Previous editions of the ZooLinK newsletters have described in great detail the various facets and strands that are being investigated under the ZooLinK project, including the list of 15 disease-causing pathogens that are under investigation including parasitic, bacterial and viral agents in livestock and humans. Further, these newsletters have described the state of the art equipment that have been set up at the field laboratory in Busia, KEMRI and at ILRI to support diagnostic work for the various pathogens. Some of the technologies that have been mentioned include ELISA and DNA-based assays (real time PCR, LAMP and sequencing).

These previous editions have offered highlights on what is currently going on in the project within these study sites and what still remains that would guide further investigation to unravel the disease mysteries that are occurring at the human and livestock interphases in western Kenya. It is also important to note that the project team have so far managed a well designed information feedback loop to the various study participants including local government agencies such as hospitals and health clinics, opinion leaders within livestock markets and communities to share results on zoonoses that have been identified, with important messaging on how the affected communities can respond to occurrence of these diseases to mitigate their impact on the affected communities. In addition to sharing valuable information with the communities on how they can mitigate occurrence of these zoonoses, the information provided to local government agencies is useful in shaping policies around the control of both livestock and human diseases. For example, the lessons learned from the study could be instrumental in shaping the response strategy to the latest challenge of Rift Valley fever outbreak in Northern Kenya amongst pastoralists, who although practising a different type of production system from the sedentary mixed system that is characteristic of western and other parts of Kenya; have strong attachment to livestock rearing activities, including handling of livestock products that gets to the local markets.

It is also important to note that when this letter was being written, a case of rift valley fever was reported in a human patient who was admitted to a health clinic in Siaya County that neighbours Busia with symptoms of excessive bleeding, which case was positively diagnosed at the KEMRI laboratory. This work was highlighted by one of our team member (Cook et al).

Given that food systems are constantly evolving, new challenges also arise that may act as barriers for effective delivery of sustainable health systems. This is reminiscent of activities that occurs at national or territorial boundaries such as is the case at Busia Kenya and Uganda border where effective border controls is important for disease control, but which is often compromised because of the so many shared cultural practises between communities living on either side of the border. For example, there have been

http://www.zoonotic-diseases.org/project/zoolink-project/
reported outbreaks of bird flu in Uganda, but to-date the disease has never been reported in Kenyan chicken flocks despite the continuous cross border supply of poultry products from Uganda into the Kenya market. But a recent study of the occurrence of African swine fever within the Busia agro-ecosystem revealed that the possibility of the disease spreading between the border of Kenya and Uganda was high because Uganda has more pigs produced in the extensive farming systems as compared to farms in Kenya, and the markets in western Kenya tended to consume more pigs from Uganda, which present a gradient through which disease agents can flow between connected systems and farms.

These interactions between human and other systems such as wildlife and livestock systems will be further investigated under the HORN project (http://onehealthhorn.net) that will have some research activities carried out at the Busia field study site and at Oloitokitok field site between the Kenyan and Tanzanian border which has the inter-phase between people, wildlife and livestock systems, all interacting.

A potential area where future research should focus on is emergence of consumer consciousness on animal welfare and its implications on disease control and spread. One of the fundamental freedoms of animal welfare is freedom from pain and suffering which may result from the occurrence of diseases in livestock, including zoonoses. People’s consciousness that animals are sentience beings may greatly contribute to adoption of disease control and prevention policies that would in turn benefit both disease control in human and livestock, especially along the various inter-phases of interactions.

I look forward to engaging the various project team members both in ZooLink and HORN projects in investigating how best to mitigate impacts of these zoonoses under the emerging livestock systems in Kenya, which may offer lessons to the world on the control of zoonoses.

Dr. Joshua Onono takes over as the ZooLinK Co-Principal Investigator from Prof. Ersatus Kan’gethe, following his retirement at the University of Nairobi.
Dr. Lilian Wambua: Post-Doctoral Fellow–Diagnostic Assays

I am delighted to join the ZooLink team and am certainly looking forward to interactions with all members!

I will be working across the project laboratories in Nairobi and Busia, mainly to develop a diagnostic platform for detection of multiple zoonoses for integration into disease surveillance systems.

I previously worked in ICIPE, developing diagnostic assays for the surveillance of: i) Zoonoses and other emerging diseases from wildlife, wildlife products and insect vectors; ii) Detection of illegally traded wildlife products; and iii) Diseases in livestock forages e.g. Napier grass.

