International Livestock Research Institute

Workshop report

Development of a protocol for the Rift Valley fever case study,

Dynamic Drivers of Disease in Africa: Ecosystems, livestock/wildlife, health and wellbeing project

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List of abbreviations

CDC Centers for Disease Control and Prevention

CICES Common International Classification of Ecosystem Services

CVM Contingent Valuation Methods

DDDAC Dynamic Drivers of Disease in Africa: Ecosystems, livestock/wildlife,

health and wellbeing

DDSR Division of Disease Surveillance and Response

DNA Deoxyribonucleic Acid

DVS Department of Veterinary Services

ECMWF European Centre for Medium Range Weather Forecasts

ELISA European Environmental Agency
ELISA Enzyme linked immunosorbent assay

FAO Food and Agriculture Organization of the United Nations

FEWSNET Famine Early Warning Systems Network

FGD Focus Group Discussion
GEF Global Environmental Facility
GPS Global Positioning Systems

ILRIInternational Livestock Research InstituteInVESTIntegrated Valuation of Ecosystem ServicesIPCCIntergovernmental Panel on Climate ChangeIUCNInternational Union for Conservation of Nature

KEMRI Kenya Medical Research Institute
KMD Kenya Meteorological Department
KNBS Kenya National Bureau of Statistics

KWS Kenya Wildlife Services
LULC Land Use Land Cover
MA Millennium Assessment

MCMC Markov Chain Monte Carlo methods

NASA National Aeronautics and Space Administration

NDVI Normalised Difference Vegetation Index

NGOs Non-governmental organizations
PCR Polymerase Chain Reaction

RVF Rift Valley fever RVF Rift Valley fever

TRMM Tropical Rainfall Measuring Mission

UoN University of Nairobi
WHO World Health Organization
WWF World Wildlife Fund

Summary

The International Livestock Research Institute (ILRI), in partnership with the Kenya Medical Research Institute (KEMRI), Institute of Anthropology, Gender and African Studies, University of Nairobi (UoN), Department of Veterinary Services (DVS), and the Division of Disease Surveillance and Response (DDSR), Ministry of Public Health and Sanitation is implementing a Rift Valley fever case study that aims to evaluate linkages between climate and land use changes with occurrence and transmission of RVF and other infectious diseases associated with febrile symptoms in humans. This is one of the case studies funded by the project: *Dynamic Drivers of Disease in Africa: Ecosystems, Livestock/Wildlife, Health and Wellbeing*.

The case study held a workshop on 30th August 2013 to:

- (i) assess whether the proposed case study activities and workplans align with the project objectives and hypotheses,
- (ii) identify ways of integrating activities across themes and teams, and,
- (iii) review the expected results and ways of utilizing them to improve health and wellbeing of the target population.

Participants were drawn from the partner institutions (names and affiliations are outlined in Annex I). Key observations made at the meeting include:

- The case study sites were confirmed to be Bura and Hola irrigation schemes in Tana River County and Ijara and Sangailu divisions, Garissa County.
- Progress that had been made with the implementation of the case study was reviewed. This
 included the development of the situation analysis report and the analysis of secondary data.
 The secondary data/analyses that were reviewed include:
 - Secondary data obtained from hospitals in the study sites suggesting that malaria, brucellosis and typhoid are the most prevalent febrile infections in the area;
 - Spatial data on land use changes, the distribution of large mammals and settlements between 1970s – 2000s showing a gradual reduction in the density of elephants, elands and other large herbivores in these areas over the period. A final report on this work will be submitted in December;
 - A dynamic systems model that has been developed for evaluating RVF transmission dynamics which illustrates that standing water masses promotes RVF endemicity if conditions that favor RVF virus transmission are maintained (e.g. availability of reservoir hosts, etc.);
 - Initial data from focus group discussions (FGDs) that were used to pretest the FGD checklists. Types of ecosystem services captured during these sessions were highlighted;
- The case study objectives, study design and activities under each of the five project themes
 adopted by the case study (i.e., local system contexts and interactions, disease-ecosystem
 interactions, multi-scale drivers, social, economic and environmental values and political

- economy of knowledge and policy) were discussed in detail. The outcomes of these discussions are discussed in Chapter 2 of the report.
- The case study implementation work plan and the teams that would be involved in the implementation of activities were formulated.

1 Introduction

Anthropogenic land use changes cause unprecedented transformation of ecosystems, modifying the types of ecosystem services that affected ecosystems can provide. The conversion of natural habitats to crop land causes a decline in regulatory and supporting services, leading to a reduction in the capacity of affected ecosystem to control disasters such as floods, soil erosion and the emergence and transmission of infectious diseases. Climate change can accelerates these changes by altering biodiversity, vegetation communities, biome boundaries and animal habitats (Intergovernmental Panel on Climate Change [IPCC], 2007).

The DDDAC project evaluates the effects of climate and land use changes on the occurrence and transmission of emerging infectious diseases, including Rift Valley fever (RVF) in Kenya, Lassa fever in Sierra Leone, trypanosomosis in Zamba and Zimbabwe, and Henipa virus in Ghana. The RVF case study is being implemented by the International Livestock Research Institute (ILRI) in collaboration with the Division of Disease Surveillance and Response (DDSR), Kenya Medical Research Institute (KEMRI), University of Nairobi (UoN), and the Department of Veterinary Services (DVS). The case study will use two sites with contrasting land use patterns — one where irrigated agriculture is the main economic activity and the other with nomadic pastoralism (Annex II). The case study hypotheses are:

- The creation of new permanent water masses through irrigation alters vector biodiversity and abundance, populations of livestock and humans and frequency of interactions between hosts and vectors, driving RVF emergence and transmission;
- The occurrence of RVF in these novel, relatively intensive, systems has impacts on health, wellbeing and economy that differ quantitatively and qualitatively from impacts in minimally altered ecosystems.

The case study adopts the five research themes described in the DDDAC project document:

- (a) Local system contexts and interactions
- (b) Disease-ecosystem interactions
- (c) Multi-scale drivers
- (d) Social, economic and environmental values,
- (e) Political economy of knowledge and policy.

The case study initially planned to focus on RVF. However, the number of pathogens to study was later expanded to include Chikungunya virus, Crimean Congo Hemorrhagic fever virus, Dengue fever virus, West Nile virus, malaria, *Brucella* spp., *Leptospira* spp., and *Coxiella* spp in order to:

- increase the sensitivity of the study on its attempts to evaluate the effects of land use changes on infectious disease occurrence and transmission, and
- (ii) generate evidence on the types of pathogens that should be considered for differential diagnosis of febrile syndromes in humans.

This workshop was convened just before the case study field activities were commenced. Its objectives were:

- (i) to assess progress made in the implementation of the case study,
- (ii) to assess the proposed case study activities and workplans against the main project objectives and hypotheses,
- (iii) to identify ways of integrating activities across themes and teams, and,
- (iv) to review the expected results and ways of utilizing them to improve health and wellbeing of the target population.

Discussions were structured by theme (see the workshop program given in Annex III), focusing on objectives, design, activities and progress that had been made in the implementation of activities.

2 Research themes

2.1 Local system contexts and interactions

2.1.1 Objectives

The theme will utilize participatory and social science methods to assess complex interactions that link peoples' livelihood practices and landscape features with risk and vulnerability to disease. Key questions that will be addressed include:

- How do the local people understand and experience the target diseases, and how do they contribute to the health burden in general?
- How do the local people interact with ecosystems as they pursue their livelihoods and how does this influence the risk of exposure to these diseases?
- How, and to what extent do the local people understand interrelationships between ecosystem change and disease?

2.1.2 Design

Participatory surveys will be implemented at the village level in all the villages that will be recruited for the study. There will be at least 22 villages in the Tana River study sites (Bura and Hola) and 19 in the Garissa County study sites (for Ijara and Sangailu). Each season will involve 6 to 12 persons with diverse socio-economic backgrounds, gender and religion. These surveys will be conducted in two stages:

- Cross-sectional surveys to be implemented between September and December 2013 to characterize the study sites based on livelihood practices, socio-economic status, ecosystems, and level of awareness on the case study diseases;
- ii. Follow up surveys as from January 2014 in a few villages to verify some of the information collected during the cross sectional survey and to follow up any emerging issues.

At the end of these surveys, stakeholder workshops will be held with key informant from the area to formulate participatory models.

Data collected from these surveys will be triangulated through key informant interviews, narratives, observations and local stakeholder workshops. A checklist for the subsequent interviews (stage II) will be developed at the end of the first phase of the study. Activities described below, therefore, focus on those planned for the cross sectional phase of the study.

2.1.3 Activities

Table 1 summarizes the types of socio-economic data and participatory techniques that will be used in the cross sectional survey. A checklist for the initial cross sectional survey has been developed and pretested (Annex IV).

Table 1. A summary of the type of data required and participatory techniques that will be used in the cross sectional survey (not ordered)

Data required	Participatory technique
Livelihood practices	Semi-structured interviews
Wealth classes	
Types of livestock species kept	
Knowledge on infectious disease in livestock	
Knowledge on infectious diseases in people	
Types of ecosystem services	
Proportion of people in different wealth classes	Proportional piling
Relative proportion of livestock species	
Perceived prevalence and case fatalities of infectious diseases	
Point of provision and consumption of ecosystem services	Resource mapping
Changes in ecosystem structure and services over time	Historical timeline
Distribution of diseases by season	Seasonal calendar
Livelihood activities by season	

2.1.3.1 Wealth ranking and livelihood practices

To determine the relative proportions of people/households in different wealth classes, participants will be asked to identify and characterize wealth classes in their neighborhoods. Classes that could be identified include: rich, moderate, poor, very poor. Participants will then be given 100 beans to distribute to the listed items (wealth classes) to determine the relative proportion of households in the neighborhood that falls within each wealth class using proportional piling technique. Circles will be drawn besides each item on a flip chart to guide the participants on where to place a pile of counters for a class. Probing will be used to understand livelihood practices of the members of each wealth class and the types and quality of social services that they can access (education, health, security, etc.). This activity will determine how exposure to diseases is influenced by livelihood practices and socioeconomic status.

