



Centre for  
Tropical Livestock  
Genetics and Health

# Protecting, preserving and restoring African poultry biodiversity

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4<sup>th</sup> International Workshop on Regulatory Approaches for Agricultural Applications of Animal Biotechnologies

12-16 September 2022 – São Paulo, Brazil



**TPGS**  
Tropical Poultry  
Genetic Solutions





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## Headlines

1. The state of Africa's poultry biodiversity and threats
2. Biobanking cPGC and chimera production at CTLGH/ILRI
3. Restoration of poultry biodiversity and dissemination of potential elite lines using the DDX4 KO and iCaspase-9 surrogate technologies
4. Up Scaling potential of poultry conservation for better livelihoods
5. Compliance from local biodiversity to global animal biotechnology





# The state of Africa's poultry biodiversity and threats



**South Africa:** Venda, Ovambo, Potchefstroom, kaalnekke, ...

**Algeria:** Kabyle, ...

**Ethiopia:** Tillili, Horro, Cefe, Jarso, Tepi, ...

**Nigeria:** Fulani, Yoruba, ...

**Kenya:** Molo Mushunu, ...

**Tanzania:** Ching'wekwe, Mbeya, Morogoro Medium, N'zenzegee, Singamagazi, pemba, Tanga, Unguja...

**Cameroon:** Dzaye, Dongwe, Tsabatha, Zarwa,...

**Burkina Faso:** konde, Fulani, Djelogodji, Dori, kolocissai, No Liguidi, joub-kole...

**Burundi:** sekaganda, Imangubo, Injogori,...

**Mali:** Wassa-Che, Semba-Che,...

**Botswana:** Tswana,...

**Chad:** Djided Baladi, Karmout,...

**Niger:** Goggori, Kolonto, Dourgou, Tchagara, Gousou-gousou, ...

**Egypt:** fayoumi, Biggadi/Ramadi, Dandarawi, Baladi Beheri, Sina, ...

Etc.

- Need to feed the African growing population
- Production increasingly based on a limited number of breeds.
- Within breeds Genetic diversity in decline.
- Roles of multipurpose breeds undervalued.
- Their genetic tolerance potential is important for the control of diseases and heat stress.





OPEN ACCESS

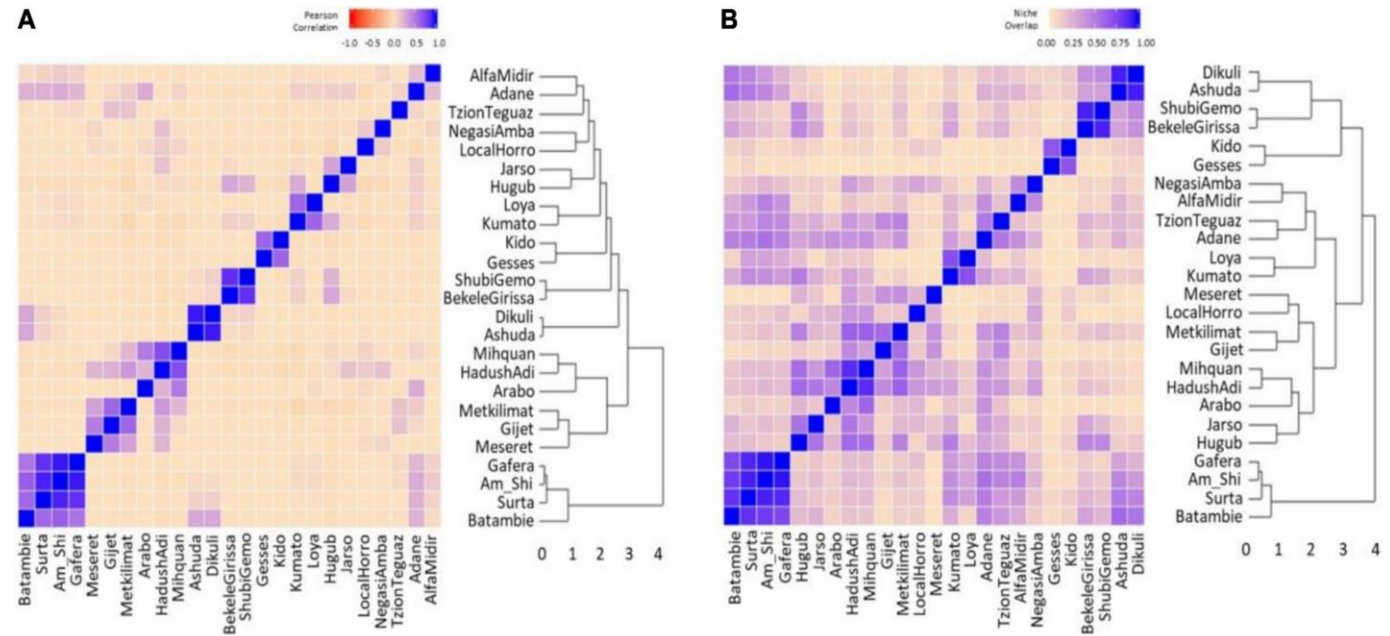
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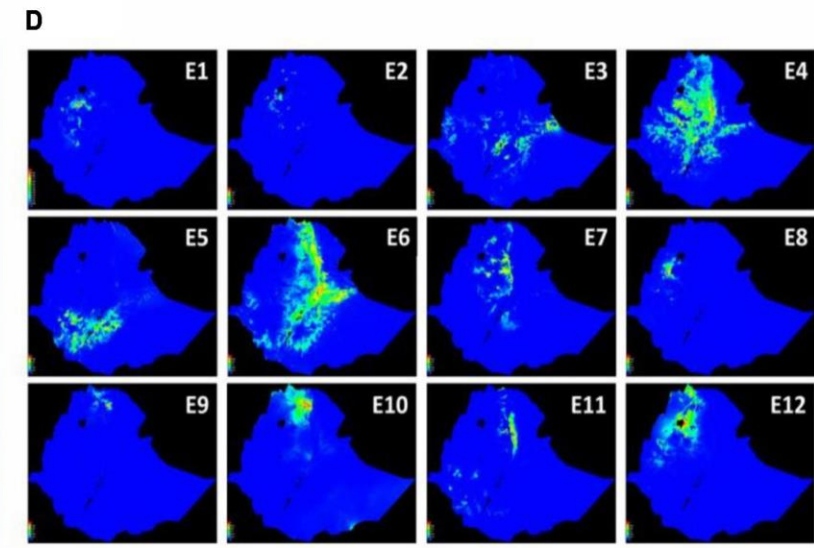
## Ecological niche modelling for delineating livestock ecotypes and exploring environmental genomic adaptation: The example of Ethiopian village chicken

