Resistance to trypanosomiasis: a use-case for genome modification in livestock

Steve Kemp
Program Leader
Livestock Genetics

Virtual Workshop Series on Regulatory Approaches for Agricultural Applications of Animal Biotechnology

Thursday 8th October 2020

ILRI
INTERNATIONAL LIVESTOCK RESEARCH INSTITUTE

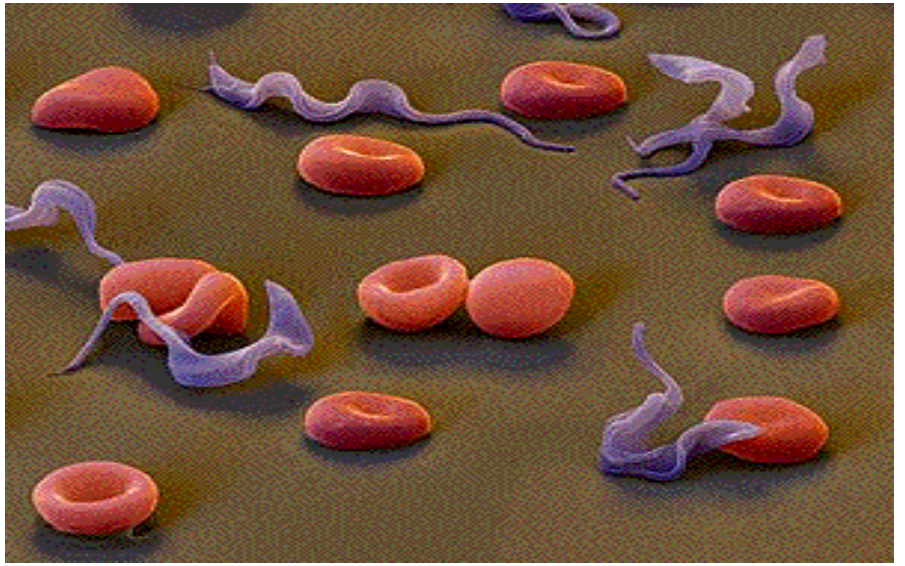
CGIAR

UKaid
from the British people

BILL & MELINDA GATES foundation

Centre for Tropical Livestock Genetics and Health

African trypanosomiasis



- Caused by extracellular protozoan parasites – *Trypanosoma*
- Transmitted between mammals by Tsetse flies (*Glossina* sp.)
- Prevalent in 36 countries of sub-Sahara Africa.

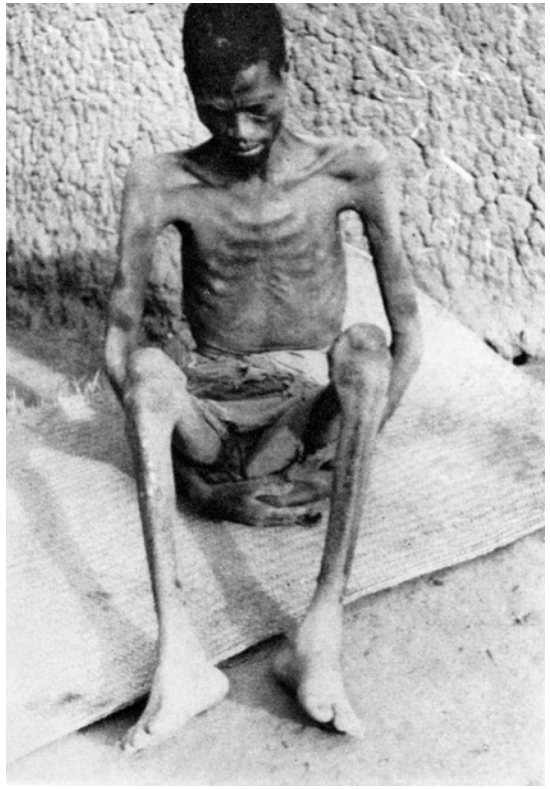
In cattle

- A chronic debilitating and fatal disease.
- A major constraint on livestock and agricultural production in Africa.
- Costs US$ 1 billion annually.

In human (Human Sleeping Sickness)

- Fatal
- 60,000 people die every year
- Both wild and domestic animals are the major reservoir of the parasites for human infection.