2.1.3.2 Livestock ownership and relative proportion of species

Participants will be asked to determine the proportion of households in each village/sub-location that own livestock. Subsequently, they will be asked to list the types of livestock species kept, and to determine their relative proportions using proportional piling. Probing will be done to inquire why some species are abundant or preferred in some ecosystems and not others. Diseases/syndromes for the most important livestock species will be identified and their relative prevalences determined using proportional piling. For each syndrome/disease identified, probing will be done to determine clinical signs they manifest and proportional piling will be used to estimate case fatality rates. In additional, seasonal calendars will be used to determine the distribution of these diseases by season. Participants will also be asked to narrate how they manage each disease and ways in which veterinary services can be improved in their areas. In cases where the target diseases are not mentioned by the participants, leading questions will be used to prompt responses.

2.1.3.3 Ecosystem services

Participants will be guided to develop resource maps of their areas indicating areas used for human settlements, grazing, crops, watering, roads and service centers e.g. towns, forests, etc. These maps will be used to characterize ecosystems in the villages and how they are used by the local people. Pictures of the local vegetation types will have been produced and used as visual aids for characterizing the ecosystems. These pictures will be superimposed on the participatory maps to aid discussions.

Participants will then be asked to list services/benefits and disservices/harms generated from each ecosystem identified. They will also be asked to state whether the diseases described earlier can be associated with some ecosystems. Historical time lines will be used to determine how those ecosystems have changed over time. The time lines will commence from the 1970s to the present in order to include the time before the irrigation schemes were initiated in Tana River County. Historical timelines will also capture the phases that the schemes have undergone e.g., 1980s to the 1990s when the scheme were at their optimal capacity, 1993 to 2012 when the schemes had collapsed and 2012 to the current period when the schemes were being resuscitated. The types of ecosystem services generated during each of these phases, biodiversity changes (including the types of wildlife present in the area) and disease incidence/distribution will be investigated.

2.1.4 Data management and analysis

All the discussions held with the participants will be recorded using digital voice recorders. Notes will also be made and transferred to a database at the appropriate time. These data will be analyzed using NVivo, a software that has been developed for analyzing qualitative data. For semi-quantitative data (scores), non-parametric descriptive statistics will be used to rank items scored. These tests will attempt to determine whether there would be some agreement between groups on the ranks obtained.

Data obtained from this theme will also be used to develop participatory models described under Synthesis section.

2.1.5 Progress

At the time of the workshop, reconnaissance surveys had been done in the study areas to introduce the project and pretest the FGD checklist. Four FGDs in total had been carried out in Ijara (Hara and Rugha villages) and Bura irrigation scheme (Village 6 and 8).

Observations made in these surveys include:

- The FGD would take about 1 to 1.30 hours to administer. Participatory tools such as mapping, proportional piling, seasonal calenders and time lines could be implemented successfully
- Pictures are very useful for eliciting information on ecosystems and tradeoffs in ecosystem services
- Seasonality: The community in Ijara have four seasons: Haghai (Dry and cool)-January-March, Der (Warm wet season) April-June, Operhet (Dry and hot) July-September and Gu (Cold wet season)- October- December
- The season for crop farmers begins in September with land preparation and planting of commercial maize (Kenya seed) which takes between 90-120 days to mature. Cotton is planted in February to March while subsistence farming is practiced alongside the commercial farming, but in smaller portions of land allocated elsewhere
- There are distinct gender roles among livestock keepers but not for crop farmers
- Ecosystem services vary by ecosystems. It is perceived that land use changes that have occurred in irrigated areas have allowed rodent and primate infestation on cultivated areas

2.2 Disease-ecosystem interactions

2.2.1 Objective

In this theme, ecological and epidemiological processes that influence emergence and transmission of infectious diseases will be identified and analyzed using dynamic systems model. The model, once validated, would be used to analyze multiple scenarios that influence disease risk as well as the effectiveness of intervention strategies.

2.2.2 Design

A simple spatial RVF transmission model has been developed and described under the Healthy Futures project report (pg. 9)¹ to simulate RVF epidemics associated with climate change and variability. The key components of the model are: (i) the environment or landscape on which the simulation takes place, (ii) mosquito vectors, and (iii) livestock. Transmission models for the other target diseases will be implemented through graduate fellowship programs.

¹

The RVF model has been use to obtain initial outputs but it needs to be refined further to fit with the objectives of the theme. The components that will be refined include:

- i. Hydrology needs to be incorporated into the model to better understand how irrigation patterns influences population dynamics of mosquitoes and other vectors;
- ii. Hosts and vectors the model needs to factor in the role of other hosts and vectors given that land use changes alter the density and diversity of hosts and vectors. Consequently, the risk of the disease might decline (due to dilution effect or zooprophylaxis) or increase as biodiversity decline (and hence vectors obtain most of their blood meals from the prevalent hosts)
- iii. Human exposure the current model does not include human hosts. This is because humans get exposed to the disease through multiple processes that are difficult to model e.g. through animal slaughtering, taking care of the sick animals, etc. The risk of human exposure will be estimated based on questionnaire surveys that identify risk practices by gender, age and socioeconomic status.

Empirical studies that will be implemented under this theme to support the modeling work include:

- Ecology to assess the drivers influencing biodiversity changes, water utilization/hydrology,
 and to determine the relative densities of various types of hosts in the study areas
- Entomology to determine relative densities, species diversity, and blood meal sources of mosquito vectors as well as infection prevalences involving the target pathogens
- **Animal health** to determine the incidence and risk factors of zoonotic diseases (RVF, brucellosis, coxiellosis and leptospirosis) in livestock and selected reservoir hosts
- **Human health** to determine the incidence and risk factors of the target diseases in humans

Ecological and entomological studies will be implemented in the village level while animal and human health studies will be implemented at the household level to explore human-livestock interactions and pathways for the transmission of zoonotic diseases.

2.2.3 Ecology

2.2.3.1 Objectives

- To evaluate the effects of land use changes on biodiversity, livelihood practices, soils, water, human demographics and settlement patterns and poverty using historical data covering the period 1970s to the present,
- ii. To ground truth the patterns identified from objective through field surveys,
- iii. To map, quantify and value ecosystem services and tradeoffs at small spatial scales using some of the tools that have been developed for such analyses, such as Integrated Valuation of Environmental Services and Trade-offs (InVEST).

2.2.3.2 Activities

i. Secondary data analysis

Table 2 outlines the type of data that will be used for secondary data analysis. Data on land use and biodiversity changes will be analyzed together with those on disease incidence to determine associations between biodiversity changes and disease incidence.

Table 2. Types and sources of primary and secondary data that would be used for ecological analyses

Data type	Source	Purpose
Land use and land	Multi-spectral satellite images – e.g. LandSat	To assess changes in vegetation
cover data/maps,	Data Archive dating back to 1972	cover - such as species
aerial photos and	Reports from government and non-government	composition, crown cover, forest
satellite imagery	organizations. These include Kenya Wildlife	age and productivity
	Services (KWS), Kenya Forestry Service (KFS),	To analyze the impacts of
	Fisheries Department, Tana and Athi River	conversion of natural landscapes
	Development Authority (TARDA), National	to crop lands
	Irrigation Board (NIB), Water Resource	To assess the degree of
	Management Authority (WRMA), Department	intensification in livestock and
	of Resource Survey and Remote Sensing	settlement
	(DRSRS)	To determine historical
	Other low resolution imagery datasets e.g.	wildlife/livestock distribution
	available from the Regional Visualization and	(wet/dry season) dating back to
	Monitoring System (SERVIR)	1977 or before
Seasonal variations	Historical Climate data from Kenya	To analyze relationship between
in climate and	Meteorology Department (KMD), Tropical	biodiversity, climate change and
other	Rainfall Measuring Mission, NASA (TRMM),	variability, and disease
environmental	European Centre for Medium Range Weather	emergence
variables	Forecasts (ECMWF)	
	Reports of NGOs, local and international	
	conservation and development agencies with a	
	record of project involvement in the area - e.g.	
	World Bank and the Global Environment Facility	
	(GEF), International Union for the Conservation	
	of Nature (IUCN), World Wildlife Fund (WWF),	
	Conservation International, Wildlife	
	Conservation Society, Nature Kenya, Terra Nova	
Environmental and	Government and Non-Government	To analyze changes in livelihood
livelihood change	Organizations reports	practices over the last 30-50
	Famine Early Warning Systems Network	years
	(FEWSNET)	

Human	National population censuses reports/housing	Human population/demographic
demographics,	and socio-economic surveys	trends going back as far as
inter-decadal	Published and grey literature	records can allow
trends	Kenya National Bureau of Statistics, Ministry of	For socio-economic
	planning	characterization of communities
		living in the study sites
Medical records	Division of Disease Surveillance and Response	To be used as an outcome
	Unit (DDSR)	variable while studying impacts
	Local hospitals and dispensaries	of land use change on human
	Centres for Disease Control, Kenya	health
Veterinary records	Department of Veterinary Services (DVS)	To be used as an outcome
	Non-Governmental Organizations such as Food	variable while studying impacts
	and Agriculture Organization, Vétérinairs Sans	of land use change on livestock
	Frontièrs - Belgium, World Vision	health and productivity
Other physical and	National Irrigation Board	To study changes in climate and
productivity data	Department of Veterinary Services	soil properties over time
from the study sites	Ministry of Agriculture	To estimate food and livestock
		productivity

ii. Field surveys to ground truth maps developed

Field surveys will be held within each village/sub-location to ground truth the secondary data collated in Activity 1. This will be done by generating 1 km² grids and randomly selecting a few that will be characterized based on vegetation type and density, topography, soil type, soil moisture, surface water, types of birds and animals, settlement patterns and livelihood practices among others. Judgment will then be made as to whether the primary data collected matches with those obtained from the secondary data sources.