Adriana Vallejo-Trujillo<sup>1\*</sup>, Adebabay Kebede<sup>2,3</sup>,  
Maria Lozano-Jaramillo<sup>4</sup>, Tadelle Dessie<sup>2</sup>, Jacqueline Smith<sup>5</sup>,  
Olivier Hanotte<sup>1,2\*</sup> and Almas A. Gheyas<sup>5\*</sup>



**Figure.** Delineation of ecotypes based on niche similarity among 25 Ethiopian chicken populations examined. **(A)** Dendrogram and heatmap of pairwise Pearson correlation ( $I$ ) among 25 populations; **(B)** dendrogram and heatmap of pairwise niche overlap statistic ( $r$ ) among 25 populations; **(C)** 12 delineated ecotypes and their population composition; and **(D)** suitability maps of 12 delineated ecotypes.

Ecotype	Populations
E1	AmeshaShinkuri, Gafera, Surta, Batambie
E2	Ashuda, Dikuli
E3	BekeleGirissa, ShubiGemo
E4	Adane, LocalHorro
E5	Loya, Kumato
E6	Mihquan, HadushAdi, Hugub, Jarso
E7	NegasiAmba, AlfaMidir
E8	Gesses, Kido
E9	Meseret, Gijet
E10	Metkilimat
E11	Arabo
E12	TzionTeguaz





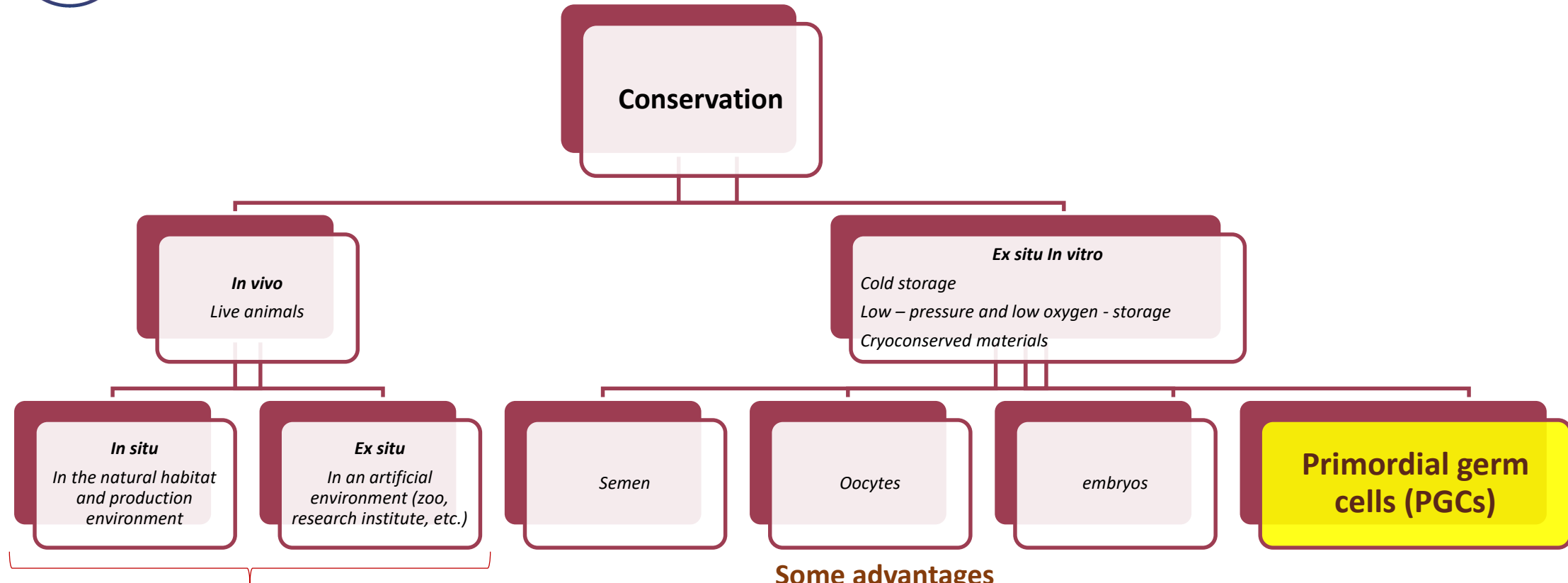
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# Major threats to poultry genetic diversity in Africa

- The rapid spread of homogenous large-scale intensive production;
- Inappropriate management, conservation and development policies & strategies;
- Disease outbreaks,
- Negative selection and inappropriate breeding programmes;
- Various types of disasters and emergencies.



# Multiple options for the conservation of poultry biodiversity



Conservation through improved use

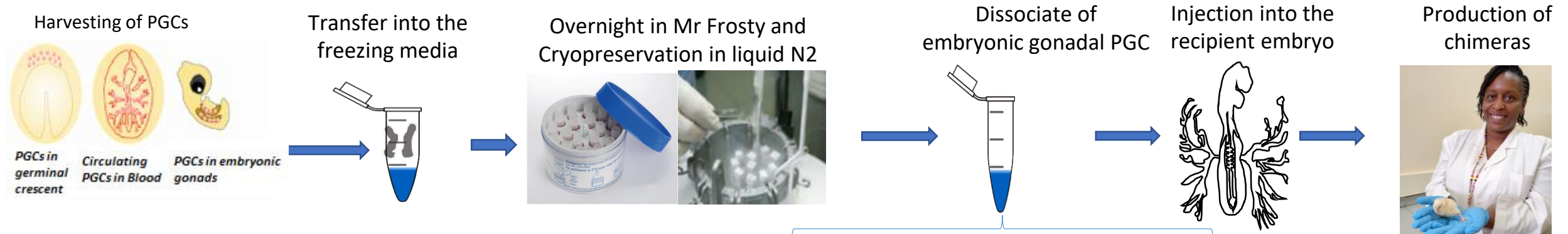
## Some advantages

- ✓ Large diversity can be preserved in a small space.
- ✓ Maintained in an environment free from pathogens.
- ✓ Protected against the nature's hazards
- ✓ Large number of chicken can be revived when needed.