Livestock are also reservoirs of human disease

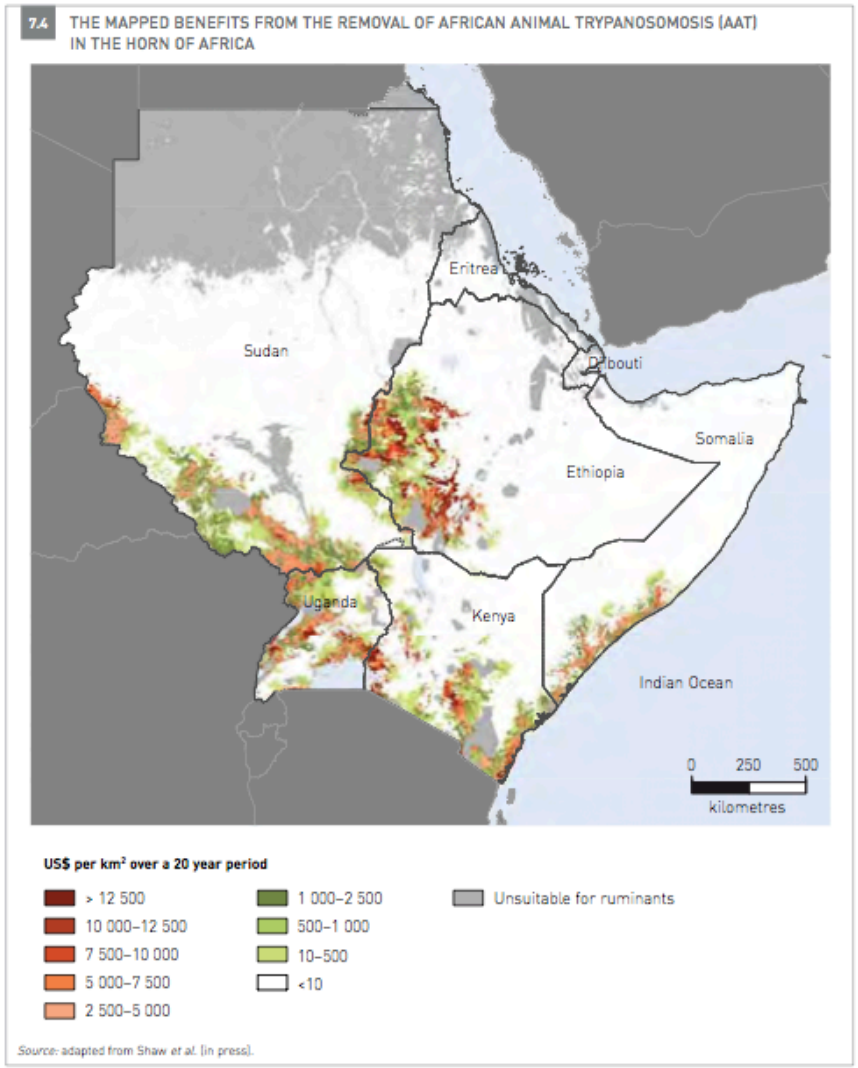


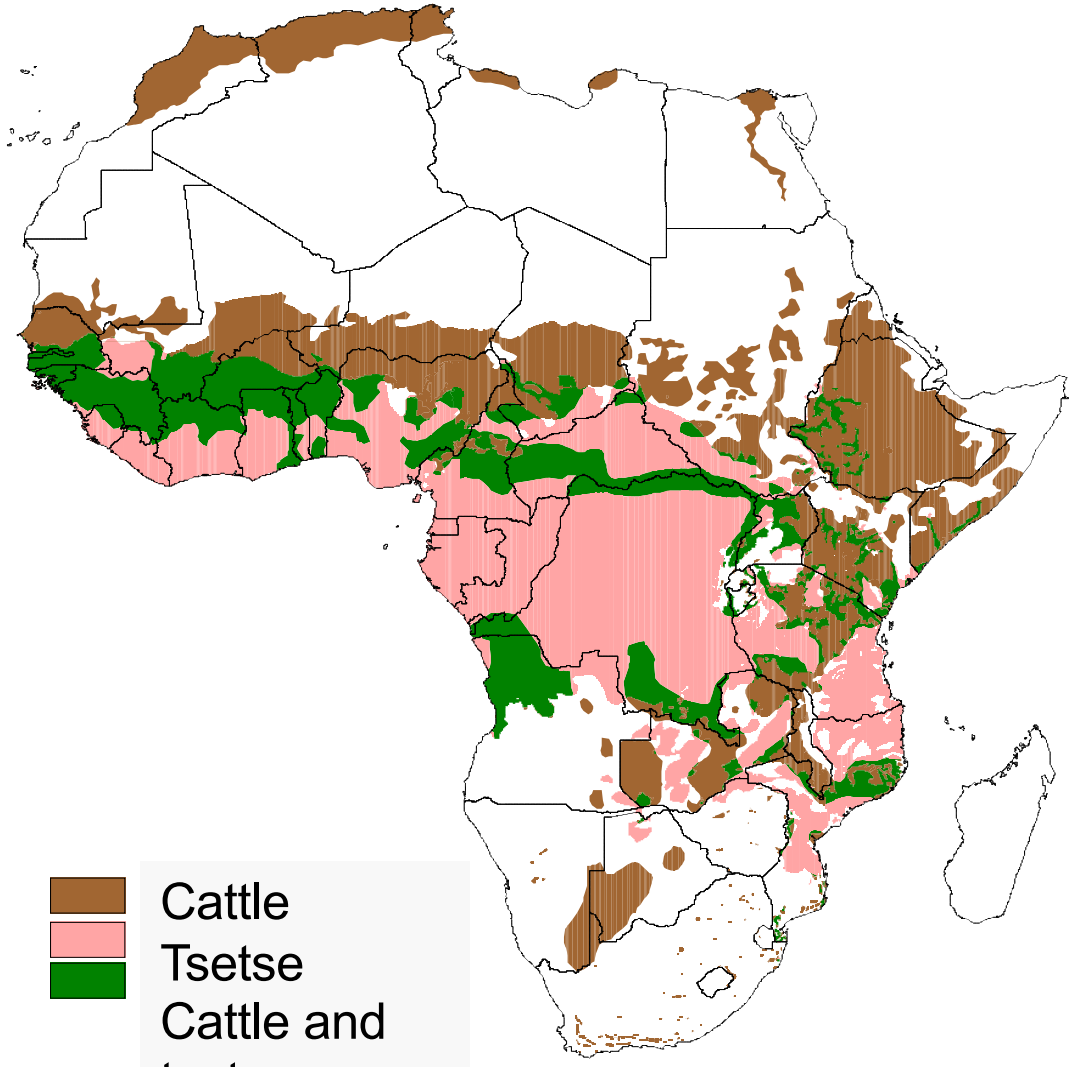
T brucei rhodesiense T brucei gambiense