Some of the surveys will also be designed to ground truth information generated from the FGDs conducted under theme 1 (Local System contexts and Interactions). While pretesting the FGD guide for instance, farmers from irrigated areas indicated that rodents and baboon infestation on cultivated areas was high. Some of these animals (especially the rodents) will be sampled to assess their role as reservoirs for the target diseases; the protocol that would be used was developed after the workshop and it is shown in Annex V. Findlay (1931) established that primates, ruminants, and rodents are more susceptible to RVFV than other vertebrate hosts.

iii. Mapping and quantification of ecosystem services

The Millennium Assessment [MA] (2005) framework is one of the earliest and most widely acknowledged system for classifying ecosystem services (as provisioning, regulating, cultural and supporting). The framework, however, presents both the final services of an ecosystem (benefits)

and intermediate services or ecosystem functions in the same dimension, an approach that has been criticized as not being consistent with regard to definition of concepts. It has also been criticized for double counting. The European Environmental Agency (EEA) has developed a standardized framework entitled Common International Classification of Ecosystem Services (CICES) (http://cices.eu/) which addresses some of the limitations associated with the MA (2005) framework. In the new framework, redundancies and overlapping structures have been removed. This classification system will be used in this study since it focuses on "final outputs" of ecosystem services, therefore limiting double counting. In this classification system, supporting services have been dropped since they are regarded as being intermediary services.

Ecosystem services will be mapped and analyzed using ArcGIS and the Integrated Valuation of Ecosystem Services (InVEST) modeling tool using data collected in Activity 1 and validated using information from Activity 2. The levels of these services will be estimated based on land use and land cover patterns. The InVEST models handle habitat quality and rarity data as proxies for biodiversity, ultimately estimating the extent of habitat and vegetation types across a landscape and their state of degradation.

Key assumptions of the InVEST model are:

- areas with high quality habitat will better support all levels of biodiversity and that
 decreases in habitat extent and quality over time means a decline of biodiversity, its
 persistence, resilience, breadth and depth;
- threats to biodiversity emanate from a change in ecological equilibrium resulting from anthropogenic activities;
- legal protection of land is effective and that all threats to a landscape are additive.

Patterns in biodiversity are characteristically spatial; they can therefore be estimated at a defined resolution based on land use and land cover (LULC) maps. This case study will use a resolution of 1km² and will also allow for the determination of external threats and influences, i.e., land use and climate change at different temporal and spatial scales. Variables which will be required for biophysical modelling will include:

- Baseline LULC map –a GIS raster dataset of LULC types on some baseline landscape with a numeric LULC code for each cell;
- Current LULC map a GIS raster dataset, with a numeric LULC code for each cell;
- Threat data a table of all threats to be considered by the model to consider, i.e., land use and climate change effects;
- Future LULC map to represents future projection of LULC in the landscape depending on the threats defined above, and if future projections are to be estimated;
- Habitat types and sensitivity of habitat types to each threat a table of LULC types, whether or not they are considered habitat, and, for LULC types that are habitat, their specific sensitivity to each threat.

Other variables that will be considered include amount of soil eroded/sedimentation, management practices, number and type of pollinators and other insects; number of ungulates, rodents, primates, birds; number of livestock; human population; distances of anthropogenic development (activity) sites from habitats, water, etc.

The analysis will generate habitat quality and rarity maps, degradation maps, land use change maps and biodiversity status which will be interfaced with medical and veterinary (preferably georeferenced) to determine linkages between ecosystem change, biodiversity and health outcomes.

For analysis, species distribution model will be used to evaluate factors that affect the distribution of each animal/rodent/bird species identified.

2.2.3.3 Progress

Maps of the study area indicating the location of major towns, rivers, settlement patterns and distribution of major mammals including livestock over the period 1970s – 2000s have been generated. Figure 1 gives the maps of the study area including settlement patterns while Figure 2 shows the distribution of large mammals over time.

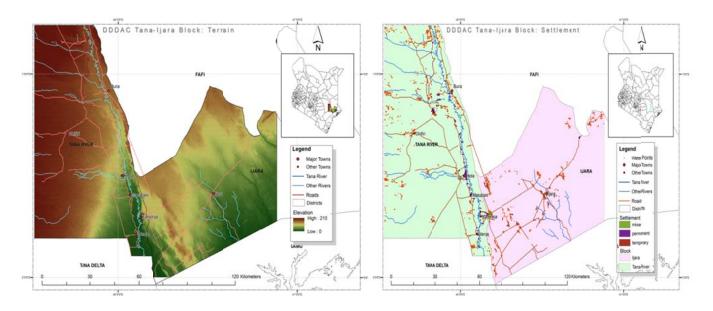
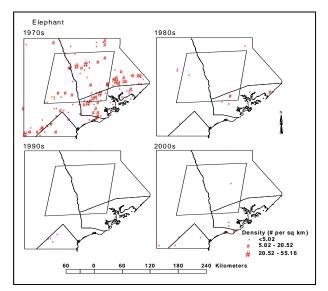
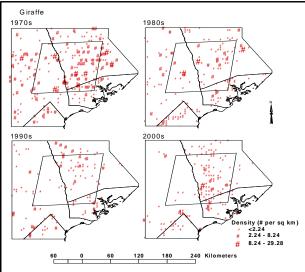
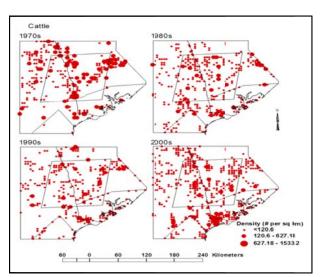


Figure 1: Map of the study area indicating the locations of the major towns, roads, rivers as well as the elevation [A] and settlement patterns [B].







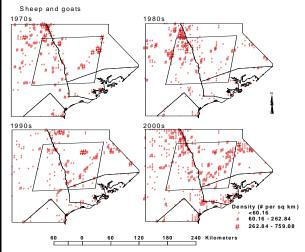


Figure 2: Maps showing the distribution of large mammals - elephants, giraffes, cattle and sheep - in the study area between 1970s and 2000s.

Spatial data showing the distribution of cattle, sheep and goats, buffaloes, elands, giraffes, grant gazelles, impala, kongoni, lesser kudu, waterbucks and zebras between 1970s and 2000s have been mapped out. Some of these maps are displayed in Figure 2. In general:

- The populations of elands, elephants, impala, kongoni and zebras have generally declined in the entire study area over time
- The distribution of giraffes has shifted slightly and seems to be found more in the Somali/pastoral areas than Tana River
- There have been little changes in the distribution of livestock

2.2.4 Entomology

2.2.4.1 Objectives

- Determine species diversity and relative densities of vectors involved in the transmission of RVFV and other arboviruses in the DDDAC study sites;
- Identify and map mosquito-breeding sites;
- Determine and compare the prevalence of the selected arboviruses in the vector populations in the study sites;
- Determine vectors' blood meal sources by area and season;

2.2.4.2 Activities

i. Trapping

Vectors will be trapped using CDC miniature light traps placed in the livestock night sheds, bushes, houses and forested areas. Up to 10 traps will be deployed per village and they will be set in the evening (6 pm) and left overnight and gathered the following morning. Trapping will be done in each village for 3 consecutive days.



Plate 1: CDC miniature light trap that will be used for mosquito trapping

ii. Vector processing

All mosquitoes sampled will be transported to KEMRI's arbovirus laboratory where they will be identified and pooled by species. After this, they will be homogenized in level 2 biosafety cabinets and homogenates centrifuged in a refrigerated centrifuge, and the supernatant cultured. Cultures that show some viral activity will be further screened with the respective QIAmp viral kits (Qiagen, Velencia, CA) using the manufacturer's instructions. Viral fragments amplified by RT-PCR will be gel purified (Qiaquick

Gel Extraction Kit, Qiagen) and sequenced on an ABI Prism 310 Genetic Analyzer (PE Biosystems, Foster City, CA) using a BigDye Terminator Cycle Ready Reaction Kit according to the manufacturer's instructions to confirm the identity of the virus.

iii. Identification of blood meal source

Special resting boxes will be used to trap fed mosquitoes. Sources of the blood meals obtained from (blood) fed mosquitoes will be analysed to determine host preference. Blood meal DNA from the engorged mosquito abdomen will be used for cytochrome b and c PCR analysis. Published degenerate cytochrome b Primer sequences specific for both avian and mammals will be used for PCR amplification (Molaei et. al., 2006) using specific parameters for each primer set. All amplification products will be resolved in 1 per cent agarose gels purified using Qiaquick PCR purification kit (Qiagen) following manufacturer's instructions.

iv). Identification of and mapping breeding sites

Mosquito breeding sites will be identified, characterized and mapped by physical inspection of potential habitats for mosquito larvae. Larvae will be harvested from these habitats and cultured for species characterization. Mosquito presence will be recorded and the GPS coordinates taken for positive breeding sites. Scoring of mosquito types in the habitat will be done to genus level (*Anopheles, Culex and Aedes*).

2.2.4.3 Progress

At the time of the workshop, no field work had been initiated. Preparatory activities were however underway, including establishment of field teams and purchase of field materials.

2.2.5 Human health

2.2.5.1 Objectives

- To determine whether land use changes associated with the development of irrigation schemes
 promote the occurrence and persistence of infectious diseases such as Rift Valley fever (RVF),
 Crimean Congo hemorrhagic fever, leptospirosis, yellow fever, dengue, Ngari virus, coxiellosis,
 brucellosis, and malaria;
- To assess whether the impacts of infectious diseases in irrigated areas (Bura/Hola) differ both
 quantitatively and quantitatively from those observed in a pastoral area (in Ijara/Sanagailu) with
 minimal land use change;
- To build capacity among the local public and veterinary health service providers on surveillance and diagnosis of febrile illnesses. In most cases, the local health centers over-treat for malaria since most febrile infections are often misdiagnosed as malaria;
- Foster the development of one health communities and practices at the community level.

2.2.5.2 Survey design

The study will be implemented in two main phases: an initial cross sectional study to characterize the study sites and identify entry points for a subsequent longitudinal phase that will involve repeated sampling to investigate cause-effect relationships.

Two main sampling strategies will be used:

- hospital-based sampling -- which will involve a systematic random sampling of patients seeking medical attention in the local health centers for febrile illnesses, and
- household survey -- will utilize cluster sampling to determine factors that affect the prevalence of the diseases identified using households as the primary sampling units. Up to five members of a selected household of more than 5 years will be sampled.