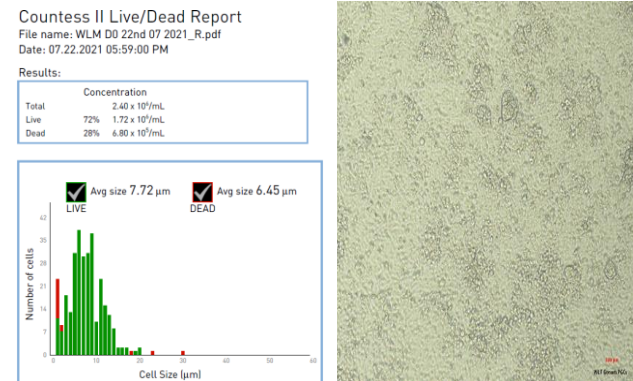


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# Biobanking PGC and chimeras production at CTLGH / ILRI



PGC propagation and characterization



Chimera hen showing some phenotypic characteristics of the local donor







# Biobanking from gonads PGC and chimeras' production at CTLGH / ILRI

From blood and blastoderm

From gonads (work in progress)

Country	Ecotype	Number cell lines
KENYA	White leghorn	43
	Karen (Nairobi)	7
	Narok	14
	Bomet	42
	Siaya	23
	Migori	20
	Homabay	8
	Kakamega	13
ETHIOPIA	Arobe	59
	Horro	29
	Hawassa	32
TANZANIA	Shinyanga	84
	Mwanza	11
	Mbeya	43
	Morogoro	69

No.	Kenyan Ecotypes	Male lines	Female lines
1	Laikipia (LP)	44	43
2	Bungoma (BG)	9	18
3	Kilifi (KF)	4	1
4	Kakamega (KK)	14	13
5	Bomet (BM)	1	1
6	Homabay (HB)	21	19
7	Siaya (SY)	13	5
8	Kwale (KW)	0	1
	<b>Total</b>	<b>106</b>	<b>101</b>

No.	KALRO Improved lines	Male	Female
1	KC1	61	63
2	KC2	75	41
	<b>Total</b>	<b>136</b>	<b>104</b>

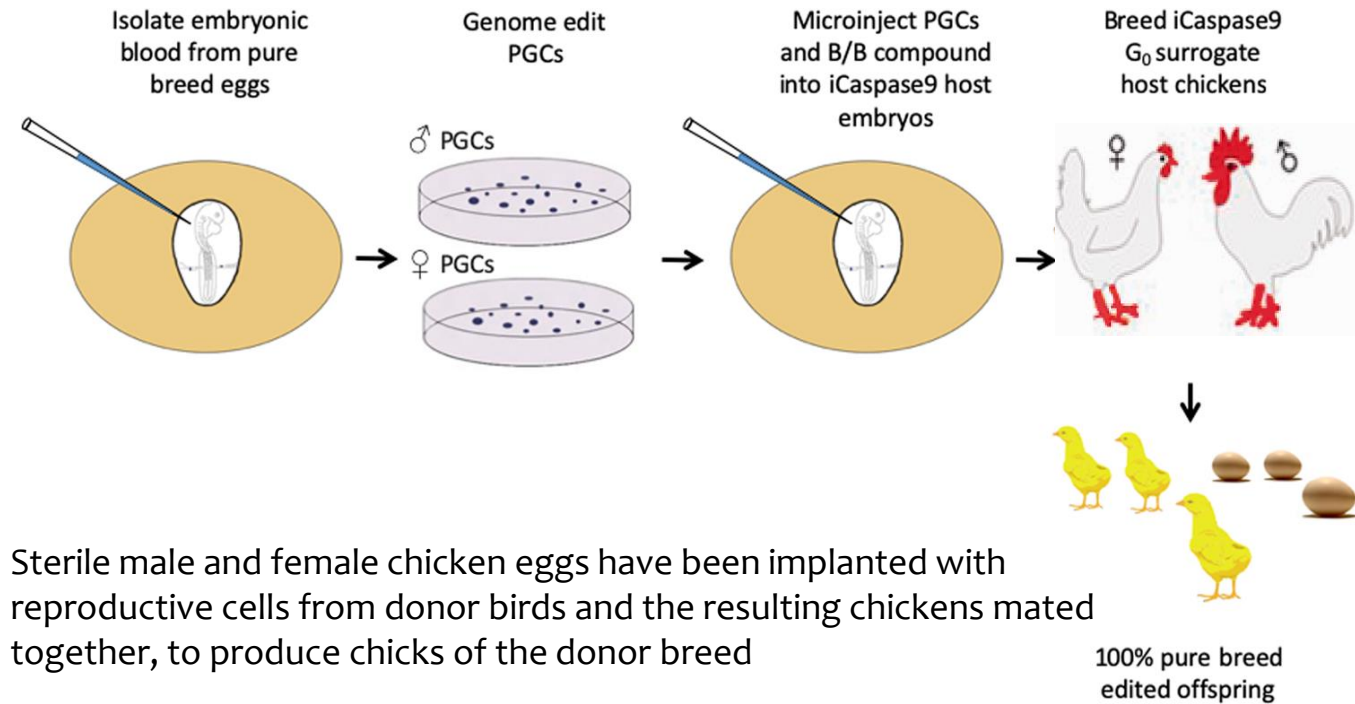
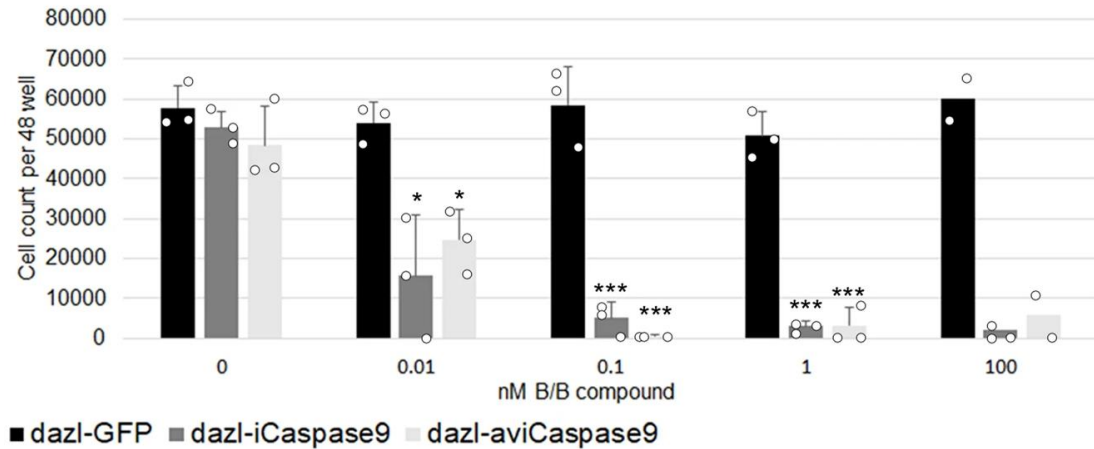
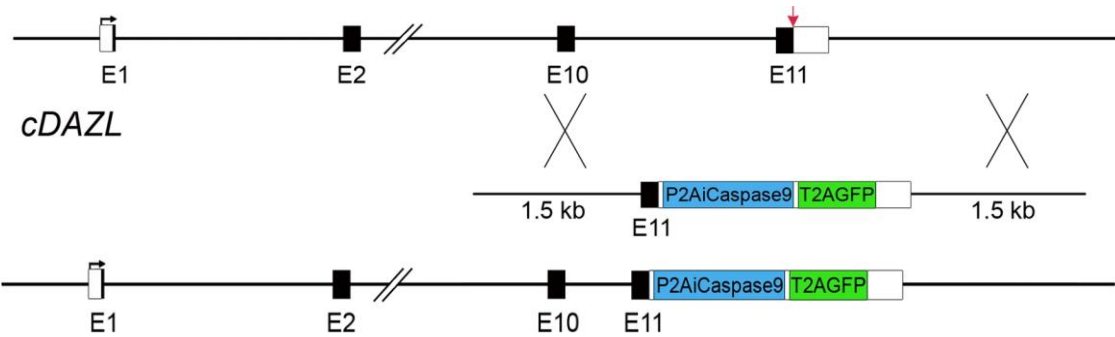