My goal as a researcher is to apply molecular tools to innovate low-cost, field-deployable diagnostic interventions for control of zoonoses.

I hold a PhD in Genetics and management of biological resources (Italy), MRes in Advanced Genetic analysis (United Kingdom) and Bsc in Biomedical Science and Technology (Kenya).

Dr. Daniel Omondi Ong’are: Kenya Field Epidemiology and Laboratory Training Programme (FELTP), Resident

Daniel is a veterinarian by profession working for the devolved unit of government in Tana River County. He heads the veterinary division in Tana Delta (Garsen) Sub County, a position he has held for the last 3 years.

Daniel's core mandate is disease control but also implements disease surveillance exercises and coordinates veterinary public health activities (meat hygiene).

He holds a Bachelor's degree in Veterinary Medicine from the University of Nairobi, 2008 and currently pursuing a master's degree in applied field epidemiology from Moi University School of Public Health.

His main area of interest is One Health. Daniel’s thesis project is on antimicrobial resistant patterns in Escherichia coli isolates producing extended spectrum beta lactamase enzymes in unpasteurized milk sold in Busia. The main aim is to demonstrate the molecular composition of the E. coli strains and associated factors in pasteurized milk sold in Busia and associated factors that cause the spread of these resistant strains along the milk value chain.

Dr. Gerald Munai: Kenya Field Epidemiology and Laboratory Training Programme (FELTP), Resident

Gerald is a veterinarian currently pursuing a master's degree in applied field epidemiology under the FELTP programme in collaboration with Moi University's department of Epidemiology and Biostatistics and a graduate fellow under the Zoolink project at ILRI.

His study seeks to estimate the economic burden of acute gastroenteritis in patients attending health care facilities in Busia County and will be supervised by Dr. Lian Thomas a post-doctoral research associate with the Zoolink project.

I studied veterinary medicine at the University of Nairobi and have particular interest in food safety.
Establishing a serum bank of samples from confirmed cysticercosis positive and negative pigs

This serum bank will serve as a platform for future development and validation of diagnostic tools that will allow for a quicker and more accurate diagnosis of porcine cysticercosis. The disease is zoonotic, meaning that it can be transmitted between humans and animals (pigs). The tapeworm, *Taenia solium*, causes taeniasis in people and can cause abdominal pain, diarrhea, nausea and indigestion. The larval stage of the worm can infect both pigs and people. In people, the larval stage can become encysted in the brain and/or spinal cord, causing neuro-cysticercosis. This is an important cause of acquired epilepsy – a debilitating disease. The signs of the disease in humans include seizures, chronic headaches, dementia, and may result in death.

We have organized to visit and buy pigs from 13 slaughter slabs spread across Busia and Kakamega Counties. The process involves contacting a trader/farmer at the slaughter house to deliver a pig on site. On the day of slaughter, intricate bargaining with the trader/farmer to ensure value for money ensues. This is a complex process given that the pricing is usually fluid, with no clear parameters to determine the price. The prices are usually based on the physical appearance of the pig which requires a lot of experience. Once the prices have been settled, photos of the pig are taken, and demographic information, such as age, heart-girth measurement and back length, are recorded. The blood is collected at ante-mortem and lingual palpation is performed.

The pig is slaughtered and weighted perimortem, and then skinned. This is a source of amusement among the butchers who have christened this ‘naked pig carcass’ as Mbuzi ulaya loosely translating to a ‘European goat’ (Fig. 1). The carcass, together with the head, lungs, liver and diaphragm, are chilled overnight and sliced in the morning. The slices ought to be at least 3mm thick to ensure any cyst present can be exposed. This is laborious process that usually takes 3-4 hours to complete.

The most recent studies carried out in the same region recorded a prevalence of 37.6% using a serological method, and 34.4% by lingual palpation. It is such findings, combined with an increase in pig keeping and consumption, that call for such a study. Currently, there exist several serological tests which detect circulating *T. solium* cyst antigens in humans and animals. Yet most of these tests have poor specificity, leading to a large number of false positives and hence, limiting their diagnostic capacity.

We look forward to share more insights from this project in subsequent newsletters.

Article by;
Dr Maurice Karani, ZooLink Research Assistant and Field Coordinator.

*The project aims to establish a bank of serum samples from confirmed cysticercosis positive and negative pigs.*
Kevin Spiegel, DVM/MPH Student, University of Georgia-USA

I am very excited to get the chance to work with ILRI-ZooLink. I have spent a long time in school learning about the steps that go into field work but now it is my chance to gain the experience.