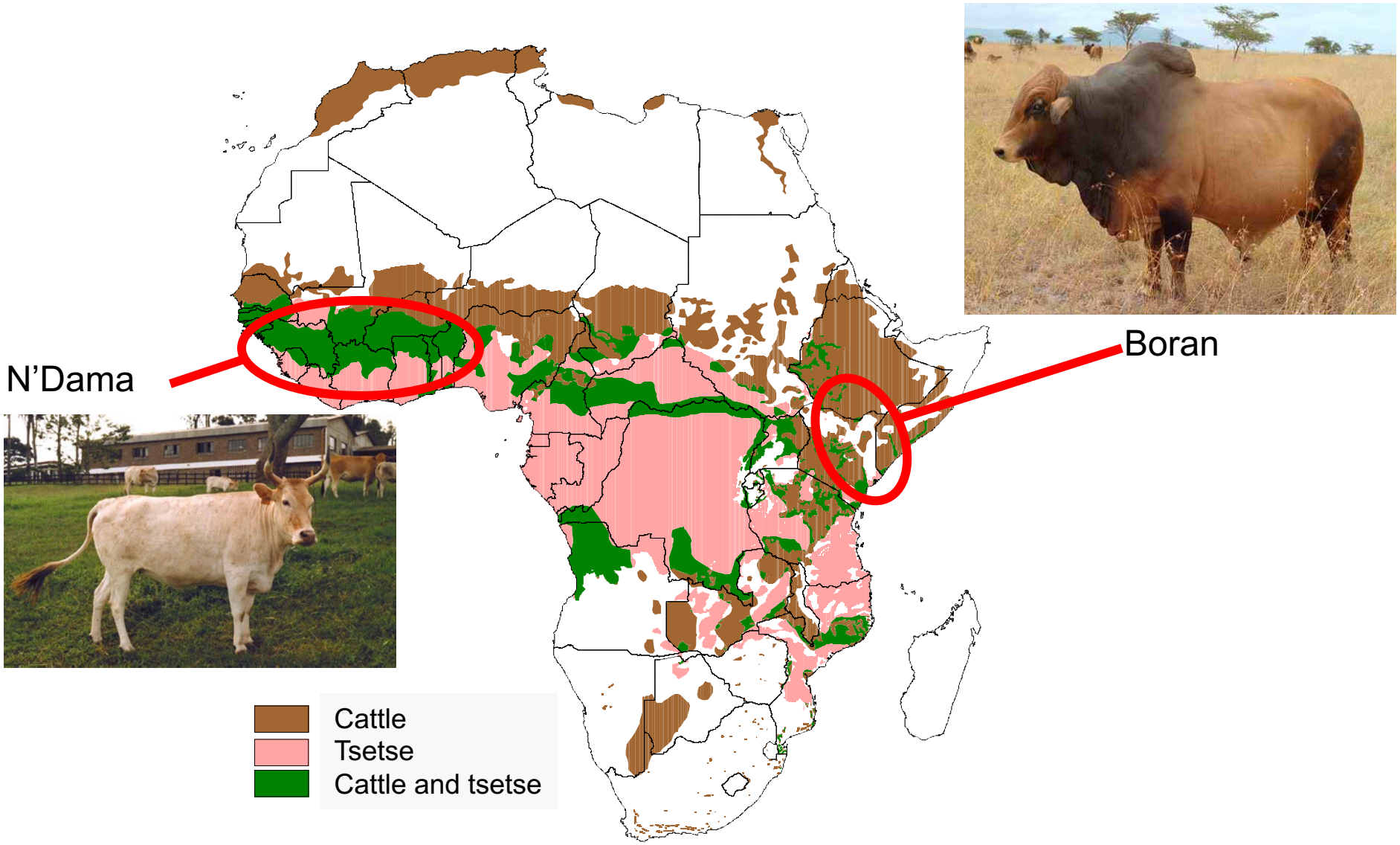
T. congolense,

T. vivax

Trypanosomiasis in livestock costs billions of dollars and excludes livestock from much of Africa