# Restoration of poultry biodiversity and dissemination of potential elite lines using the DDX4 KO and iCaspase-9 surrogate technologies

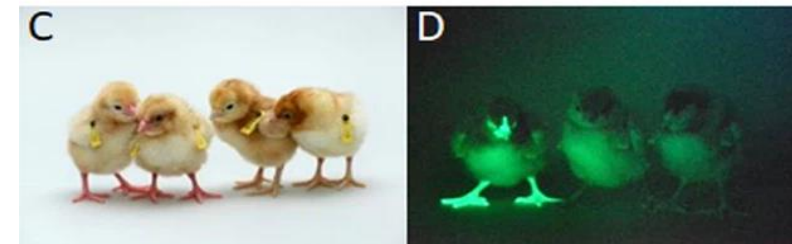
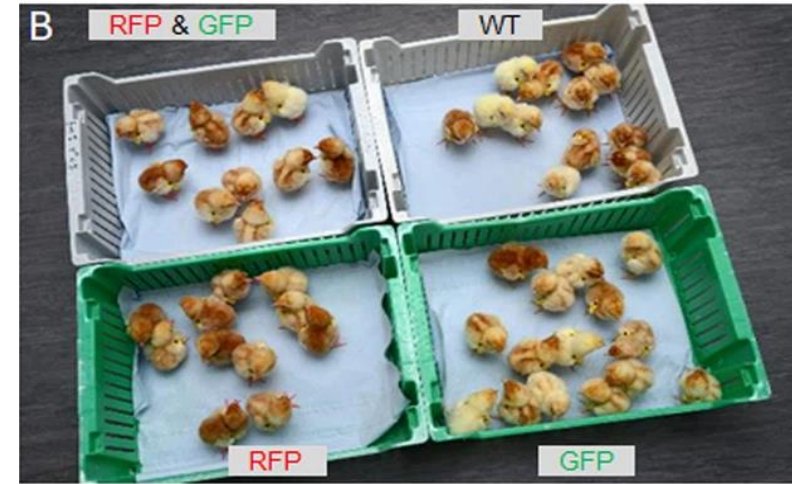
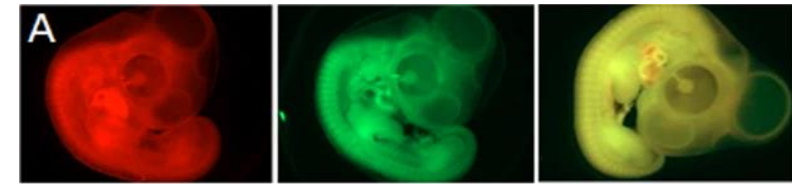
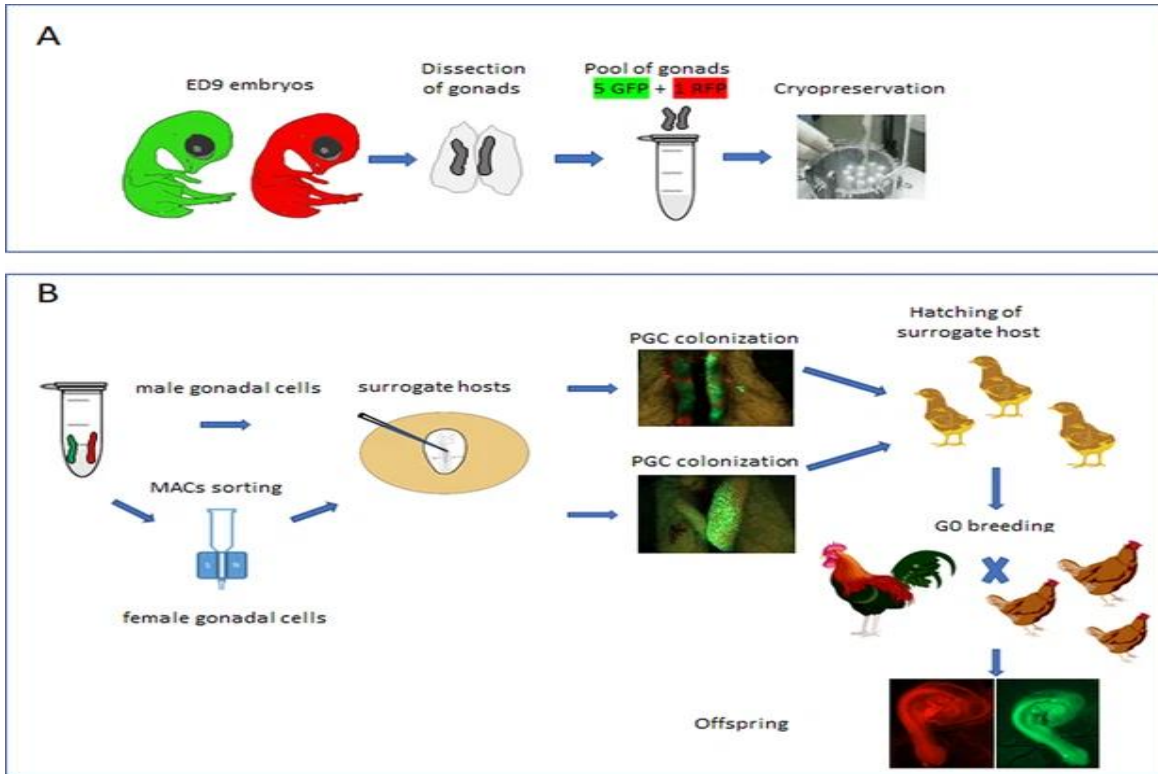
## Production of sterile surrogate host