It was a long road to get to Kenya but, after all the necessary paperwork was signed by both the University of Georgia and ILRI, I finally began my work collecting information about the ongoing zoonotic surveillance system.

My project was to build the data collection tools being used and to identify the scope at which we can measure cost. I spent the first two weeks of my time here in Kenya at the ILRI office in Nairobi identifying the measurable aspects of the ZooLink surveillance system and building the data collection tools. Using Qualtrics I was able to construct multiple team specific tools, that allow me to select the activity being performed by the field teams while simultaneously assigning a time stamp for the task. Using this data we will calculate the time cost for each measurement and identify the tests that are not economically viable for a surveillance system when it expands to the national level.

Since joining ZooLink in Busia I have accompanied the animal and hospital teams into the field to collect samples. While they collected samples, I collected time stamps for each activity performed. After collecting some preliminary data, I calculated preliminary results identifying important measurements to focus on. The goal of these measurements is to determine what measurements are superfluous and can be discarded. If taking a measurement takes a long time, costs a lot of money, and does not increase the probability of detecting disease, then it is unnecessary for surveillance.

Unfortunately, I am now at the end of my time here at ZooLink. I am so grateful for the team making me feel so welcome and embracing my work with open arms. They have accommodated me in so many ways and I cannot say thank you enough. ZooLink has been an amazing introduction to field work and lessons I have learned from this internship will continue to have lasting effects on my career.

Our participation in the ZooLink suite of projects will remain memorable. We have acquired sufficient knowledge and experience through the exposure given to us by ZooLink staff and our participation in the target areas of the project.

Since we joined the project in May 2018, we have rotated among the three functional units of the project, namely: (1) veterinary team who visit the livestock markets and slaughterhouses; (2) laboratory team and (3) clinicians team who visit the health centres. The following report will focus on the veterinary team. It describes the activities carried out therein and their relevance to the project.

A normal ZooLinK day begins with packing the field car with the required consumables a day before the field. Such consumables include: red and purple topped vacutainers, nasal swabs, digital thermometer, heart girth measuring tape, ziplock bags, barcodes, consent forms, faecal pots, gloves, disinfectant, water, coveralls and gumboots etc.

In the field, the veterinary team splits into two groups; one group works at the livestock markets and the other at the slaughterhouse.

Upon arrival, at the livestock market, the animal is randomly selected and the owner identified to seek consent for sampling the animal and to answer a few questions. If he/she agrees, he/she signs two consent forms one of which goes with the animal owner while the other one remains for ZooLinK records.

Before sampling, the animal is humanely restrained to ensure safety of the animal, handler and the person collecting the samples. Physical examination begins before the actual sample collection. Which entails checking for any abnormal discharges from the mouth, eyes, genitals and nose. On the skin swellings and injuries are recorded when present. Nature of the ocular mucous membranes is assessed and recorded, the mouth is checked for any lesions and sores as well the aging is done from the dentation. The pre-scapula lymph nodes are palpated on both sides to ascertain any enlargement. Lifting of the loose skin of the neck is done to test for skin elasticity. The body condition of the animal is scored in a scale of 1-5. The fleece condition is recorded as either rough or normal and a tape measure used to measure the heart-girth to estimate the weight of the animal. The temperature is taken per-rectal.

After physical examination, the actual collection of the samples begins. Blood is collected from the jugular vein into a red top vacutainer (plain blood) for serology and an EDTA-purple top vacutainer (uncoagulated blood) for parasitology and hematology. Nasal swabs (Fig.1) are used to collect swabs from the nose. Nasal swabs are later cultured in the lab and used to test for presence of Staphylococcus aureus. Fresh faeces are collected per-rectal and placed into a fecal pot. The fecal sample is cultured in the lab to determine presence of E. coli, Salmonella and Campylobacter. External parasites like ticks, lice etc. are also collected if encountered.

The same procedure takes place in the slaughter houses but in addition, post-mortem lesions like cysts, flukes, are recorded and collected inclusive of mesenteric lymph nodes from the pigs.

We are glad to declare that our internship has equipped us with adequate disease surveillance skills in the animal field that will help us to extend the knowledge of disease control to farmers and other stakeholders back at home.

"...our internship has equipped us with adequate disease surveillance skills in the animal field that will help us to extend the knowledge of disease control to farmers..."