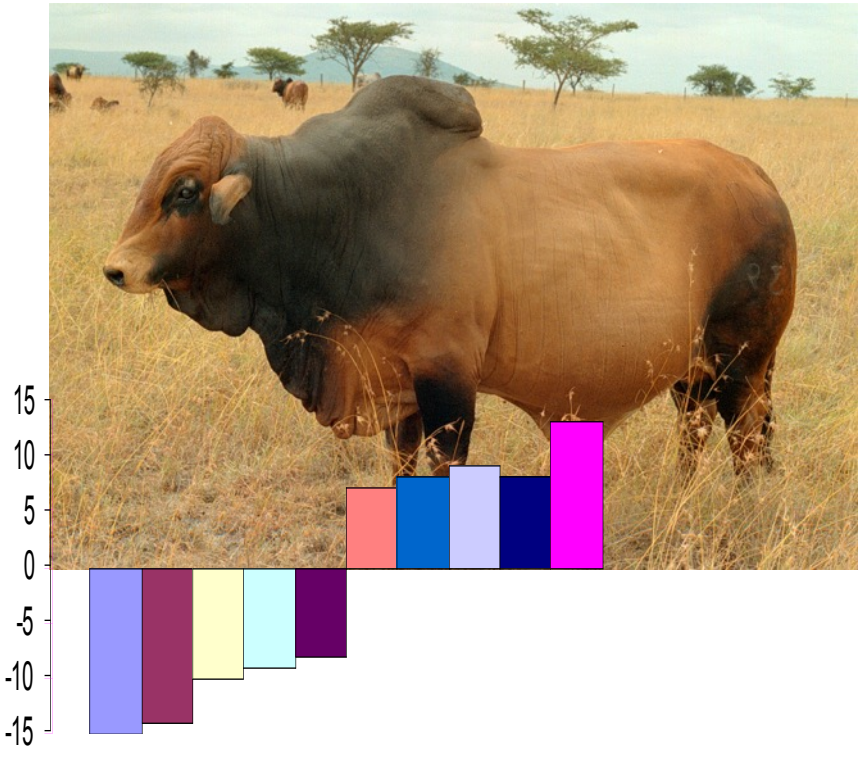




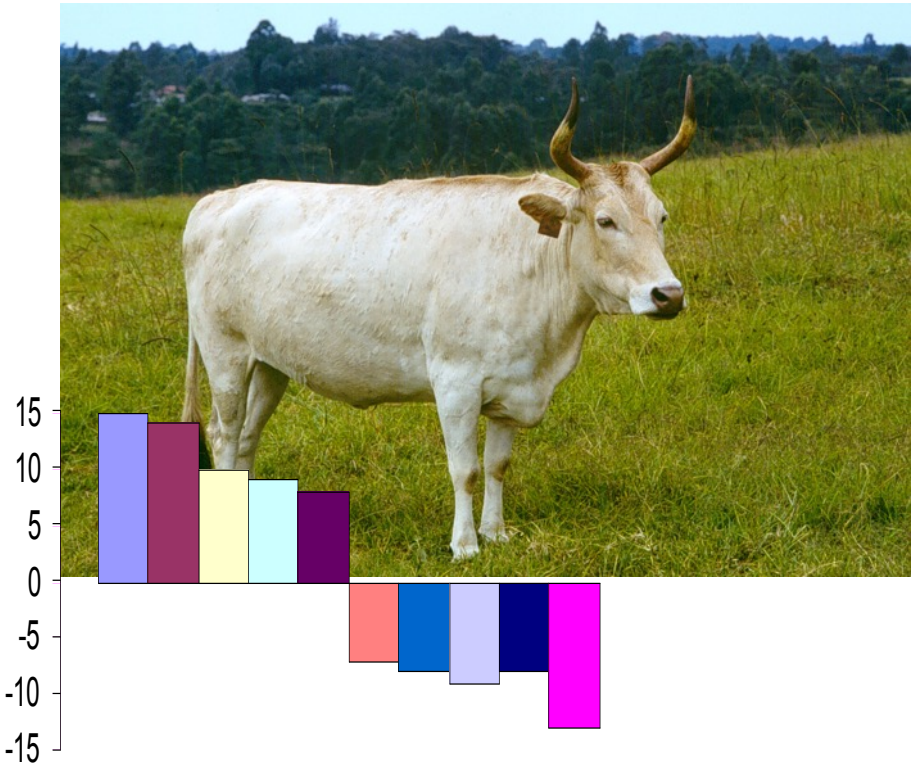

N'Dama
Boran
Cattle
Tsetse
Cattle and tsetse

Contribution of 10 genes from Boran and N'Dama cattle to reduction in degree of trypanosomosis

Boran (relatively susceptible)



N'Dama (tolerant)



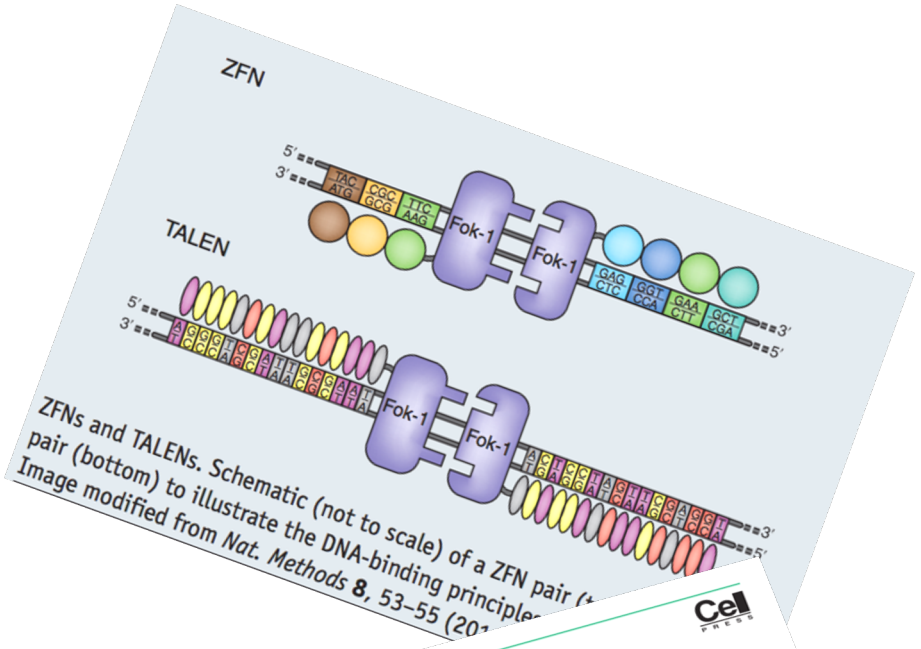
The N'Dama and Boran each contribute trypanotolerance alleles at 5 of the 10 most significant QTL, indicating that a synthetic breed could have even higher tolerance than the N'Dama.

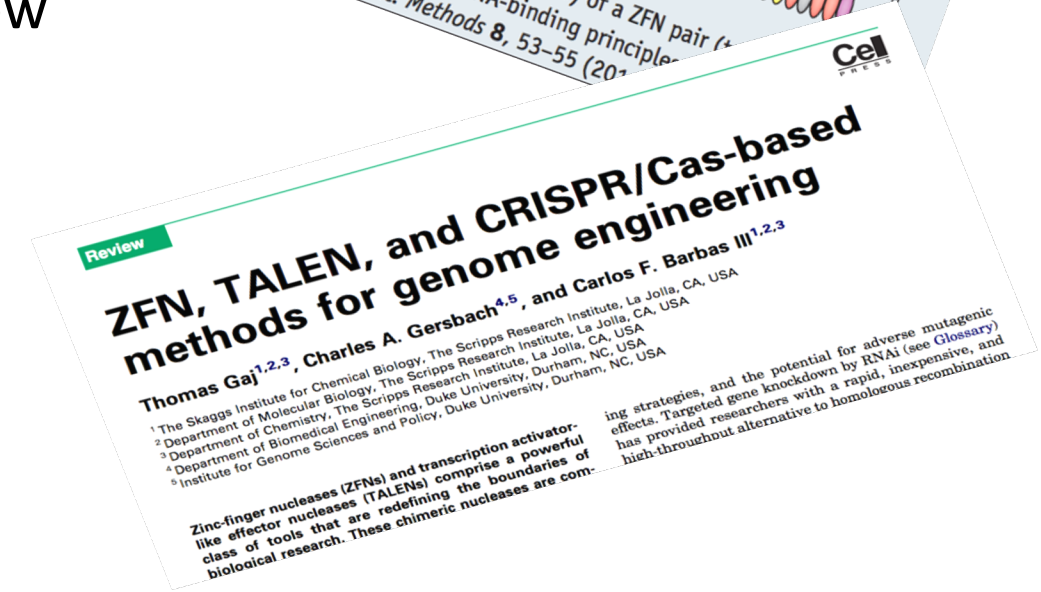
Time for a new search for variation underlying tropical adaptation and productivity

Identify and make use of the genetics underlying natural variation.

There has been no systematic search for the genomic basis of adaptation. Because until now we have had no validation tools and no delivery tools.

New Genome Editing tools change the landscape.





