



Toll-like Receptors in Farm Animals – Evolutionary Lineages and Application in Disease Resistance

Indian Council of Agricultural Research
National Agricultural Innovation Project



Consortium leader



Madras Veterinary College, Tamil Nadu Veterinary and Animal Sciences University, Chennai

Consortium partners



**Indian Veterinary Research Institute
Mukteswar**



**National Bureau of Animal Genetic Resources
Karnal**

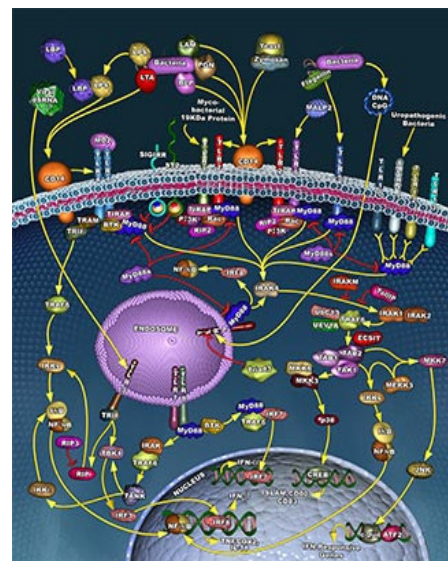
2008 - 2012

National Agricultural Innovation Project (NAIP)

NAIP of Indian Council of Agricultural Research (ICAR) accords high priority to generation and transfer of innovative agricultural technologies. The overall objective of the NAIP is to facilitate an accelerated and sustainable transformation of the Indian agriculture, so that it can support poverty alleviation and income generation through collaborative development and application of agricultural innovations by the public organizations in partnership with farmers, the private sector and other stakeholders. The specific objective of the NAIP under the **Component 4** is to build capacity to undertake basic and strategic research in frontier areas of agricultural sciences. The present project is funded by the NAIP to study the Toll-like receptors in farm animals-Evolutionary lineages and application in disease resistance

Toll-like receptors

Whenever a pathogen invades a host, the host mounts an innate immune response primarily to keep the pathogen under check before the specific immunity consisting of antibodies and immune cells fights the pathogen more precisely. One of the components of this innate immune system is the toll-like receptors (TLRs). They recognize conserved components among various classes of pathogens / chemicals / molecules. TLRs are capable of identifying different classes of pathogen-specific structures. For example, TLR 4 recognizes lipopolysaccharide moieties in Gram negative bacteria. This recognition in turn stimulates a sequence of signaling mechanisms resulting in the production of various protein effector molecules (cytokines). These molecules serve as a link between innate and specific immune mechanisms. Thus TLRs (immune cell-origin) or their specific ligands usually of pathogen-origin, can be exploited to modulate the immune response culminating in a more beneficial outcome to the host.



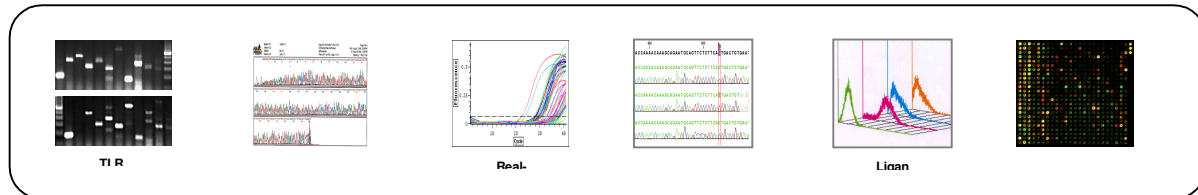
Objectives

- Sequence characterization of toll-like receptor genes in buffalo, goat, mithun and yak
- Ligand screening and expression profiling of TLR genes in different breeds of buffaloes and goats
- Detection of polymorphism in TLR genes of buffalo and goat and their association with expression levels
- Correlation of TLR mRNA expression levels with a bacterial, viral and a parasitic disease resistance models

Brief project description

- TLR genes of buffalo, goat, mithun and yak would be amplified and sequenced to determine how these genes have evolved among different animal species.
- The basal expression levels of TLR mRNAs of three distinct breeds of buffaloes and goats would be assessed using real time PCR to determine whether these species / breeds are susceptible / resistant to pathogens by virtue of their reduced or enhanced levels of TLR mRNAs
- If enhanced levels of TLR mRNAs are found in some breeds, then the reason for such enhancement would be assessed by screening for any favourable mutations (SNPs) in their coding genes or promoter regions.

