Among the various pathogens studied in the ZooLinK project is Salmonella species, a zoonosis often associated with foodborne transmission in most circumstances; but here in sub-Saharan Africa some species of Salmonella, especially Salmonella enterica serotype Typhimurium is responsible for severe bacteremic illness among adults with HIV and among children under 5 years of age co-infected with HIV, malaria, sickle cell disease or with malnutrition, with incidence in children ranging from 166-568/100,000 cases pyo. In developed countries the infections caused by similar species but different sequence types are typically self-limiting foodborne illnesses that spontaneously end within 48-72h. So why the difference? The local strains, through unique evolutionary pathways, have lost a repertoire of genes so as to become more host-adapted for humans and hence the tendency to cause invasive typhoid-like disease especially in immune-suppressed individuals. We expect that studies on this organism within the ZooLinK theme on bacterial zoonoses will further unravel the mysteries about its reservoir status in livestock and help track transmission routes and provide data on key risk factors and potential prevention strategies in the farming communities. Just to point out that we now have a number of new initiatives aimed at developing new vaccine candidates for the most common non-typhoidal salmonella species (S. Typhimurium and S.Enteritidis) and these will have a great impact towards the prevention of this life-threatening illness. We have a host of other important bacterial zoonoses under investigation in the ZooLinK project, including Campylobacter, E. coli, MRSA, etc. These studies will no doubt unravel more information regarding local epidemiologic risk factors for emergence and transmission between different ecosystems in the One-Health paradigm. Similarly, antimicrobial resistance, a key component of risk factors for severe prolonged illness, longer hospitalization and increased mortality will be an important part of our studies. We expect to map resistance and are already mapping usage of antimicrobials in our target farming communities and provide critical data on transmission dynamics and possible intervention strategies for improved animal husbandry.

I wish all our field and lab teams continued great success in their work and we look forward to a most successful new year for the project.
Introducing ZoolinkK students

**Dr. Robin C.A. Omedo**
PhD student

Robin is a lecturer at Masinde Muliro University of Science and Technology (MMUST). He holds a BVM degree from the University of Nairobi and an MSc from Kenyatta University.

His PhD research within the Zoolink project will focus on phenotypic and genotypic characterization of non-Typhoidal Salmonella (NTS) serovars from Humans and domestic animals, and determining the risk factors that predispose people in Busia County in Western, Kenya to NTS infections.

**Titus Mutwiri**
PhD student, ILRI graduate fellow

Titus has joined ZoolinkK project as a PhD student on a German DAAD scholarship. Prior to joining ZoolinkK, Titus worked as an assistant lecturer at Kenya Methodist University, Department of Medical Laboratory Sciences.

His PhD research focuses on “Cystic Echinococcosis in Western Kenya: Distribution and Genetic Diversity”. For the next few years, Titus will be working to understand infection of Cystic echinococcosis (CE) in livestock, dogs and humans.

His interests extend to economic losses incurred by Echinococcosis also known as hydatid disease. The study site is western Kenya with ILRI field lab in Busia being the base of activities and laboratory work.

**Tom Turbine Ouko**
PhD student

Tom, is a microbiologist working with Kenya Medical Research Institute Centre for Microbiology Research (CMR). Tom is currently supervising the KEMRI Microbiology thread of the Urban Zoo project. In the Zoolink project, Tom is a PhD student registered with Jomo Kenyatta University of Agriculture and Technology (JKUAT). His research work will focus on genetic diversity, risk factors and drug resistance of Campylobacter from Human and Poultry in Busia, Kakamega and Bungoma Counties in Western Kenya.

**Dr Robert Kiplangat Rono**
FELTP resident

Robert is a medical officer working in the ministry of health. He is currently pursuing a masters in science in Field Epidemiology and laboratory training program, (FELTP).

His thesis project is entitled "Prevalence, risk factors and Antimicrobial susceptibility pattern of Campylobacter infection in Children under five years presenting with Acute Diarrhea in Busia County Hospital, Kenya". This is one of the projects under the ZooLink project.
Challenges associated with tracking the movements of people and their livestock.

By Jessica Floyd, PhD student, University of Southampton

Phase two of this study (detailed in the previous Zoolink newsletter) began in November. Over the last two we revisited 27 households that we collected GPS data from in phase one in order to track the movements of the same people and livestock as we did in July and August of this year. Briefly, this involves visiting randomly selected households in Busia County and asking the participant to wear a small GPS tracker on a lanyard around their neck or alternatively, to keep it in a pocket on their person for one week. During the same visits we also attach an identical GPS unit on a collar to one of the livestock belonging to the household. After the week is over we return to the household to collect the trackers and to ask a few questions about the experience.