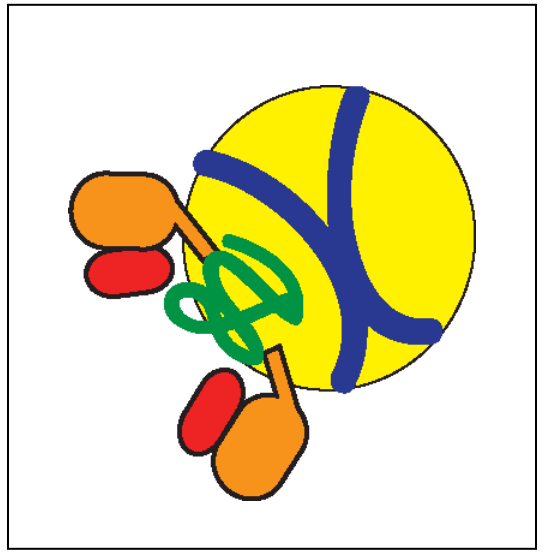
Centre for Tropical Livestock Genetics and Health

UKaid from the British people

BILL & MELINDA GATES foundation

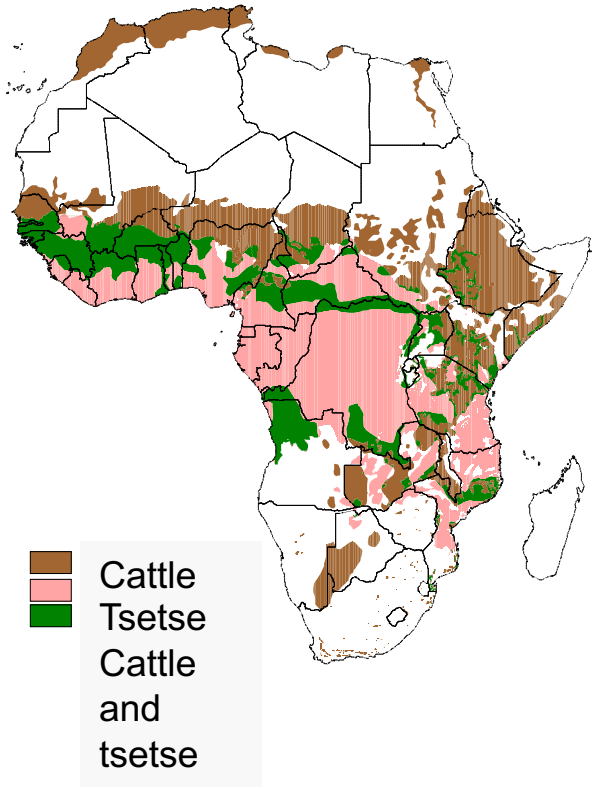
The Mzima Cow Project

A transgenic approach to trypanosome resistance



Primate trypano lytic factor (TLF) has been shown to protect mice from trypanosomes in a transgenic system.

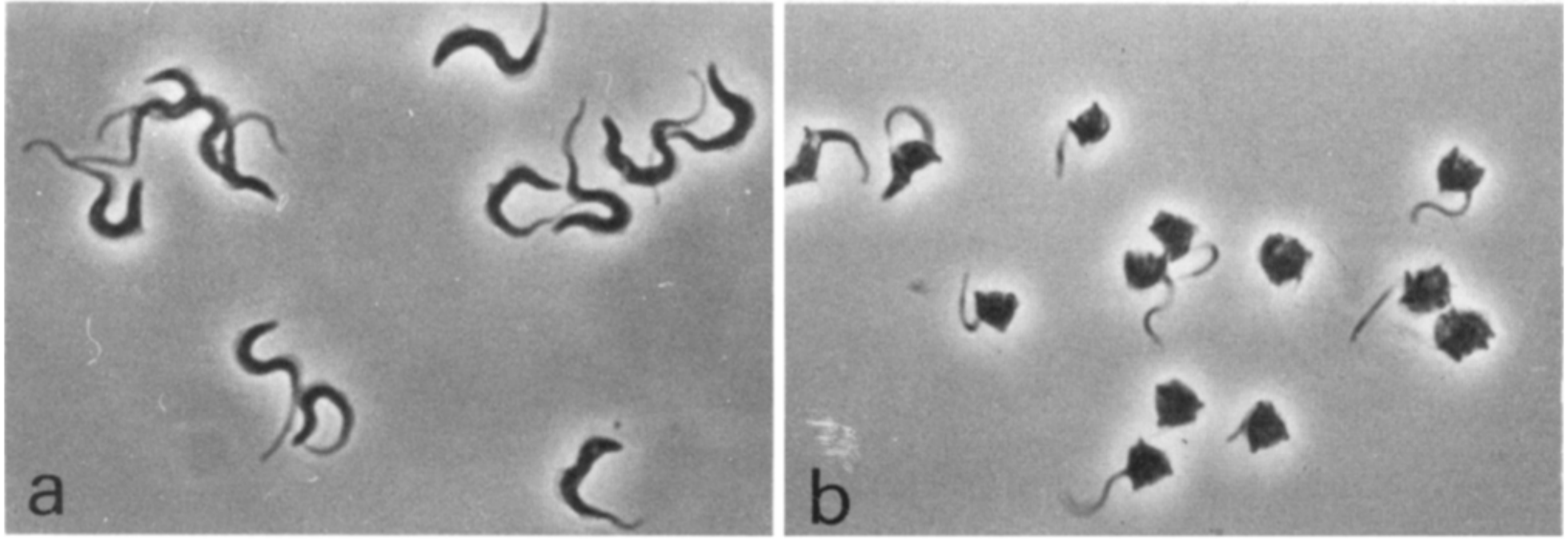
We are attempting to make a cow with a synthetic TLF



Trypanosomes lyse when exposed to primate serum, which contains the pore-forming toxin APOLI

Mouse/Cow/Goat Serum (no APOLI)

Primate serum (with APOLI)