In both cases (hospital and household surveys), a short questionnaire will be administered to each participant to determine risk factors and frequency of exposure to pathogens being studied followed by blood sampling. For the longitudinal phase of the study, sampling will be repeated at quarterly intervals to capture seasonal trends.

i. Systematic random sampling in hospitals

At least 2 health centers from each site will be recruited and involved in the sampling of patients seeking treatment for medical conditions that present with fever as one of the symptoms. These centers will include Ijara and Sangailu health centers in Ijara District and Bura and Hola health centers in Tana River area. Within each site (irrigated verses pastoral), 474 patients will need to be sampled to reliably compare the levels of exposure to at least one of the pathogens. This sample size was estimated using the formula (Dohoo et al., 2003):

$$n = \frac{(Z_{\alpha}\sqrt{2\overline{pq}} - Z_{\beta}\sqrt{p_1q_1 + p_0q_0})^2}{(p_1 - p_0)^2}$$
 (Equation 1)

Where:

n is the required sample size per site

 Z_{α} is the value of the Z statistic that corresponds to the desired significance level

$$\overline{p} = \frac{p_1 + p_0}{2}$$
 and $\overline{q} = 1 - \overline{p}$

 Z_{β} is the value of the Z statistic that corresponds to the desired power of the study p_1 denotes the prevalence of at least one of the diseases in the irrigated site and $q_1=1-p_1$ p_0 denotes the prevalence of at least one of the diseases in the pastoral site and $q_0=1-p_0$

Assumptions made for the sample size estimation are:

- The proportions of patients infected with at least one of the pathogens being studies in irrigated and pastoral study site (p_1 and p_0) are 10% and 5%, respectively,
- Level of confidence that the difference between these proportions is not due to chance, Z_{α} , is 95%,
- The power of the study to find a difference in the prevalences, $Z_{\rm g}$, is 80 per cent,
- Correlation between measurements within a time period is assumed to be negligible as patients are expected to come from different villages.

The estimation of the sample size was dine using the *sampsi* command in STATA version 11.1 which is based on the formula set out above (Equation 1). Its output is:

```
. sampsi 0.1 0.05, power(0.8)

Estimated sample size for two-sample comparison of proportions

Test Ho: p1 = p2, where p1 is the proportion in population 1 and p2 is the proportion in population 2

Assumptions:

alpha = 0.0500 (two-sided)
power = 0.8000
p1 = 0.1000
p2 = 0.0500
n2/n1 = 1.00

Estimated required sample sizes:

n1 = 474
```

The number of subjects that will be sampled from each health center per site will be derived using sampling fractions estimated from the relative number of patients served by each center on an average day. Within each facility, a systematic random sampling technique will be used for participant identification and recruitment. This will involve using random sampling technique to identify the first subject, followed by sampling of *n*th individual in the queue depending on the number of febrile conditions a facility handles in a day. Only those patients above 5 years that will be suffering from febrile (malaria-like) illness and would be in stable health status will be included in the study. This condition will be strictly observed in adherence to the World Medical Association declaration of Helsinki which recommends, among other things, that participant selection should recognize the fact that health and wellbeing of the subjects take precedence over research interests.

Subject selection will be done by a clinical officer attached to the project. All the clinical officers and assistants who will be involved in the work will be trained on the study design including subject selection criteria and ethical requirements. Subject selection criteria specifying the symptoms that a patient will need to have to be enrolled to the study (e.g., fever, headache, dizziness, abdominal pain) will be developed. This will be used at the triage to identify potential subjects. On completion of the triage screening, a clinician attached to the project will seek consent from the preselected subjects and only those who would accept the conditions specified on the form and sign it will be recruited to the study. Guardians will need to give consents for patients below 18 years of age. All the consents will have to be counter signed by a witness.

Blood samples will be obtained from the subjects in the health center laboratory after being examined. The patient will be treated as per the hospital's standard operating procedures and a short

questionnaire will also be administered to the subject after treatment or while the patient is waiting for his or her lab results. This process could take a few days to a few weeks to attain the estimated sample size. Facilities required for sample processing and storage (e.g. liquid nitrogen cylinders) will be distributed to all the sampling centers.

ii. Cluster sampling using a household as the unit of analysis

Household surveys will be done to determine the incidence of the diseases being targeted at the community level. A household will be used as the unit of analysis since it thought that some infectious diseases cluster at this level. The distribution of malaria, for instance, is influenced by the type of housing and types of outdoor occupations such as herding cattle. All the members of a selected household will be sampled. For the purposes of this study, a household will be defined as a group of people who live together and share common livelihood activities under a common household head.

The number of units to be used will be determined using the same formula given in Equation 1 though the estimate obtained will be adjusted to account for clustering of observations at the household level. This adjustment will be done using the formula (Dohoo et al., 2003):

$$n' = n(1 + \rho(m-1))$$
 (Equation 2)

Where:

n is the adjusted sample size n is the unadjusted sample size ρ intra-household correlation coefficient m is the average household size

Assumptions used for the sample size calculation are:

- The proportions of patients infected with at least one of the pathogens being studies in irrigated and pastoral study site (p_1 and p_0) are 10% and 5%, respectively,
- Level of confidence that the difference between these proportions is not due to chance, Z_{α} , is 95%.
- The power of the study to find a difference in the prevalences , $Z_{\scriptscriptstyle\beta}$, is 80 per cent,
- Correlation between measurements within a household, ρ , is 0.04 (there is no information on the magnitude of intra-household correlation of these infections 0.04 is therefore a guestimate. An analysis will be done when data become available to determine the significance of using 0.04 in sample size determination), and
- The average size of a household, m, is 5.

Based on these assumptions, the study will use at least 110 households (or 550 persons) in each site (Bura, Hola, Ijara and Sangailu). This was also estimated in STATA version 11.1 as demonstrated below:

```
. sampclus, obsclus(5) rho(.04)

Sample Size Adjusted for Cluster Design

n1 (uncorrected) = 474

n2 (uncorrected) = 474

Intraclass correlation = .04

Average obs. per cluster = 5

Minimum number of clusters = 220

Estimated sample size per group:

n1 (corrected) = 550

n2 (corrected) = 550
```

During the initial phases of the study, a census of households in the study sites will be drawn up with the help of the local leaders including the village headmen. Bura and Hola irrigation schemes for instance have 10 and 12 defined villages within the schemes – it will be easy therefore to enumerate households within each village. Lists generated will then be used as a sampling frame for a random selection of households. Since two irrigation schemes will be involved in the study, the number of households to be selected within each irrigation scheme will be based on proportionate allocation.

Before identifying the households, community meetings will be held in the study areas to introduce the project and to discuss its design. This will include a discussion of the participant selection procedures and the benefits of implementing the research. These meetings will involve the local leaders, partners and project scientists.

At each site, an additional sample of 30 people that are believed to be having relatively higher high risk of exposure to RVF will be drawn and included in the study. These would include herdsmen, butchers, community animal health workers, and watchmen. A sampling frame comprising such high risk people will be made and random sampling technique used to select those who would be recruited for the study.

2.2.5.2.1 *Activities*

Each subject/household recruited for the study will undergo two sampling steps: (i) blood sampling and (ii) questionnaire survey.

i. Blood sampling

Each participant will be adequately prepared for blood sampling after signing a consent form; the form describes the procedure to be performed including the amount of blood to be drawn. They will also be asked to sit in a comfortable position while sampling is being done. They will also be reassured about the need for a technician to wear protective clothing.

Up to 20 ml venous blood will be obtained from patients above 10 years and 15 ml from those between 5-10 years. Preferably, the left median cubital vein will be used and the site to be injected will be disinfected with 70% isoprophyl alcohol just before the procedure is performed. Sterilized butterfly needles and vacutainer tubes will be used and a tourniquet will be applied 3-4 inches above the

venipucture site to facilitate identification of the vein as well as blood collection. Half of the blood sample will be collected in heparinised vacutainer tubes for screening using targeted multiplex PCR techniques while the other half will be collected in non-heparinised vacutainer tubes for serum extraction and serological analysis. After sampling, punctured sites will be bandaged using adhesive tapes.

Blood samples collected using non-heparinised tubes will be allowed to clot and later centrifuged at 3000xg for 10 minutes to harvest serum. Samples collected will be kept in dry ice both in the field and while being transported to ILRI Nairobi where they will be kept in nitrogen tanks until analyzed. Samples will be barcoded and linked to other metadata. Individual subject's identification details will not be included in the general database.

For household surveys, all members of a selected household above 5 years of age will be sampled. Written consents will be obtained from each one of them. For persons below 18 years, consents will be obtained from them as well as their household head. Independent witnesses will also be expected to sign the consent form.

ii. Questionnaire survey

For the hospital-based survey, a short questionnaire will be administered to each subject to identify risk factors for febrile infections being studied. The questionnaire will cover the following topics:

- Participant identification and characterization including, name of the hospital, name of the patient, location, village, gender, occupation, age, level of education, and the size of his/her household
- Potential risk factors of each of the diseases including type of housing used, source of water, ownership of livestock, contact with livestock,
- Previous exposures to febrile infections that required medication,
- Knowledge on ways of preventing each disease,
- Access to public health services

Questionnaires will be coded to correspond with blood samples that will be collected from each patient following the ethical requirements.

For household surveys, a structured questionnaire will also be administered to the household heads as well as to each person sampled after being sampled. The questionnaire will collect data on:

- Ownership of livestock
- Farm/livestock management practices
- Hygiene levels (through observation rather than by asking)
- Exposure factors e.g. contact with livestock, consumption of raw animal products, division of labor
- Other livelihood practices, housing and type of vector control measures used
- Sources of water
- Frequency (if any) of previous febrile conditions and whether medical attention was sought

Prevention of RVF and access to public health and veterinary services

Questionnaires will be numbered and linked to a household.

iii. Laboratory procedures

All the samples will be initially screened using ELISA tests and those found positive will be subjected to further analysis using multiplex PCR techniques. Serological screening will be done at KEMRI using commercial ELISA kits. Procedures recommended by the manufacturers of these kits will therefore be followed. However, some of the samples will be shipped to arbovirus and leptospirosis reference laboratories for more rigorous diagnosis. In all these screening stages, samples will be anonimised as per the ethical requirements described below.