Sterile male and female chicken eggs have been implanted with reproductive cells from donor birds and the resulting chickens mated together, to produce chicks of the donor breed



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(A) Embryonic day (ED) 9 gonads are isolated from embryos, pooled by sex, and cryopreserved in liquid N<sub>2</sub>. (B) The frozen gonads are thawed, dissociated, and injected into sterile surrogate host embryos. The surrogate host embryos are incubated and hatched and bred to hatch donor gonadal offspring.

(A) Embryonic day (ED) 5 embryos from Sire Dam Surrogate (SDS) mating displaying representative red, green, and 'yellow' fluorescence. (B) Hatched chicks from SDS mating grouped according to fluorescence. (C) RFP fluorescent chicks were apparent (chicks on left) under visible light. (D) GFP fluorescent chick (left) visualised under GFP illumination.





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# Restoration of poultry biodiversity and dissemination of potential elite lines using the surrogate technology




27-Jul-22  
\*\*\*

**Dr Evans Ilatsia**  
Deputy Director General, Kenya Agricultural and Livestock Research Organization  
Institute Director / Dairy Research Institute,  
P.O. Box 25 - 20117, Tel: + 020 2390930, NAIVASHA  
E-mail : [Kairo.Poultry@kairo.org](mailto:Kairo.Poultry@kairo.org)

Dear Sir,

**REQUEST TO HOST THE BACKUP POULTRY FLOCKS FOR THE COLLABORATIVE WORK ON CHICKEN CONSERVATION AT KALRO NAIVASHA**

I have the honor most respectful to request the establishment at KALRO-DRI and within the poultry unit, of a backup flock of white leghorn and other poultry breeds we are currently keeping at ILRI-Nairobi farm.

This is to ensure good continuity and efficiency of the ongoing work, while reinforcing collaboration between our two institutions for the conservation, recovery, and dissemination of the African chicken genetic resources.

While the main laboratory work will still be conducted from CTLGH-ILRI lab, the establishment of the flock at KALRO will imply frequent movements of eggs and chicks between ILRI-Nairobi and KALRO-Naivasha. The logistic for that purpose will be catered for by the laboratory activities.

Best Regards,


*Ababu lociny*  
*This is approved - please ensure with Dr. Tiamba on the logistics of doing this*  
*27/07/22*  
*KAG-L*

**K. Tiamba, Christian (ILRI)**  
Scientist  
Co-Lead, Reproductive Technologies  
CTLGH / Livestock Genetics -ILRI  
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Patron: Professor Peter C Doherty AC, FRCGS, FRCR  
Animal scientist, Nobel Prize Laureate for Physiology or Medicine-1996

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ILRI has offices in East Africa • South Asia • Southwest and East Asia • Southern Africa • West Africa



**NATIONAL BIOSAFETY AUTHORITY**  
NBISS/001/BUILDING 23/1/1/0001  
UPPER KABETE, OFF WAIYAKO WAY  
P.O. Box 28251 - 00100, Nairobi. T:Tel: +254 20 267 8667  
Email: [info@biosafetykenya.go.ke](mailto:info@biosafetykenya.go.ke) Website: [www.biosafetykenya.go.ke](http://www.biosafetykenya.go.ke)

When replying please quote:  
Our ref: NBA/GMO/C09/98/(19)      17<sup>th</sup> May 2022

**The Director General,**  
International Livestock Research Institute (ILRI),  
P. O. Box 30709-00100  
**NAIROBI, KENYA**

Attn: Dr. Vish Nene, Chair, ILRI IBC

**APPROVAL DOCUMENT: APPLICATION TO INTRODUCE GENE EDITED SURROGATE HOST CHICKENS FOR TRIALS ON BIOBANKING AND RECOVERY OF INDIGENOUS CHICKEN BREEDS**

Reference is made to your application seeking authorization to demonstrate the use of surrogate technology (DDX4 and iCaspase9 chicken) to revive cryopreserved Primordial Germ Cells (PGCs) from elite African chicken genetic resources to enhance resilience, productivity and support future response to new breeding requirements.