Resistance to trypanosomiasis

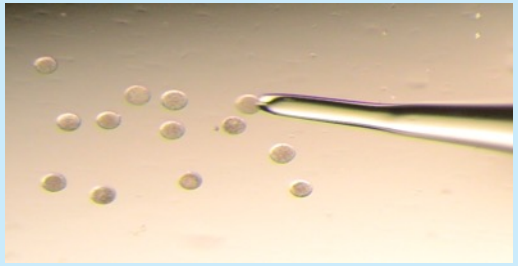
The Mzima cow project

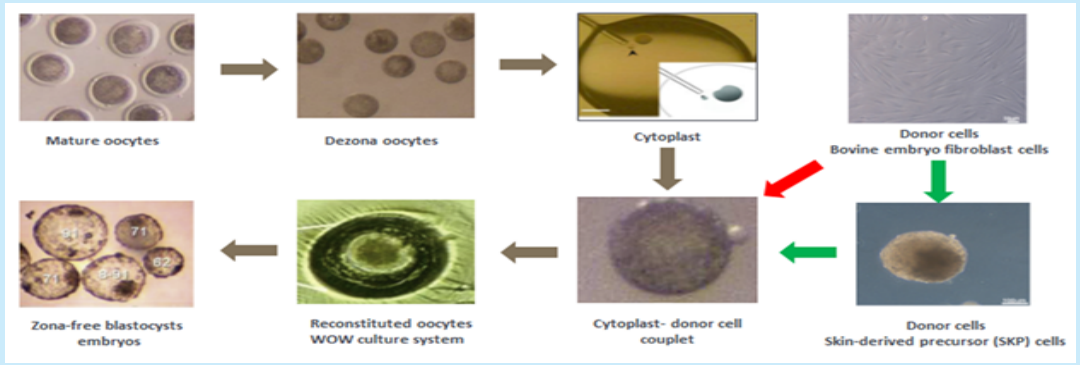
The Mzima Cow Project

A transgenic approach to trypanosome resistance

Currently, the Mzima cow methodology is based on CRSPR-Cas9 with Somatic Cell Nuclear Transfer SCNT using material derived from embryo skin cell lines.

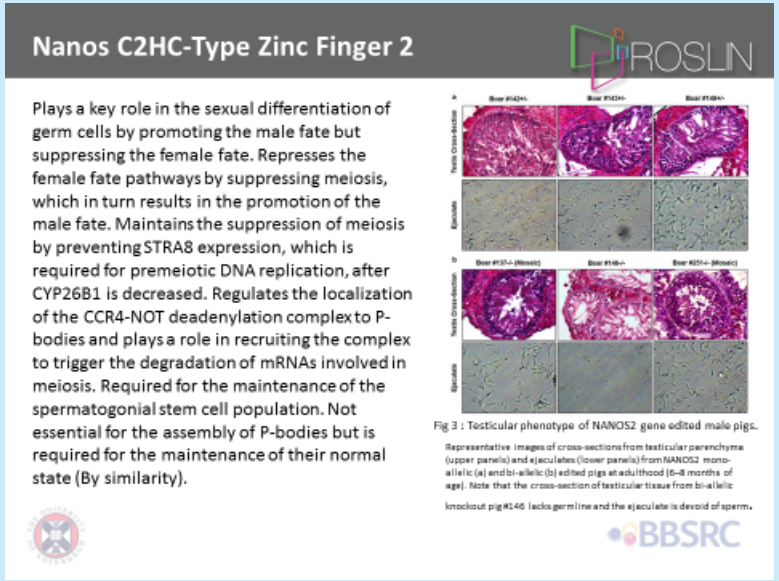
- Produce bulls transgenic for your target gene





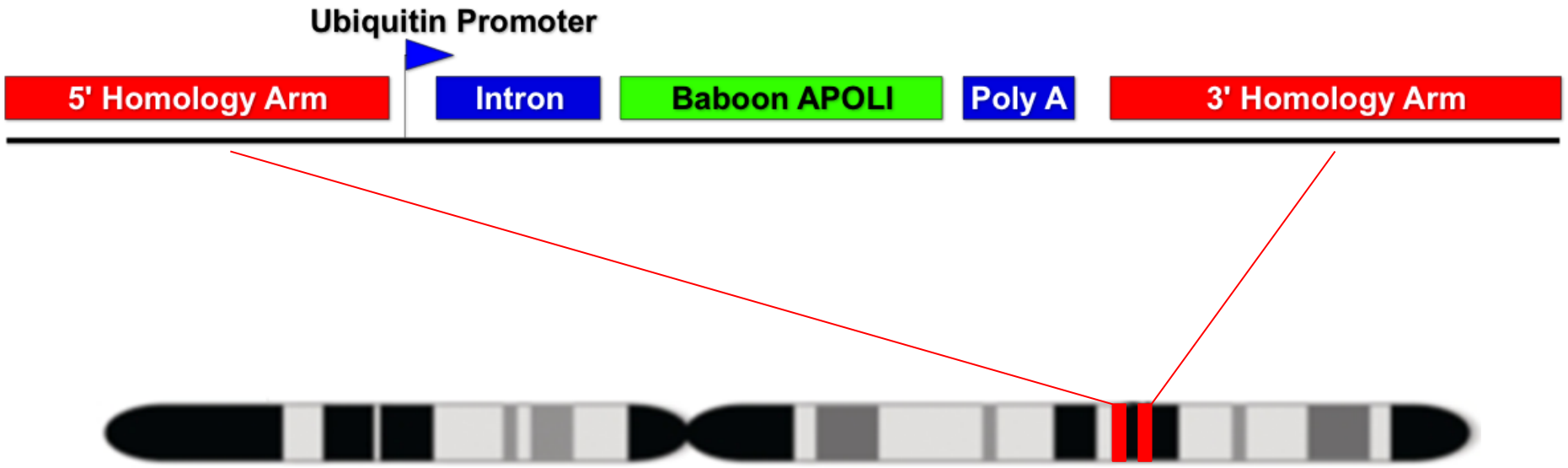
An interesting alternative approach is use of spermatogonial stem cell lines(SSC's)

- Produce *modified* males capable of hosting sperm carrying 'any' target gene
- Requires a host male with his own sperm production ablated