Samples collected from subjects with fevers and those found positive on serology will be screened further using metagenomics approaches to identify possible causes of illness. A metagenomics approach will be used on the selected samples using second and third generation sequencing platforms to characterize biological profiles of selected samples. There will be a possibility of identifying multiple pathogens in individual samples including those not indicated in the list above. This will generate fruitful information indicating the range of pathogens that could be prevalent in the study areas.

iv. Training of research assistants and piloting

Clinicians, medical technicians, other research technicians and postgraduate students who will be involved in the surveys will be trained on the project design and data collection procedures. An initial training will be done at the start of the project in the course of pre-testing the survey tools. This will be done in the research sites and it will take at least 5 days. Topics that will be covered include:

- Methods for subject selection and the need to strictly follow the sampling design
- Communication techniques including ways of approaching the community
- Administration of the questionnaires/focus group discussion checklists and the need to understand the focus of each question
- Data recording and questionnaire/FGD identification systems
- Ethical issues and the need to identify appropriate times when questionnaires can be administered
- Analysis of data generated

2.2.5.3 Ethical issues

Guidelines for ethical conduct of biomedical research involving human subjects in Kenya have been outlined at https://webapps.sph.harvard.edu/live/gremap/files/ke NCST guidelines.pdf. One of the requirements is that biomedical research should be conducted by qualified persons under the supervision of a clinically competent medical person. The public health component of the project will be supervised by Dr. Ian Njeru, the head of Disease Surveillance and Response unit based at Kenyatta

National Hospital. He is a medical doctor registered with the Kenya Medical Practitioners and Dentist Board and a member of International Society of Infectious Diseases.

Sampling will be done by qualified and registered clinicians and medical laboratory technicians based at the local health centres recruited for this work. Other regulatory requirements are captured in the National Council for Science and Technology research permit (based on Science and Technology Act 1979). This act indicates that research on human subjects should conform to universally accepted scientific and ethical standards.

There are a number of ethical issues that have been addressed already e.g., the study design, etc. Three of them (i.e., confidentiality, informed consent, and compensation for time spent on the research) however require special attention.

i. Confidentiality

All the biological samples will be identified using barcodes since this identification system retains its integrity through all the storage and processing stages and provides a reliable scheme for concealing personal information associated with each sample. Each barcode will be associated with a unique number that will be used to identify questionnaires. A separate register of the subject names will be developed and stored under lock and key. Consent forms will also be stored together with the register. Number codes will be used to link various types of data (questionnaire, lab results) in an SQL database that will be developed for data storage and analysis. The database will therefore use codes and not names as the primary record identifier.

ii. Informed consent

Clinicians attached to the project will be responsible for obtaining informed consents. Consents for subjects below 18 years will be obtained from them as well as from their guardians/parents. Each consent form will be signed by the subject and an independent person who would witness and confirm that adequate information will have been provided to allow a subject to make an informed choice.

Information that would be included in the consent forms includes:

- the purpose and objectives of the research and how participant identification will have been done
- The rights of a participant to participate/decline participation
- Procedures that would be undertaken including the administration of a short questionnaire and types and amounts of specimens to be collected
- Management of the data obtained and confidentiality of the information obtained
- Benefits that the research would generate for individuals as well as the entire community
- Risks and discomforts associated with participation
- The fact that results obtained from the screening process would be relayed back to them through their clinicians as soon as they are obtained
- That some of the samples will be stored in the lab for future screening and that these samples will be anonimised

iii. Compensation

Participants will not be paid for participation as this might be mistaken for enticement. However, any monetary costs incurred due to participation, especially travel costs and time, will be reimbursed. It will be ensured that the amount paid, method of payment and timing will not be coercive or provide undue advantage to any party. This will also not be contingent on the participant completing the study. Compensations will also be provided for adverse effects that can be directly linked to participation. Information on types of compensations to be provided will be fully disclosed to the participants while preparing them for enrolment. Participants will also be given contacts for ILRI and AMREF ESRC that can be used to lodge claims or report any malpractices or mistreatments. Such complaints would be handled by independent investigators at ILRI/AMREF who will not be participating in the DDDAC project implementation to ensure transparency and accountability.

2.2.5.4 Gender issues

Attempts will always be made to encourage the participation of the various gender groups in the study e.g. by stratifying focus group discussions by gender so as to capture gender-specific issues that might get dampened in general discussions. In addition, for the household survey, responses on the gender specific questions obtained from the household head will be validated with the other gender groups in the household before on completion of the interview. The main respondent will be notified in advance that some of the responses will have to be corroborated some of his/her family members.

2.2.5.5 Data analysis

Data will be stored in a SQL database with limited access rights. Data will be disaggregated by gender and other variables to allow for risk factor analysis. Statistical and dynamic systems models will be used to analyze the data. Statistical models will be used to identify risk factors and estimate transmission parameters while dynamics systems models will be used to extrapolate findings from the case study. Multiple models will be developed for the various target diseases as outcomes as well as with combinations of various diseases to determine risk factors for multiple infections. Random effects logistic regression models will be used so as to account for clustering within households as well as repeated sampling by area. Predictors that will be considered include:

- Area-level factors: e.g., land use, weather variables, population density, prevalent wildlife species, etc.,
- Household-level factors: e.g., livestock kept, size and composition of household, housing, use of preventative measures, education level and socio-economic status of the household head, etc.
- Subject-level factors: e.g., age, sex, education level, occupation, vaccination status, etc.

Qualitative data indicating drivers for disease occurrence will be coded and used as predictors. In addition, data obtained from FGDs will be used to build causal web models; such models can be programed as qualitative/participatory models that can be used to analyze qualitative scenarios.

2.2.6 Animal health

2.2.6.1 Objectives

- To determine the prevalence of RVF virus in livestock using both antigen and antibody detection techniques;
- To determine the prevalence of other zoonotic pathogens, mainly *Coxiella burnetii*, *Leptospira* spp and *Brucella* spp., that are important for differential diagnosis of RVF in humans;
- To investigate the impacts of RVF on livestock productivity building on the work that has been done in the past that focused on the impacts of the 2006/7 epidemic.

2.2.6.2 Design

All the livestock owned by households selected for the human health's household survey will be screened for RVF virus, *Brucella* spp., *Coxiella* spp. and *Leptospira* spp. The same households will be used for both human and animal sampling to permit the determination of human - animal interactions that promote the transmission of these zoonotic diseases.

The minimum number of household to use has been described above to be 110 per site. Each of the four areas (the two irrigation schemes in Tana River district and two pastoral sites in Ijara district) will be treated as independent sites given that they have contrasting ecological features – Bura has a denser vegetation cover and many water reservoirs than Hola and Sangailu is closer to the Boni forest that Ijara. This implies that a total of 440 households - 220 in Tana River district and 220 in Ijara district will be used.

2.2.6.3 Activities

For blood sampling, up to 20 ml venous blood samples will be obtained from the jugular vein from adults and 10ml from your animals less than 6 months. Half of this sample will be collected using non–heparinised vacutainer tubes for serum preparation while the other half will be collected using heparinized vacutainer tubes. Clotted blood (in non-herparinised tubes) will be centrifuged and serum harvested into cryo tubes. Both serum and whole blood and serum samples will be transported to ILRI in dry ice. Standard serological tests will be used for screening the serum samples while metagenomic approaches will be used to screen the whole blood.

Sampling forms will be completed in each household for each animal sampled indicating the sex, age, breed, body condition score of the animal.

Laboratory procedures provided by the manufacturers of the test kits that will be used will be followed. Kits that will be used for serological tests include:

- Serion Elisa classic Coxiella burnetti Phase I IgG, Wurzburg Germany for Coxiella burnetti
- Svanovir Bruella-Ab C-Elisa, Svanova Biotech Sweden for Brucella abortus

- Panbio IgM Leptospira IgM Elisa Australia/New Zealand for Leptosira interogans serovar Hadjo
- BDSL RVF C-Elisa, Scotland for RVF virus

2.2.6.4 Responsibilities:

- The project will also train the technicians on sampling, record keeping and other relevant procedures. These technicians will be required to use protective clothing while sampling. These include hand gloves, overalls, and gum boots, etc.
- All the samples will be analyzed at Biotechnology Unit, ILRI Nairobi. Experts from the Biorepository Unit at ILRI will also train all the technicians involved in the project on sample collection, labeling, shipment and storage.
- The local veterinary officers who would be involved in the sampling will help with the mobilization of farmers for animal screening and in translation.

2.2.6.5 Data analysis

A number of regression models will be used to identify risk factors for each of the study diseases. Analyses will also be done to identify factors that promote exposure to multiple pathogens, focusing on those animals that would be diagnosed with more than one pathogen.

Dynamic systems models will also be used to simulate transmission patterns of these diseases in space and time. These models will be parameterized using data generated from the field work. The models will evaluate linkages between presence of standing water masses in irrigation schemes with the development and persistence of mosquito populations, hence endemicity of RVFV. In pastoral areas, the models will assess how livestock movement dynamics and climate change contribute to RVF transmission dynamics. In both scenarios, socioeconomic factors captured through participatory modelling will be used to guide exposure patterns and farming practices.

2.3 Multi-scale drivers

2.3.1 Objectives

This theme explores empirical relationships between RVF emergence, incidence and spread and socio-economic, environmental and biodiversity features of a landscape. This work will be done at the national level.

2.3.2 Design

Previous work on this topic has utilized limited socioeconomic, environmental and biodiversity data to model the occurrence of RVF in Kenya. The types of data that have been used (both as the outcome and predictor variables) are outlined in Table 3.