The National Biosafety Authority has reviewed your application in line with the provisions of the Biosafety Act, 2009 and the Biosafety (Contained Use) Regulations of 2011. The Authority approved your application to conduct the contained use activity and spelt out conditions that you are required to observe.

Attached herein, please find approval document reference number **NBA/GMO/C09/18/44**. Following this approval, it is expected that the project will commence within twelve (12) months from this date, failure to which the project will be deemed as terminated.

We look forward to a harmonious working relationship with you as the project commences.

*[Signature]*  
**DR. ROY B. MUGIIRA, PHD, MRSB**  
AG. CHIEF EXECUTIVE OFFICER  
NATIONAL BIOSAFETY AUTHORITY

Copy to: **Dr. Obadiah N. Njagi**  
Director, Directorate of Veterinary Services (DVS)  
Veterinary Research Laboratories, Kabete  
Private Bag 00625  
**KANGEMI, NAIROBI.**

National Biosafety Authority is ISO 9001:2015 Certified



**Target :** To return to the local providing communities their conserved elite breeds to improve livelihoods

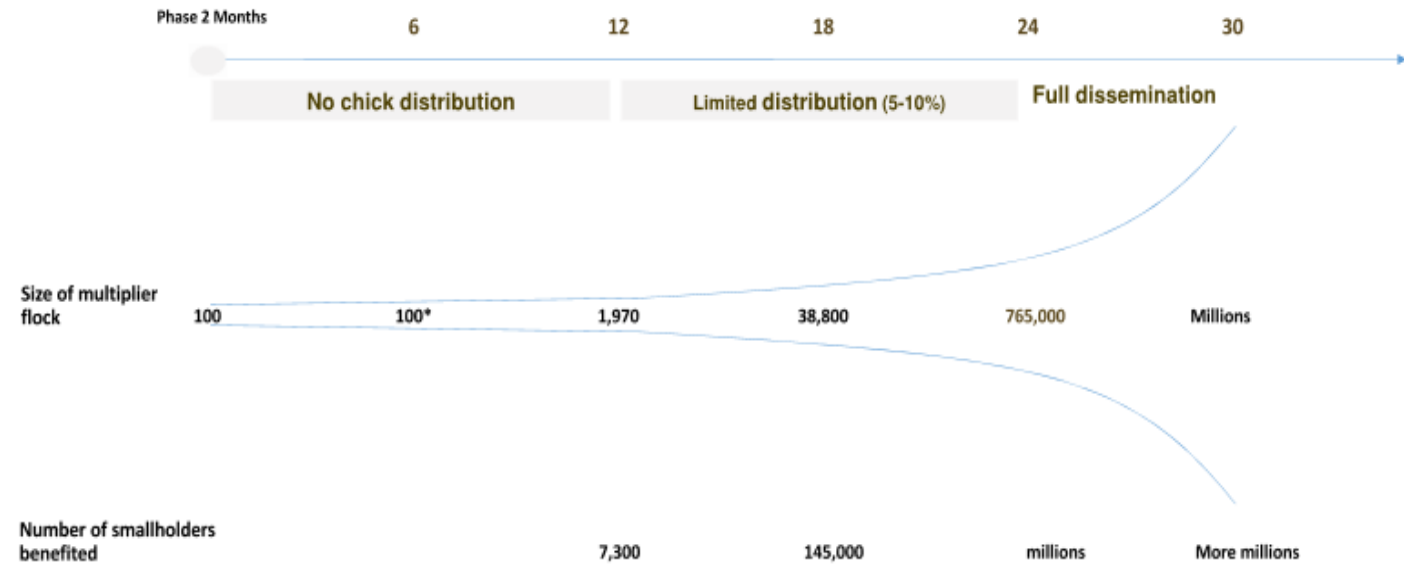
Following NBA inspection, Authorizations to import and to host the Surrogate chicken and the ILRI's backup flock at KALRO



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# Up Scaling potential of poultry conservation for better livelihoods: the TPGS model



## NB:

- Chicks recovered from PGCs are 100% of donor's genotypes
- The biological machinery of exotic surrogate could enable rapid scaling up in distribution of elite adapted local lines.
- The TPGS solution model support emergence of local poultry industry





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# Biobanked PGCs to support to Poultry research and development

## PGCs in Biorepositories and Gene banks

Low throughput



Medium throughput



High throughput



Various sequencing Platforms/services



- ❖ DNA seq. : functional annotations
- ❖ RNA seq. : Gene expression data
- ❖ Proteins seq. : protein structures
- ❖ Metabolites: small molecule structures



- ❑ Physical redundancy (through a RAID array)
- ❑ Logical redundancy (through the GlusterFS storage supervision software that implements logical replicas)
- ❑ INSDC databases (ENA, GenBank, DDBJ) for most sequences
- ❑ GISAID for SARS-CoV-2 sequences