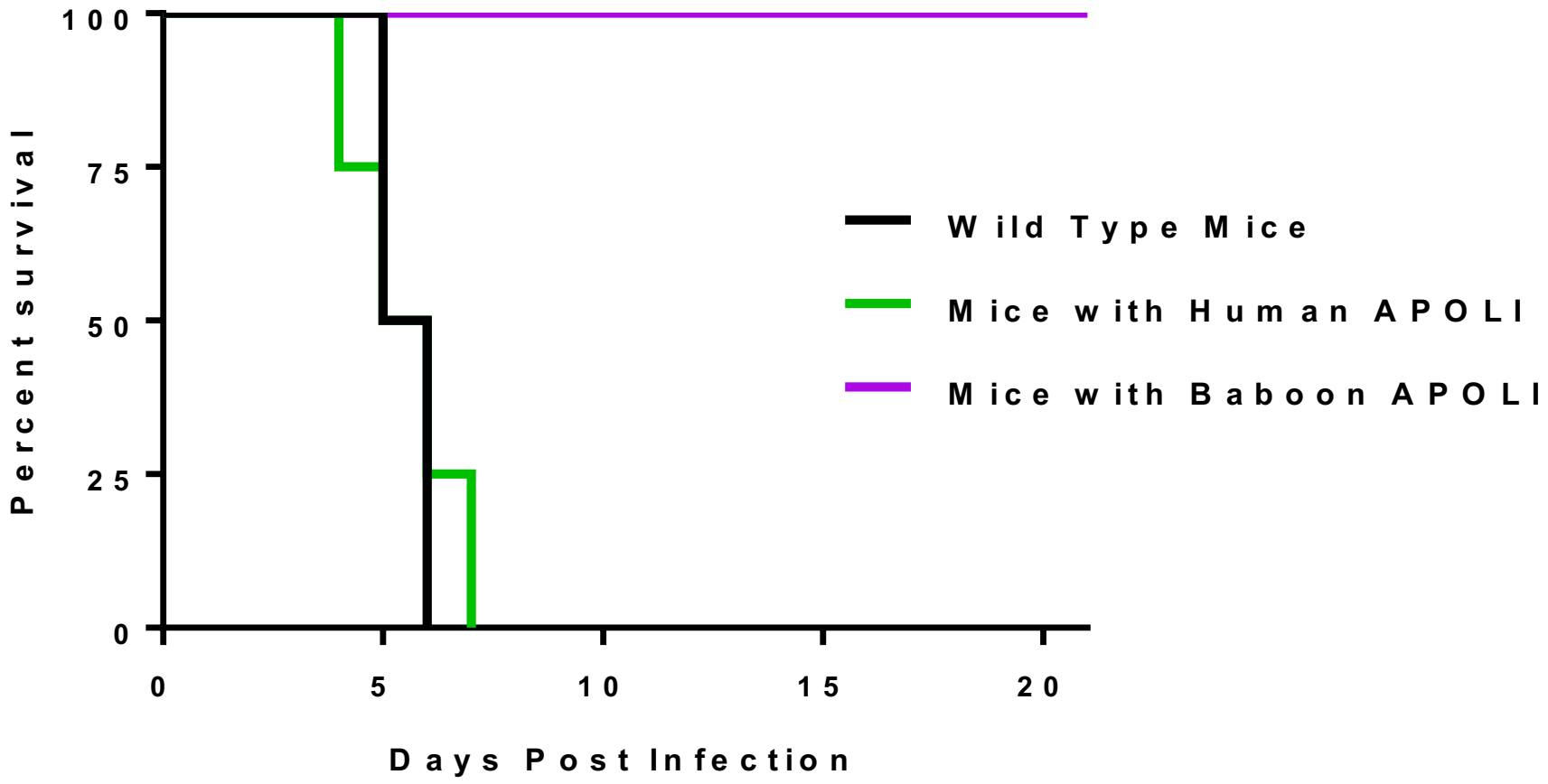
YY

Targeting the Transgene to the ROSA26 Locus Using CRISPR/CAS

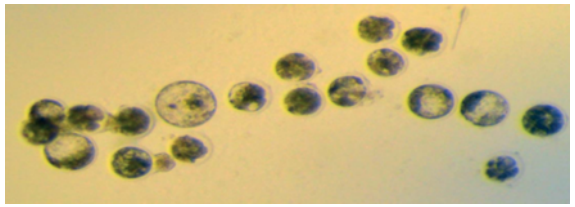


Are transfected mice protected from infection by African trypanosomes?

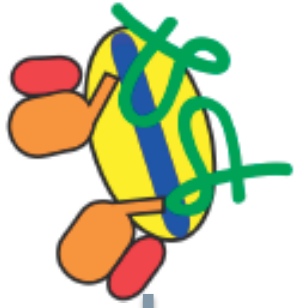
Mice expressing baboon APOLI are immune to infection by the human infective *T. b. rhodesiense*

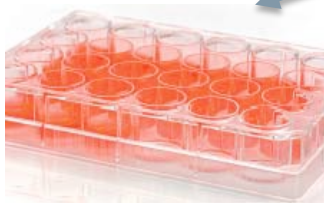


Mzima cow Experimental plan

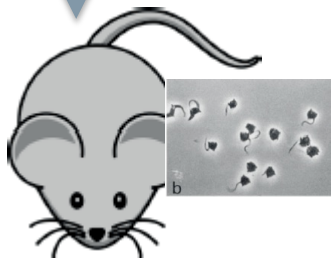


Design a synthetic TLF that could kill all African Trypanosomes





Validate the protective capacity of the genes in genetically modified mice



Bovine embryonic fibroblast cell culture system

Transfection of bovine targeting construct and blastocyst implantations

Assemble ideal bovine targeting construct

Establish bovine cloning system

Phenotyping and field studies

Stakeholder Engagement

The Mzima Cow

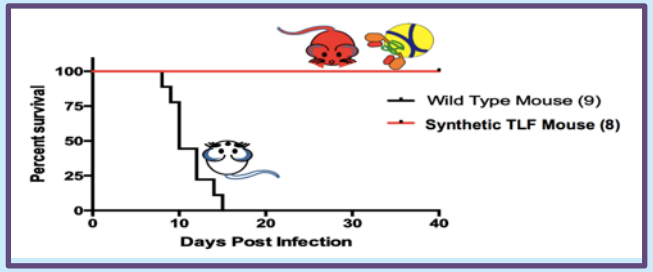
A transgenic approach to trypanosome resistance