Table 3. Types and sources of data that have been used for geostatistical analysis of RVF occurrence and distribution in Kenya

Variable	Description (Source)
RVF outbreak data	Surveillance data (Department of Veterinary Services, DVS)
Livelihood zones	Livelihood practices (FEWS NET)
Land cover	Global land cover data (FAO)
Precipitation	Monthly minimum, maximum and average (ECMWF)
NDVI	Monthly average, minimum and maximum values (SPOT VEGETATION)
Human population	Human and household census (KNBS)
Elevation	CGIAR Consortium for Spatial Information

There are some limitations associated with the use of surveillance data on RVF outbreaks in the analysis for multi-scale drivers. First, the available data have low resolution since outbreak sites have not been georeferenced; instead, data is presented at the area level. Secondly, the distribution of the data corresponds with the surveillance effort, implying that areas that did not report outbreaks are least represented. In this case, some of the areas might be erroneously classified as being disease-free. These limitations could bias the analysis and predicted patterns of the disease.

In this study, all the known RVF hotspots will be georeferenced and the data generated analysed using species distribution models (SDM).

In addition, the available historical data on RVF epidemics for the period 1931 to 2007 will be reviewed and classified into: (i) areas that have continued to report outbreaks following prolonged/persistent rains (mainly El Nino rains), and (ii) areas that have become dormant/no longer report cases as from the 1990s. After this, a comparative analysis will be done to determine factors that could be associated with continued occurrence of epidemics verses dormancy. Factors that will be considered as drivers for these changes include – land use change including urbanization, intensification of agriculture, increase human population, changes in the precipitation patterns, etc.

2.3.3 Activities

Most of the known and accessible RVF hotpots will be visited, georeferenced and a questionnaire administered to the local leaders to characterize the hotpots by:

- Livelihood activities
- Types of livestock kept and production systems used
- Types of wildlife common in the area
- Land use/vegetation types
- The last time the disease was reported from the area and whether both livestock and human were affected
- Measures that the communities are implementing to mitigate future outbreaks

Secondary data on socio-economics, poverty indices, access to public health and veterinary services will be obtained from the Central Bureau of Statistics for each of the smallest administrative units in the country and overlaid with the disease risk map to determine how these incidences correspond with the disease risk.

2.3.4 Data management and analysis

MAXENT/GARP ecological niche model will be used for the analysis. MAXENT is however more sensitive to variation in the spatial resolution of the data and GARP would be used more brodly.

These analyses will also compare predicted present and future distributions of RVF to determine hotspots that are likely to contract, expand or shift based on land use and other temporal dynamics such as climate change. Further assessments will be made to determine the degree of overlap between the current and future predictions.

Lastly, efforts will be made to compare factors that predict the disease occurrence in livestock verses those that predict the disease occurrence in both livestock and humans. This will utilize the questionnaire survey data that characterize each hotspot as to whether the recent outbreaks that occurred there affected livestock only or both livestock and humans.

Outputs will be mapped in ArCGIS or QGIS software.

2.3.5 Progress

At the time of the workshop, a few hotspots had been georeferenced.

2.4 Social, economic and environmental values

2.4.1 Objectives

- Assess the 'baseline' economic costs of disease impacts on human health and livelihoods;
- Assess the values of key ecosystem services for livelihoods in a local system context (e.g. village or local rural setting) making use of participatory approaches;
- Assess the impacts of existing disease control measures on valued ecosystem services and therefore livelihoods, identifying trade-offs through a qualitative systems analysis involving different stakeholders.

2.4.2 Design

Part of the analyses under this theme will focus on economic costs of the disease as well as the impacts of the existing control measures while the other analyses will focus on valuation of ecosystem services.

2.4.2.1 Economic costs of the disease

The economic costs of disease impacts on the livelihoods of a range of value chain actors will be analyzed using systems dynamics model which combines production, performance, financial, risk and policy data; its conceptual framework is presented in Figure 3. Types of data that will be required to fit such a model include:

- Livestock demographics and population dynamics
- Animal movements data (Off take rate by sex/age class; movements of animals to/from region etc.)
- Elasticities (demand, supply, income)
- Value chain process variables
- Period of time taken between farm sales and market arrivals
- Period of time taken between sales from farms and slaughter (weeks)
- Inventories of meat (weeks)
- RVF epidemiological data
- Market prices
- RVF control costs
- Draught labour parameters

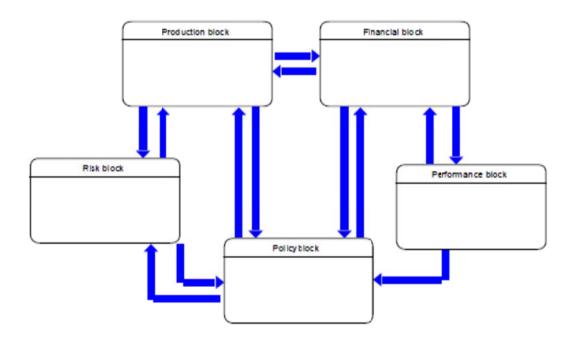


Figure 3. Conceptual framework for the systems dynamics model

The risk block will utilize the epidemiological model that will be developed as part of the disease-ecosystem interactions. System dynamics framework, therefore, provides a framework to integrate epidemiological and economic impacts in one platform. A similar approach has been used by ILRI to analyze the impacts of foot and mouth disease on livestock trade and competitiveness in Botswana.

2.4.2.2 Valuation of ecosystem services

At the community level, Contingent valuation methods (CVMs) will be used to value ecosystem services although they have also criticized as not being credible. Hausman (2012) indicates that until or unless CVMs resolve its limitations (e.g. hypothetical response bias, differences between willingness-to-pay and willingness-to-accept and problems of scope), they should have zero weight in public decision making. Other challenges that have been reported in the valuation of ecosystem services in general include overvaluation and double counting of benefits. Despite these limitations, CVMs will be used in the study to specifically identify demographic and other variables that influence how the local people value a range of ecosystem services. Toman (1998) also indicates that some of the challenges can be addressed if specified baselines and measures of change are well described during the surveys. In addition, we believe that the use of a comprehensive classification scheme (e.g. CICES) would minimize some of biases and double counting errors.

CVMs have two main approaches: willingness-to-pay (representing what respondents are willing to pay to avoid a negative outcome) and willingness-to-accept (representing how large a payment a respondent is willing to receive to accept a negative outcome). The study will use willingness-to-pay approach. Some of the steps that will be taken to develop and implement the survey include:

- developing the research questions and detailed definition of the specific services to be valued and methods of payment,
- developing and pretesting survey tools,
- administering the survey and
- cleaning and analyzing the data.

These surveys will be done when the PRAs (described under Local Systems and Interactions Theme) will have been completed because PRAs will provide a guide on what services to value.

Values generated could be used to estimate ecosystem services at a spatial scale using the InVEST tool.

Activities

- Visual aids will be developed for Contingent Valuation activities
- Questionnaires will also be developed, pretested and administered to the same households selected for the epidemiology work.

3 Synthesis

Data and information generated from most of the activities described in Chapter 2 will be synthesized using participatory and dynamic systems modeling so as to generate scenarios that can be used to engage stakeholders while proposing intervention options. Participatory modeling will be valuable for defining the socio-ecological systems that influence disease occurrence and transmission while the dynamic systems model will attempt to quantify processes within these systems so as to estimate impacts. Participatory models will therefore generate qualitative scenarios on health and wellbeing while dynamic systems models will generate quantitative estimates on disease incidence, impacts and effectiveness of competing intervention strategies.

3.1 Participatory modeling

This work will be informed by the framework developed by the Stockholm Resilience Centre that is available at http://www.espa.ac.uk/projects/ne-i00324x-1/further-information-and-project-documents. Participatory modeling involve graphical representation of the knowledge domain, linkages between variables, events and agents to better understand the structure, behavior and patterns of ecosystems being studied based on perceived knowledge and experiences. Most of the lessons/inputs that will be used for this work will be drawn from participatory studies described under the Local System Contexts and Interactions theme. It is a useful tool for inferring dependencies among variables and causal chains and therefore it adds tremendous value to models that analyze populations or behaviors of individual agents. This model will have the following key components:

- drivers of change (climate, land use, technology, and population density changes) and how they influence landscapes and ecosystems
- ecological dynamics for instance how land use changes affects biodiversity, changes in livelihood practices to cope with drivers of change, exposure to disease
- dimensions on health and wellbeing

A series of stakeholder workshops will be used for the development of the model. A template of such as model is illustrated in Figure 4.

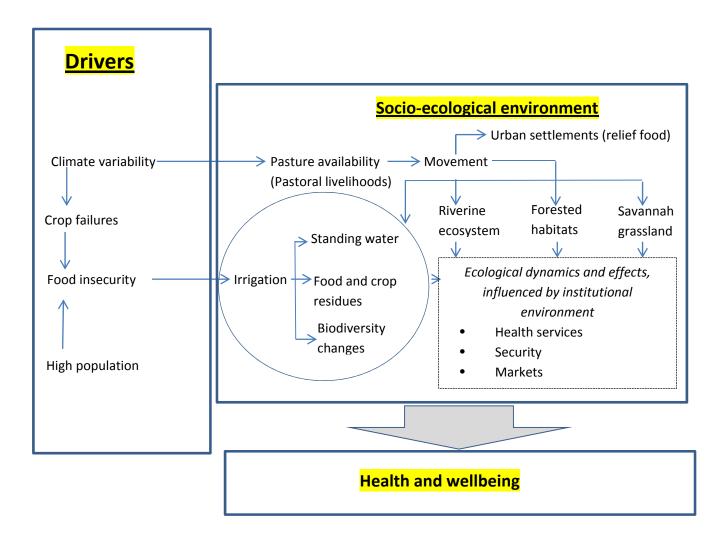


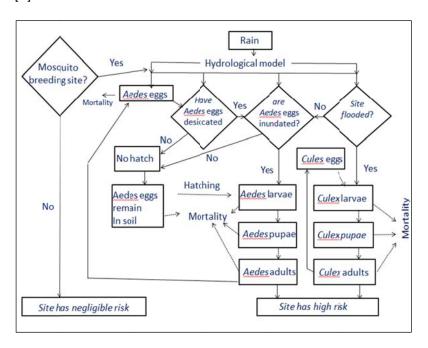
Figure 4: A draft framework for participatory model

3.2 Dynamic systems models

The procedures that will be used to modify a dynamics systems model has already been developed under the HEALTHY FUTURES² project is described under section 2.2.2 - disease- ecosystem interactions. Briefly, the model has two main modules – the vector and host modules and their frameworks are illustrated in Figure 5. Some of the outputs showing epidemic and endemic transmission patters that have been generated from the model are shown in Figure 6. This study hopes to understand the drivers of these dynamics, especially whether water availability in irrigated areas supports endemicity of RVF.