- The ligands for different TLRs and their induced cytokine profiles would be assessed in buffalo and goat immune cells to identify suitable ligands for therapeutic interventions
- The role of TLR genes in conferring species-specific disease resistance would be further confirmed using disease resistant models wherein buffalo and goats are either susceptible or resistant to bacterial, viral and parasitic diseases.



Novel approaches to be followed

- ❖ To determine how TLR genes evolved in various animal species
- ❖ To elucidate the role of TLRs using specific disease resistance models
- ❖ To find out why some classes of pathogens multiply more in certain tissues or organs and not in others
- ❖ To relate mutations in TLR genes that can be associated with increased basal TLR mRNA expression
- ❖ To design a DNA chip for assessment of the expression levels of buffalo and goat TLRs and cytokine genes
- ❖ Application of TLRs or their ligands as targets for immuno-modulation and increasing the potency of existing vaccines

The total cost of the project: Rs. 308.483 lakhs

Outputs

- ❖ About 40 TLR gene sequences of buffalo, goat, mithun and yak
- ❖ About 480 basal expression levels of various TLR mRNAs in three breeds of buffalo and goat
- ❖ Four TLR ligands capable of stimulating buffalo and goat mononuclear cells and their induced 40 cytokine profiles
- ❖ SNPs in 4 TLR genes in 3 breeds of buffalo and goat and their association with basal TLR mRNA expression levels
- ❖ Role of TLRs in a bacterial, viral and parasitic disease resistance models

Expected impact of the project

- ❖ **TLR ligands as alternate targets**- Various TLR ligands could serve as alternate targets for increasing the potency of existing vaccines and for therapeutic intervention
- ❖ **Breeding for disease resistance** – The identified SNPs responsible for increased TLR gene expression can be exploited in marker based selection programmes
- ❖ **Disease Resistance Chip** - Using this microarray based method to quantify TLR and cytokine mRNA, other breeds of buffalo and goat can be screened for disease resistance potential.

TANUVAS

Sequence characterization of TLR genes in goat
Ligand screening and expression profiling of TLR genes in buffaloes and goats
Association of polymorphism in TLR genes with TLR mRNA expression levels in buffaloes and goats

TOLL-LIKE RECEPTORS IN FARM ANIMALS – EVOLUTIONARY LINEAGES AND APPLICATION IN DISEASE RESISTANCE

IVRI

Sequence characterization of TLR genes in mithun and yak
Correlation of TLR mRNA expression levels in

NBAGR

Sequence characterization of TLR genes in buffalo
Detection of polymorphism in TLR genes of buffalo, goat and their association with TLR mRNA expression levels

Consortium Principal Investigator (CPI) and Consortium Co-Principal Investigators (CCPIs)

Name of the member	University / Institute	E mail	Phone
Dr. G. Dhinakar Raj (Consortium Principal Investigator)	Professor, Dept. of Animal Biotechnology, Madras Veterinary College, Chennai – 600 007	dhinakarraaj@yahoo.com	O: 044-25381506 Ext- 257 M: 09381036277
Dr. R.K.Singh (Consortium Co-Principal Investigator)	Head, Division of Virology and Station in-charge IVRI, Mukteswar, Nainital District -263 138	rks_virology@yahoo.com	O: 05942-286346 M: 09927438389
Dr. R.S. Kataria (Consortium Co-Principal Investigator)	Senior Scientist, DNA fingerprinting Unit, NBAGR. Karnal 132 001	katariaranji@yahoo.co.in	O: 0184-2267918 M: 09416344825

CONSORTIUM ADVISORY COMMITTEE

Chairman - Dr. H. K. Pradhan, WHO consultant, Bharathiya Kala Kendra, 1, Copernikus Marg,
New Delhi -110 001

Member - Dr. G. Butchaiah, Former Dean, Rajiv Gandhi College of Veterinary and Animal Sciences,
Kurumbapet, Puducherry –605 009

Member - Dr. K. Thangaraj, Scientist, Centre for Cellular and Molecular Biology, Uppal Road,
Hyderabad - 560 007