Tumaini (*Hope*), the world's first cloned Boran at ILRI

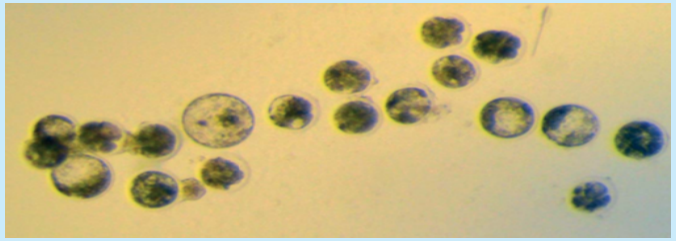
Tumaini resulted from SCNT at ILRI. He is healthy and fertile.

The APOL1 resistance gene has now been inserted into the same source cell line

CRISPR/Cas9 mediated insertion of TLF protects mice



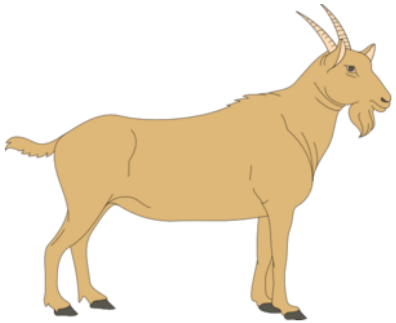
This construct is now in 'Tumaini's blastocysts' ready for implantation into recipient cows at ILRI



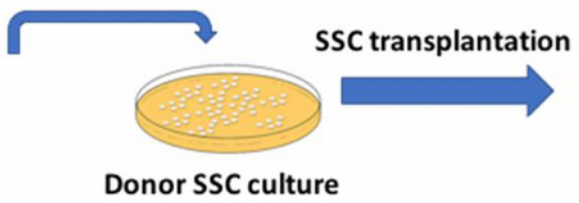
Jon Oatley

WASHINGTON STATE UNIVERSITY

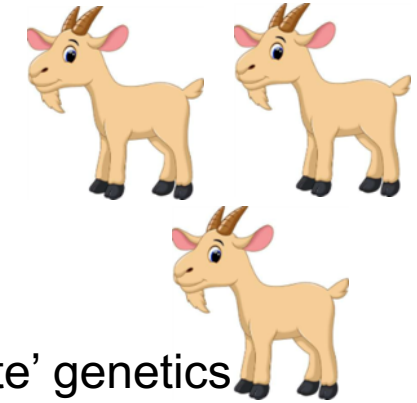
Surrogate sires to enable genetic improvement in small holder settings



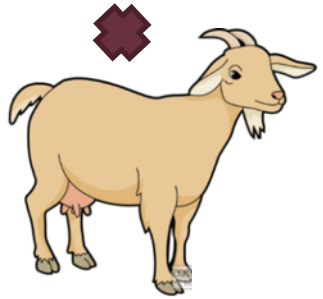
Donor ‘elite’ buck



Germline ablated recipient buck



Donor progeny with ‘elite’ genetics



Doe

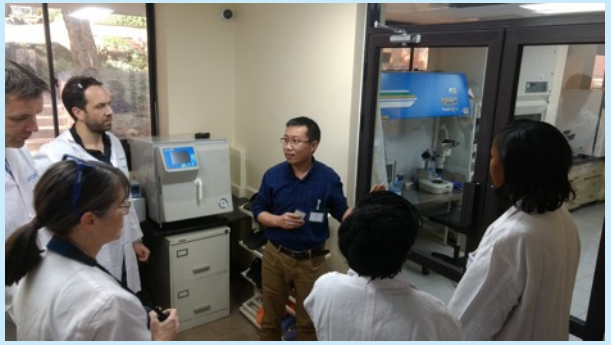
- Indigenous germ line ablated bucks carry the sperm of ’elite’ bucks.
- Instead of having one elite buck we would have thousands.
- This provides a transformative step change to disseminate ‘elite’ semen without changing the existing the infrastructure.

Schematic adapted from Oatley et al. 2017

Translating Genetic Research to Adoption & Social Value

Preparing the regulatory, safety & public awareness environment

Kenyan and regional regulators and stakeholders meet in ILRI with their global counterparts to discuss the Mzima Cow project



- Ground-breaking Science
- The Challenge of Adoption
- Defining the Testing Routemap

"Systems Change at the Speed of Trust":

Establishing trust is key to regulatory progress and the eventual adoption and understanding of genome edited livestock

ISAAA International Service for the Acquisition of Agri-biotech Applications

USDA

STI4D

The Mzima Cow

Regulatory and public awareness

Regulators, policy makers and stakeholders at the Mzima Cow Strategy & Theory of Change workshop 26-27th February at ILRI Nairobi, Kenya

In addition to organizations recognized for specific projects and outputs, we thank all donors which globally supported the work of ILRI and its partners through their contributions to the CGIAR system www.cgiar.org/our-funders

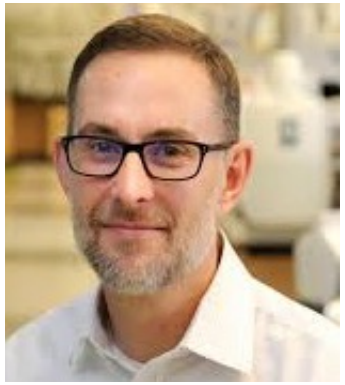
Thanks!

Mike McGrew

ROSLIN

Christian Tiambo

ILRI
INTERNATIONAL LIVESTOCK RESEARCH INSTITUTE



Jon Oatley

WASHINGTON STATE UNIVERSITY

Jayne Raper

HUNTER
The City University of New York

Centre for Tropical Livestock Genetics and Health

UKaid
from the British people

BILL & MELINDA GATES foundation

ILRI
INTERNATIONAL LIVESTOCK RESEARCH INSTITUTE

CGIAR