² A project funded by the European Union under the FP7 funds entitled: *Health, environmental change* and adaptive capacity: mapping, examining and anticipating future risks of water-related vector-borne diseases in eastern Africa

[A] Vector module



[B] Host module

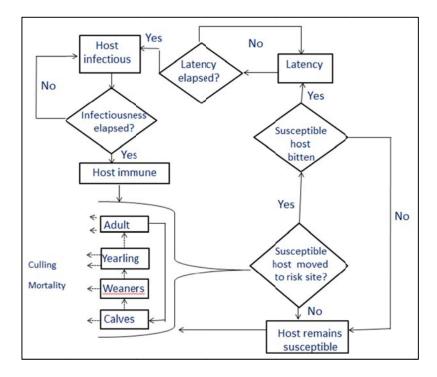


Figure 5: A framework for a Rift Valley fever transmission dynamics model

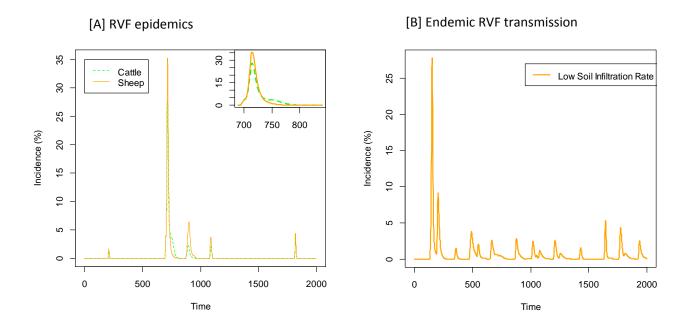


Figure 6: Preliminary outputs from an RVF transmission dynamics model on epidemic and endemic transmission scenarios. The epidemic scenario [A] is driven by precipitation data for 1st January 2005 and 23rd June 2010 (2000 days) obtained from Tropical Rainfall Measuring Mission (TRMM) and the endemic scenario [B] assumes persistence of floods after an initial inundation

4 Workplan

Theme/activity	Month																		
			2013						2014										
	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
Local systems contexts and interactions																			
Development of FGD materials																			
Cross sectional surveys																			
Questionnaire surveys																			
Participatory modelling																			
Targetted surveys																			
Disease ecosystem interactions																			
Ecology																			
Collation and analysis of GIS datasets																			
Field surveys to validate GIS data																			
Quantifiction of services using InVEST																			
Entomology																			
Cross sectional surveys																			
Labortory analyses																			
Animal health																			
Cross sectional survey																			
Laboratory screening of samples																			
Longitudinal studies																			
Human health																			
Ethical approval																			
Training of field personnel																			
Cross sectional survey																			
Laboratory screening of samples																			
Follow up surveys																			
Multiscale drivers																			
Georeferencing RVF hotspots																			
Hazard mapping																			
Vulnerability mapping																			
Sociai, economic and environmental drivers																			
Development of survey instruments																			
Valuation of services																			
RVF impact studies																			
Systems dynamics modelling																			

5 References

Hauseman J. 2012. Contingent valuation: from dubious to hopeless. *Journal of Economic Perspectives* 26(4): 43–56.

IPCC. 2007. Contribution of working group ii to the fourth assessment report of the intergovernmental panel on climate change. M.L. Parry, O.F. Canziani, J.P. Palutikof, P.J. van der Linden and C.E. Hanson, Eds. <u>Cambridge University Press</u>, Cambridge, UK. 976 pp.

Millennium Ecosystem Assessment. 2005. http://www.unep.org/maweb/en/index.aspx. Washington, DC, Island Press.

Toman M. 1998. Why not to calculate the value of the world's ecosystem services and natural capital. *Ecological Economics* 25(1): 57–60.

Annex I: List of participants

Name	Affiliation
Salome Wanyoike	Veterinary Epidemiologist, Department of Veterinary Services
Damaris Mwololo	MSc student, University of Nairobi
John Muriuki	MSc student, University of Nairobi
Salome Bukachi	Medical Anthropologist, University of Nairobi
Caroline Nganga	MA student, University of Nairobi
Millicent Liani	Medical Anthropologist, University of Nairobi
Rosemary Sang	Arbovirologist, Kenya Medical Research Institute
Joel Lutomiah	Technologist, Kenya Medical Research Institute
John Gachoya	Technician, Kenya Medical Research Institute
Francis Mulwa	Technician, Kenya Medical Research Institute
Ian Njeruh	Medical Epidemiologist, Disease
Joan Karanja	Medical Epidemiologist, Department of Disease Surveillance and Response
Purity Kianga	MSc student, University of Nairobi
Bernard Bett	Scientist, International Livestock Research Institute
Delia Grace	Scientist and Team Leader, International Livestock Research Institute

Annex II: Description of the RVF Case Study sites

Two sites with contrasting land use patterns will be used for the study – Bura and Hola irrigation schemes in Tana River County to represent areas that have are currently undergoing rapid land use changes, and pastoral areas in Ijara and Sangailu divisions in Garissa County, representing areas with minimal/slower land use changes. All the 22 villages in Bura and Hola irrigation schemes (10 in Bura and 12 in Hola), 9 sub-locations in Ijara and 8 sub-locations in Sangailu will be recruited for the study.

In general, Tana River district has low rainfall that is bimodal and erratic, ranging between 300 – 500 mm with coastal areas receiving up to 1,200 mm of rainfall. The district is sparsely populated with about 6.2 persons per square km. The human and population census of 2009 indicated that the district has 240,075 persons comprising 119,853 males and 120,222 females. There are a total of 47,414 households in the district with most of them concentrated along River Tana. There are three main communities that live in the district, the Pokomo community, who live along River Tana and practice horticultural farming, and Orma and Wardei communities who live in the hinterland and practice pastoralists. The pastoralists however use the riverine vegetation during the dry seasons while other settled groups practice recession farming following flooding of the main river Tana which flows through the district. It is estimated that 72 per cent of the total population in the district live below the poverty line. They therefore do not have access to capital as property is owned by adult males. Most of the youth are engaged in manual labour, e.g. in irrigated farms. The district has 2 district hospitals, 47 dispensaries, 5 health centres and 4 medical clinics. The doctor: human population ratio is only 1:95,500 (Tana River District Development Plan, 2005³).

Ijara district falls in V-VI agro-ecological zones (semi-arid and very arid respectively) with the southeastern part neighbouring the coastal strip falling in zone IV (semi-humid to semi-arid zone). The district is predominantly inhabited by the Somali pastoralists. The housing and population census of 2009 indicated that the district had a human population of 62,571. At an annual growth rate of 3.5%, this population was expected to reach 73,767 people by 2012. With respect to human health centers, the district has a total of 11 heath facilities including one district hospital that serves a population of 100,000 people, one sub-district hospital and three health centers, each serving a population of 30,000 people and six dispensaries, each serving a population of 10,000 people (Ijara District Development Plan, 2005⁴).

The incidence of poverty is quite high in the selected study sites due to drought, insecurity, changing livelihoods, changing climate and geographical isolation. Most of the natives are primary school dropouts and so have low literacy levels.

³ http://www.ncapd-ke.org/images/stories/districts/TanaRiver.pdf

⁴ http://www.ncapd-ke.org/images/stories/districts/ljara.pdf

Annex III: The workshop program

Time	Session	Convener	Contributors
10.00 - 10.20	General introduction	Bernard Bett	
	Objectives of the workshop		
	Objectives of the DDDAC RVF case study		
10.00 10.00	Research themes and activities		1
10.20 - 10.30	Break		
10.30 – 11.00	Theme 1: Local system contexts and	Sally Bukachi	Salome Wanyoike
	interactions - Specific objectives		Caroline Bernard Bett
	Specific objectivesComponent activities and		bernaru bett
	methodology		
	- Workplans		
	- Expected outputs – by 2013, 2014		
11.00 – 11.30	Theme 2: Disease-Ecosystem dynamics	Bernard Bett	Rosemary Sang
	- Specific objectives		lan Njeru
	- Component activities and survey		Salome Wanyoike
	designs		Shem Kifugo
	- Workplans		Enoch Ontiri
	- Expected outputs – by 2013, 2014, etc		Evans Mwangi
			Damaris/John
11.30 – 12.00	Theme 3: Social, environmental and economic	Salome Wanyoike	Sally Bukachi
	values		Shem Kifugo
	- Specific objectives		Enoch Ontiri
	- Component activities & methodology		
	- Workplans		
	- Expected outputs – by 2013, 2014		
12.30 - 13.00	Theme 4: Political economy of knowledge and	Salome Wanyoike	Bernard Bett
	policy		Ian Njeru
			Sally Bukachi
13.00 – 14.00	Lunch		
14.00 – 14.30	Integration	Sally Bukachi	All
	- Participatory modeling	Bernard Bett	
	- Epidemiological modeling	Francis Wanyoike	
	- System dynamics modeling		
44.20 45.00	- Ecological niche modeling	Dawa and	A !!
14.30 – 15.00	outputs to outcomes:	Bernard	All
	- Behavior change		
	- Capacity building on differential		
	diagnosis in hospitals - One health interventions at		
	community level		
	- Agricultural practices – how to involve		
	the irrigation board, etc.		
	the impation board, etc.	1	

Annex IV: A checklist for the initial participatory surveys

General themes for discussion

1. Livelihood issues

A. Wealth/capital ranking/household diversity profiling

Aims: -To investigate perceptions of wealth differences and inequalities in a village.