Security



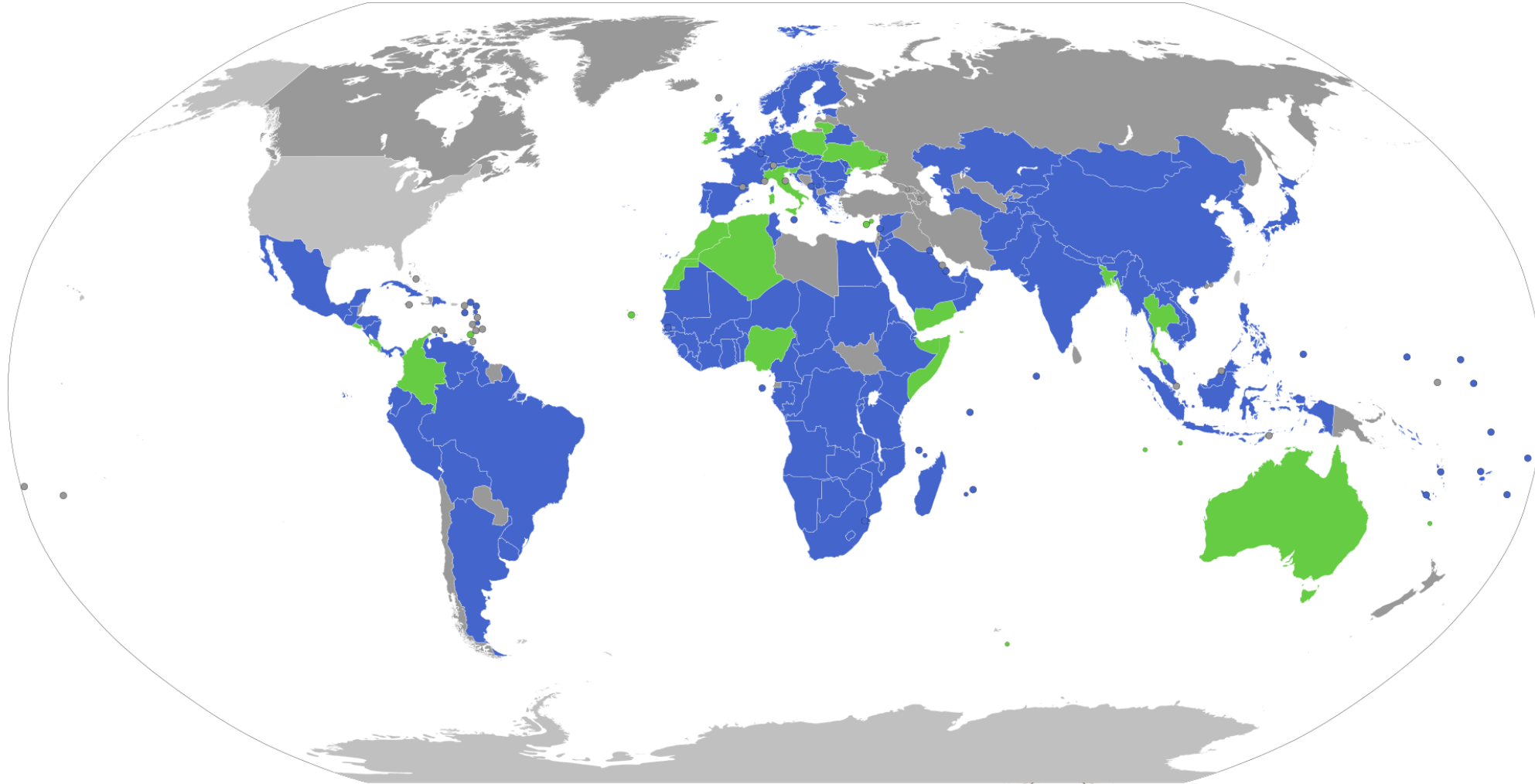
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Uploaded on various platforms



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# Regulation on ABS of the Nagoya Protocol for Protection of African poultry biodiversity



-  Parties
-  Only signed, but not ratified
-  Non signatory, but CBD party
-  Non signatory, non-CBD party







# ACCESS PERMIT PROCEDURE: e.g. from KENYA

## PRE-ACCESS PROCEDURE



Negotiate terms and conditions of the MOU or CRA

[www.nema.go.ke](http://www.nema.go.ke)  
[www.oris.nacosti.go.ke](http://www.oris.nacosti.go.ke) or  
[www.nacosti.go.ke](http://www.nacosti.go.ke)

Lead agency, individual or community in a specified locality

- Letter of Affiliation
- Partnership MOU/CRA
- Project proposal abstract.

Apply for researcher's pass (Departement of Immigration Services: Kenya Citizenship and Immigration Act, 2011).

Establish contact with appropriate GR provider through the public administration to commence the process of acquiring PIC&MAT

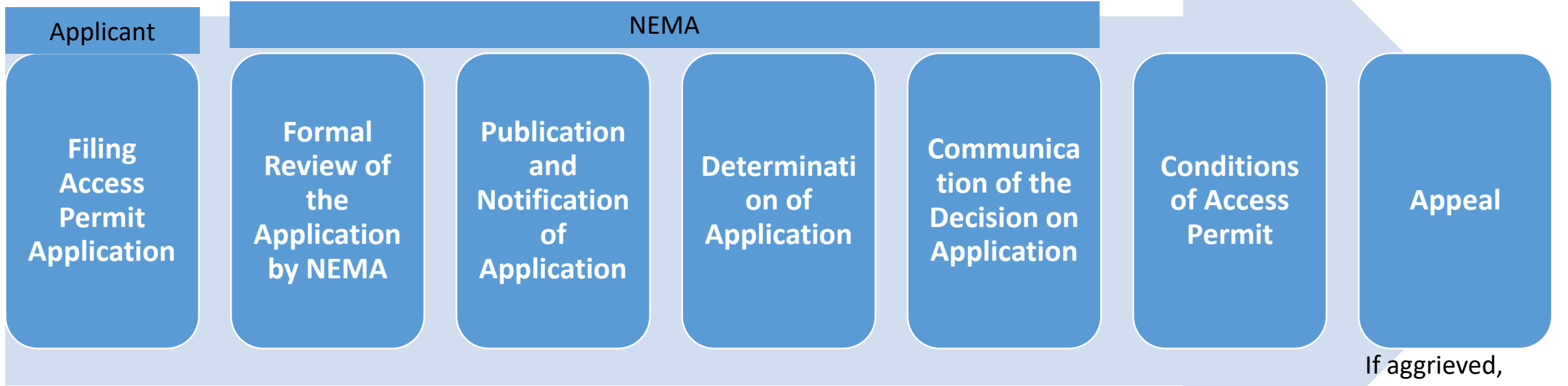






# ACCESS PERMIT PROCEDURE: e.g. from KENYA

## PROCESS FOR ACQUIRING ACCESS PERMIT



Fill the form:  
[www.nema.go.ke](http://www.nema.go.ke)  
 Attach required document, including the PIC&MAT

Publishes the application in the Kenya Gazette and in at least one nationwide newspaper

Invites ABS Technical Committee (TC) to evaluate the application taking into consideration the comments from the public and other stakeholders

Decision to grant/reject the application communicated within 60 days from the date of receipt of the application.

Permit is not transferable. Valid for one year renewable for a similar period.

