrypted locally on the device and sent to a server hosted at LSHTM. The .xlsx versions of the ODK forms are available on the OpenScience Framework [project page](https://osf.io/usjrd/).

trap nights across these visits have been completed. Study visits are scheduled four times per year (every 3 months).

Remote sensed land use classifications derived from Jung et al. and ground-truthed through input from local communities guided the selection of the trapping sites at the selected study villages. First landuse classifications for the Lassa virus endemic region of Eastern Sierra Leone was mapped with the distribution of different land classifications visualised to aid selection of adequate coverage of land types (Figure 1.). The study region was the environs of Kenema, seen as the purple region in the lower left part of the map. Subsequently the land use classifications of each study village was matched to identify potential sites to produce adequate coverage by village (Figure 2-7).

The first study village was Lalehun, a village north of Kenema, on the road between Panguma and Tonga. This village of ~1,000 inhabitants is surrounded by agricultural land with seasonal crop production. Some areas of secondary forest and fallow land remain and are the location of cultivation of cacao or used for bushmeat hunting.

Seven trap sites have been used at this village.

The trap locations are shown overlayed on these habitat maps below.

The proportion of nights trapping in each habitat gave good coverage of land use types in Eastern Sierra Leone.

The trap success rate is around 4% this is fairly acceptable based on the review I did. We have trapped 137 rodents from at least 13 species. The majority of trapped individuals have been shrews (crocidura), *Mastomys sp.*, *Praomys sp.*, *Mus minutoides* and *Lophuromys sp.*. Data from the most recent trapping activities from visit 3 during the rainy season have so far shown a dramatic drop in trap success.

Species accumulation curves have been produced for the first two visits from Lalehun and Seilama. It has not been possible to produce equivalent plots for Baiama and Lambayama due to the low number of individuals/species trapped in those locations.

As we catch an increasing number of individuals we are seeing them clustering within expected habitats.