- -To identify and understand local indicators and criteria of wealth and well-being.
- -To map the relative number of households in a village in relation to the clusters identified.

Key questions

- 1. Describe the different types of households found in your village (**Probes**: wealthy/rich, poor etc)
- 2. List the predominant characteristics of each of the different types of households in your village. (**Probes**:
 - a. Type of house structure, number of rooms, roofing materials;
 - b. Size and/or amount of land farmed;
 - c. Sources of energy (i.e. firewood, kerosene, solar);
 - d. Personal and household belongings (e.g. mobile phones, bicycles, tools);
 - e. Type and number of livestock;
 - f. Skills and education levels of both parents and children;
 - g. Livelihood activities;
 - h. Food security levels
- 3. Describe the nature of land ownership (*Probe:* subdivision, community owned etc)
- 4. Describe the features of a person who is absolutely comfortable (**Probe** on wellbeing and the characteristics)

B. Main means of Livelihood/Seasonality/gender dynamics

Aims: -To identify the common economic activities in a village.

- -To identify and understand the seasonal changes in the livelihood activities.
- -To establish the gender dimensions in the livelihood activities.

Key questions

- 1. List the main means of livelihood in the community (Probe by gender, social & socio-economic status, religion) (Ranking of most common)
- 2. List all the activities related to the main means of livelihood in the community (For livestock Probe: herding, watering, milking, treating, slaughtering, assisting in the birthing process, preparation and consumption of animal and animal products, caring for diseased animals etc: For crop farming Probes: Land preparation, planting, weeding, harvesting, post-harvest activities e.g drying etc).
- 3. Gender roles and responsibilities in different forms of activities identified in the different livelihoods.

4. Seasonal variations in main livelihood activities by socio-demographic characteristics related to livelihood activities (*Seasonal Calendars; probe for crop farming and livestock keeping*)

5. Ecosystem

Provisioning services/regulating services/cultural services

Aims: -To establish the exiting ecosystem,

-To identify the benefits of each ecosystem, the changes and drivers of these changes.

Key Questions

- 1. Using the **pictures** of the different ecosystems, let the participants identify the pictures that best exemplify/represent their village.
- 2. List all the services (provisioning, regulating and cultural) in the ecosystems representing the village. (With help of visual aids)
- 3. Changes in land use over the years: **Probe**: How has their village/ecosystem changed over the years as far back as they can remember **Probe** for changes in the ecosystems services, diseases, pests etc (*Historical timelines, community mapping/profiling*).
- 4. Establish the drivers/factors responsible for the changes

6. Diseases/health issues

Aim: To Identify the problems/diseases associated with each of the ecosystems identified and the variations over time, annually and seasonally.

To establish community linkage/connection of human diseases to animals

Key questions

- Common causes of deaths in livestock (*Probe*: causes, seasons with emphasis on diseases)
- Common animal diseases in the ecosystems identified (Use of visual aids: listing & ranking/incidence scoring/proportional piling/impact matrix scoring).
- Common human diseases in the ecosystems identified (Use of visual aids: listing & ranking/incidence scoring/proportional piling/impact matrix scoring).
- Probe how these diseases came about and what causes these diseases and how they vary over seasons and over the years (Probe for both human and animals: seasonal /historical timelines & trends).
- Category of animals usually affected by disease in the herd (*Probe:* which category and which diseases/conditions)
- Knowledge of diseases that affect both humans and animals (Listing/ proportional piling/incidence scoring).

7. Rift Valley Fever

Theme 1: Knowledge and Perceptions regarding RVF:

- Describe RVF: local names and meanings, causes, when does the disease occur, risk factors, symptoms in animals and humans, breeds of animals most affected, categories of people most affected, frequency of occurrence.
- Describe how RVF is controlled and treated in the community for both animals and humans: methods known, methods used, and preferred methods.
- Describe the reporting to relevant authorities of RVF risk: to whom is it done, why, what changes or issues signify RVF risk in the community, how is the authorities response perceived.

Theme 2: Factors associated with transmission and spread of RVF

- Risk factors for the occurrence, transmission and spread of RVF: any specific behaviour that predispose humans to acquiring RVF.
- Describe the perceived linkage of livestock practices (herding, milking, and consumption of animal products, residing with animals) with RVF.
- Describe the perceived linkage of the ecosystem with RVF (show photographs of the different ecosystems such as forests, flooded areas, and grasslands).

Theme 3: Impact of RVF on individuals, families and community

- In what ways does RVF affect individuals, families and communities? {Probe; ability to meet basic needs, impact of illness or death on families, community's response to such calamities, Government's response in regard to human and animal health, long term impacts to families and community}
- What measures would you like to see in place regarding the control of RVF?

Annex V: Protocol for Small mammal removal trapping

Equipment and supplies

Baits, heavy rubber gloves, masks, wash water, Sherman traps (box traps), clipboard, indelible markers, pencils, cotton balls (in cold water), sack or shoulder bags, apples, soap for washing hands, white labeling or tape, insect repellant, surveyor's flagging, paper, trap tally forms, habitat assessment forms, plastic collection bags, vacutainer tubes and vials for blood, tissue sample containers, GPS, needles, syringes, blades.

Preparatory procedures:

- Train the field personnel on the necessary safety requirements while conducting the exercise.
 Make them aware of the risks associated with the work and ways of minimizing them, e.g. the need to use of disinfectants, etc.
- Obtain all the necessary protective clothing and equipment
- Obtain the necessary trapping permits from the National Museums of Kenya, KWS, residents,
 NIB and other relevant institutions
- Obtain information on any endangered species in the area and plan on how to release them from the traps as early as possible in the event that get trapped
- Develop a trapping design based on landscape features that can be considered for stratified sampling. This will outline (i) areas where to place the traps, (ii) trap density and placement. The landscape can be stratified into various categories. Those relevant for the study areas include:
 - Residential areas
 - Farm/irrigated lands
 - Dry lands with perennial crops/shrubs
 - Forested areas
- Check the functionality and integrity of the equipment
- Prepare bait (a grain based would be appropriate)
- Prepare printed labels for the mammals- date, animal, number, tissue type using pre-printed adhesive labels
- Attach a strip of white tape on top of every trap
- All traps and baits must be placed on site before dark

Deployment of the traps

- Place traps in areas where there is no frequent disturbances like from vehicles, sidewalks and roads
- Indicate the beginning of the trap line with a small piece of surveyor's flagging, bearing the trap number. If the vegetation is dense, mark the location of the trap with a GPS

- Place the traps in line of 10 to 15 traps at intervals of 10- 15 meters. Maintain constant intervals between the traps for easy of locating the traps on checking trips. Each landscape can have 10 to 15 lines of traps. Keep the traps in place for 3-5 days
- The traps should be carried in a sack, the field people should walk the trap line placing the traps as required. Where necessary, clear the ground slightly by scrapping the soil using one's foot, making sure the mouth of the trap is flush with the ground
- Where possible, place traps near flush piles, fallen, rotting logs, tree bases and any items that may provide shelter. Be sensitive of areas with evidence of rodent activity
- Place each trap in line in only one habitat type
- If it is suspected to be very hot, place traps where there is no direct sunshine or cover them with a piece of cloth.
- After completing placement of traps in each line, complete the trap placement tally form.
- Complete a habitat assessment form for each trap line or group of trap lines in a distinct habitat type. Record the GPS location of the trap lines network, with altitude of the place. Information on distances to houses, roads, rivers etc can be recorded.
- Take photographs of the trapping sites for landscape description

Collecting the Mammals from traps

- Traps should be checked as early as possible, especially in the study sites being used in this study since they are known to be hot most of the time
- Crew members should wear appropriate protective clothing (heavy duty gloves, long-sleeved shirts, socks and heavy shoes)
- Each crew member should check traps he/she set out to avoid losing/missing traps in the field
- Check the traps for evidence of visitation even if the mammal was not trapped. Check for any tissue samples like fur, urine or feces. If such is on the trap, the trap should be placed in a double plastic bag for removal of the tissue material and decontamination. Such a trap should be replaced with a clean one
- When a trap is encountered with a door closed, the trap should be lifted without shaking, then the personnel should stand on the on the direction where the wind is moving from, with the trap at arms- length, push the door open just enough to peer in to check for presence of an animal. If there is no animal in the trap, the adjustment of the door should be checked and the trapped placed back in the trap line. If a non-target species is encountered in the trap, the animal should be released at the site and the trapped put back in the trap line
- If a target species is encountered, the tape on top of the trap should be marked with the trapline number, the trap carefully placed in a plastic bag and the top tied closed. Then the bagged trap should be placed in a second bag which is then tied closed. The double bagged trap should be placed on the ground to be picked on the return journey and then the crew continues checking the rest of the traps on the trapline

- Upon completion of the trapline checking, the bagged mammals should be returned to the collection vehicle and the trap tally form completed (number of capture, number of traps sprung open but nothing trapped and number of missing traps)
- Place collected mammals in a cool area (in the bags) until all traps are checked. The plastic bags must not be re-opened once they have been tied closed
- Traps may be collected to place in a second location if trapping success is more than 10% or they can be left to stay for another night
- The plastic bags containing the animals should be transported in a field vehicle to processing site, avoiding the sun as much as possible
- After placing the animals in the field vehicle, the rubber gloves should be washed in water and soap, then (the rubber gloves are) removed and the hands are washed thoroughly in soap and water

Sample collection, transportation and storage

- Animals, and traps containing animals should be kept in ventilated areas, outside and downwind to avoid inhalation of aerosolized particles
- Identify reliable processing sites e.g. a laboratory. Alternatively if electricity and tap water are not needed, outdoor processing can be used
- Rodents will be euthanized using inhalant anesthetics, weighed and measured. They will then be
 dissected; blood collected via cardiac puncture and heart and liver, lung, kidney and ceacal
 tissues collected and kept in cryogenic tubes with colored screw caps for ease of identification
- While in the field, these samples will be kept and transported in dry ice. In the laboratory, the samples will be kept in liquid nitrogen tanks

Analysis

• Initially, the samples collected will be screened for *Leptospira* spp. using standard PCR kits. The remaining samples will stored for further analyses later