If aggrieved, appeal to the National Environment Tribunal as provided for in section 11 (3) of the ABS Regulations, 2006.



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# ACCESS PERMIT PROCEDURE: e.g. from KENYA

## POST ACCESS

### Execution of Material Transfer Agreement

1. Engage the relevant lead and initiate negotiation of terms of transfer of the GRs and/or aTK
2. Negotiate and complete MTA customize it appropriately to suit the GR
3. Ensure the agreement is signed and dated by legally authorized persons in both contracting parties
4. Submit a copy of the executed MTA to NEMA for further information.

### Application for Export Permit

- \* Phyto-sanitary Certificate
- \* Animal Health Certificate

- Apply for and obtain an export permit from the relevant lead agency
- Attach the animal health clearance certificate from DVS



# ACCESS PERMIT PROCEDURE

## TRANS-BOUNDARY MEASURES

- If the same genetic resources or aTK are found *in situ* within many countries (eg: Kenya, Uganda, Tanzania, Ethiopia, etc.) the Government will endeavour to cooperate with them and involve IPLC concerned, where applicable, with a view to implementing PIC, MAT and other ABS Agreements and the objectives of the Nagoya Protocol.



# ACCESS PERMIT PROCEDURE

## INTERNATIONALLY RECOGNIZED CERTIFICATE OF COMPLIANCE (IRCC)

- The access permit issued in accordance with ABS Regulations is submitted to the CBD ABS- CH to constitute an IRCC.
- The Certificate of Compliance serves as evidence that the genetic resource which it covers has been accessed in accordance with PIC&MAT have been established, as required by the Kenyan ABS regulatory requirements.





# Implications of non-compliance: **Biopiracy**

## Non-compliance (*Regulation EU (No) 511/2014*) meaning failure to:

- Comply with the ABS laws of the provider country
- Comply with your benefit-sharing agreement as in PIC&MAT
- Comply with the AU Regulation
- Fulfill your due diligence obligations, including SKT information failing to declare due diligence;
- Stop utilization if information becomes available that you need ABS documentation for your research material and you don't have it; or
- Support a user check by the Nature Protection Agencies .



# Implications of non-compliance: **Biopiracy**

## Action against non-compliant researchers and institutions:

### 1. Remedial orders

- ✓ Prohibits further utilization and transfer of the material;
- ✓ Prohibits the publication or sharing of any results of utilization

### 2. Confiscation

- ✓ The BfN is authorized to confiscate material or research results

### 3. Fines

- ✓ Regulation (EU) No 511/2014: **€50,000**

### 4. Reputation loss

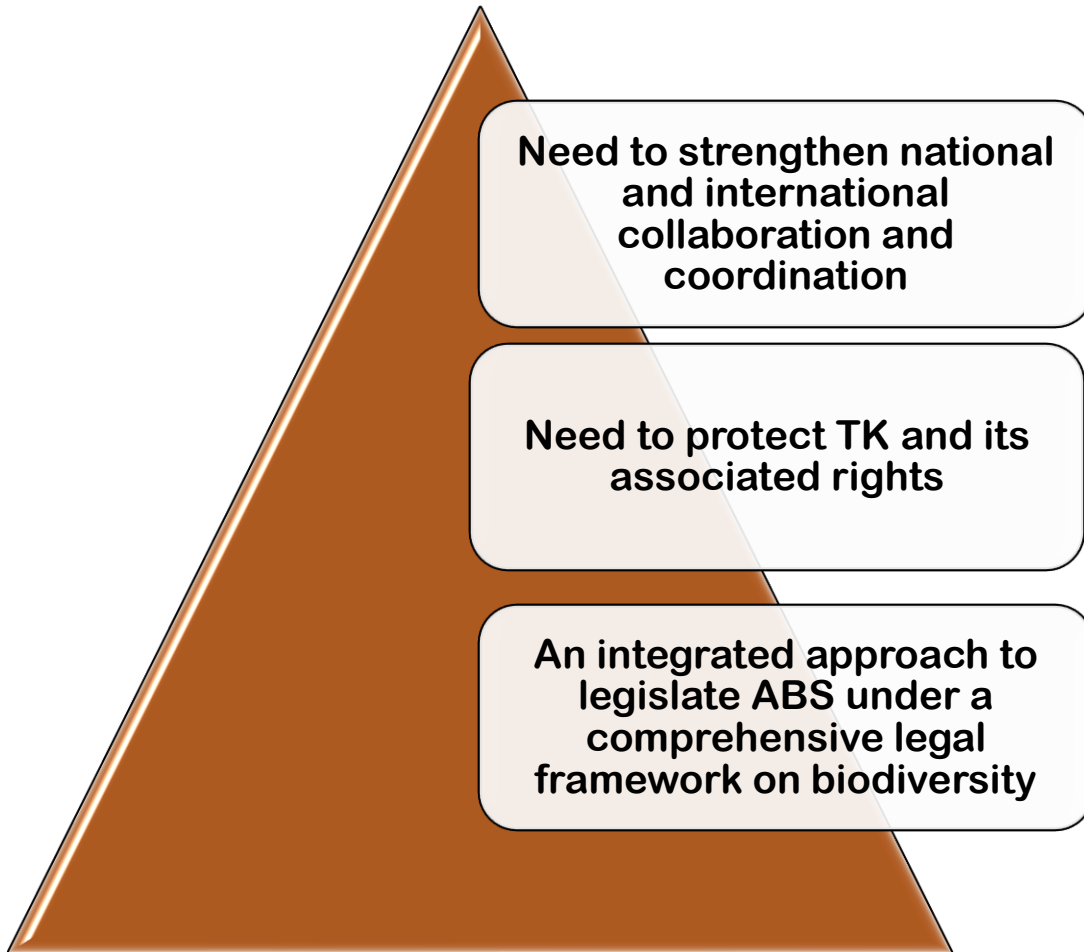
### 5. Blacklisting

### 6. Publication retracted

### 7. Refusal to deposit



# Lessons Learnt & Solutions



- CTLGH/LiveGene - ILRI APPROACH





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# Thank you

[www.ctlgh.org](http://www.ctlgh.org)

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