Although most people have been keen to participate in the study for a second time, we often hear of challenges they encountered while wearing the trackers. These are nearly always due to other people’s perceptions of the purpose of the trackers and the research. For example, many participants reported that they were questioned by people from other households, which led to participants having to convince other people of the purpose and worth of the study. In the worst cases, participants reported that other people were convinced that the tracker was listening to their conversations, was a bomb or was doing “the work of the devil”. However, it was heartening to hear that in all cases the participant attempted to explain the study to other people, with varying results. Interestingly, the intensity of the questioning by outsiders seemed to be related to the participant’s age and gender: we tended to find that young women wearing the trackers were more likely to be subjected to questioning and (attempted) persuasion to discontinue their participation in the study than others. Nonetheless, participants invariably reported that while others might be doubtful, they themselves remained convinced of the purpose of the study and continued to wear the trackers.

Sometimes it was difficult to find our participants and collect the trackers when the week was up – we would drive to a sub-location up to two hours away from Busia town, only to find that the people we wanted to visit were out and we would have to track them down, mainly by asking the villagers where our participants might be. On the bright side, this also meant that our trackers were out collecting interesting data, and has led to us stumbling upon various events within the villages, including a funeral, a circumcision ceremony and a fishing trip (see photo).

Overall, this second phase of fieldwork has been largely successful!

Fishing in village in Busia

Antigenic diversity in the African trypanosomes *Trypanosoma congolense* and *Trypanosoma vivax*

By Sara Silva Pereira, PhD student University of Liverpool.

Blood sampling

Trypanosomes are extracellular blood parasites, transmitted by the bite of tsetse flies and cause nagana, a wasting disease severely compromising both animal health and livestock productivity in Sub-Saharan Africa. Nagana remains a challenge mainly due to the process of antigenic variation, employed by the parasite for immune evasion.

I came to Busia to conduct a longitudinal experiment on natural cattle infections of *T. congolense* to better understand the process of antigenic switching. With the help of a local veterinary surgeon, we screened cattle across for trypanosomes using thin blood smears and high centrifugation technique and followed the infection in positive animals for a month, after which the animals were treated.

The collected materials will be subject to DNA and RNA sequencing and Mass Spectrometry to characterise the genetic repertoire of the parasites and the antigens expressed over time.
Non-typhoidal Salmonella (NTS) in pigs in Busia, Nairobi and Malawi

By Catherine Wilson.
MRES, Student, University of Liverpool

I am investigating the prevalence of non NTS in pigs in both Kenya and Malawi in extensive, low input production systems. The aim is to determine whether invasive NTS are present in the pig population of three study areas; one rural and one urban area in Kenya and one rural region of Malawi.

In sub-Saharan Africa, NTS is a leading cause of human mortality, particularly in the very young, old, malnourished, or those suffering from co-morbidities such as HIV or malaria.

An invasive NTS serovar has been found to be able to cause severe disease in chickens; suspicion is therefore arising that transmission between humans is not the sole route of spread of NTS, and that zoonotic transmission, especially from pigs, may have a role to play in the epidemiology of the disease.

Should this invasive strain of bacteria be found in pigs, we will assess whether the same serovar clinically affects humans in the same geographical location, using data already gathered from human hospitals. A correlation between the two would indicate that zoonotic transmission may be occurring.

The final part of this study will assess the presence of drug resistance in the strains of NTS isolated from pigs, and whether this bears any correlation to a similar antimicrobial resistance pattern of NTS to that previously detected in humans in the same area. Should antimicrobial resistance be detected, other management techniques for the swine, such alterations in husbandry and hygiene, may be trialed. In the longer-term vaccination development may be a possibility as an important method of preventing zoonotic disease transmission in the study areas, for which research is currently in the very early stages.

For sampling, both faecal and mesenteric lymph nodes samples were collected post mortem from 256 pigs in Busia and 304 pigs in Nairobi. The location in which the pigs were reared, as well as details of signalment, any previous antibiotic treatment if known and the method of transport of the pig to the slaughterhouse, were recorded for each individual pig.

ZELS grant holders workshop will be held in Arusha on the 24th to 25th January 2017.

The annual ZooLink PI meeting will be held in Western Kenya on 7th to 8th February, 2017.

Samples were processed at the Busia Field Lab and ILRI laboratories respectively. Culture and serotyping was carried out to confirm the presence of Salmonella, followed by antimicrobial susceptibility testing to a range of antibiotics. Positive isolates have then been stored for transport to the UK, where whole genome sequencing will be undertaken to identify the presence of any antimicrobial resistance genes.

Once the results have returned, analysis is planned compare antimicrobial resistance profiles of the pig samples to those of humans in the same geographical location, to assess whether zoonotic transmission may be occurring.
