

# TANUVAS TECHNICAL REPORTER

*An official publication of research and developmental activities of  
Tamil Nadu Veterinary and Animal Sciences University*



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## TANUVAS PHARMACOVIGILANCE LABORATORY FOR ANIMAL FEED AND FOOD SAFETY –A TREND SETTER IN RESIDUE MONITORING

Pharmacovigilance Laboratory for Animal Feed and Food Safety (PLAFFS), a National Facility supported by DST and APEDA is functioning under the Directorate, Centre for Animal Health Studies (CAHS), Tamil Nadu Veterinary and Animal Sciences University (TANUVAS). It offers the facility of testing and reporting the levels of Mycotoxin, Drug residues, Pesticides and GMO's from various feeds, feed stuff and livestock farmers to prepare their products as per International Standards.

The erstwhile Central Animal Feed and Food Residue Laboratory (CAFFRL) unit, the fore runner of **Pharmacovigilance for Animal Feed and Food Safety** was started as one of the service units of Central University Laboratory, TANUVAS in the Year 1993.



In 1993

During the year 2007, a scheme entitled "Pharmacovigilance Laboratory for Animal Feed and Food Safety" was sanctioned by Department of Science and Technology, New Delhi. The main objective of the scheme is to have a constant



In 2007

Pharmacovigilance on veterinary drugs in food and feed and to generate a data base on the same,

### Quality Policy

- Management complies with ISO: IEC 17025:2005 standard is maintained continually to improve the effectiveness of the Management System of Testing Services.
- Management ensures that the integrity of the management System is maintained when changes to the management system are planned and implemented.
- Management ensures that all the laboratory personnel familiarise themselves with the content of Quality Manual Associated Documentation and comply with the laid down policies and procedures, at all times.
- The management of PLAFFS is to offer the facility of testing and reporting the levels of Mycotoxins (Aflatoxin, Ochratoxin A, Citrinin, Patulin and T2) in various feeds, feedstuffs and livestock products, thereby helping livestock farmers to prepare their products as per International standards. The service of this laboratory is open to continuous improvement based on feedbacks and findings and through the application of the Quality Policy and Quality objectives focused with three 'D's - Desire, Dedication and Determination.

### Vision

- To prepare and meet out WTO Challenges pertaining to xenobiotic residues (Veterinary drugs, Pesticides and Mycotoxins).
- Labeling of livestock foods GMO/Non GMO.

- To establish poison control information service.
- Therapeutic drug monitoring.

**Mission**

- ▲ Assuring safe food for human consumption and for inland and global trading by establishing a system for testing.
- ▲ Establishment of single window system for residue monitoring.
- ▲ Capacity building.

**Mandate**

The laboratory is currently carrying out the analysis of animal feed and food as well as biospecimens for the presence of the following

- Mycotoxin – Aflatoxin, Ochratoxin, Citrinin, Penicillic acid, T2 Toxin, Zearalenone.
- Pesticides – Organochlorine, Organophosphorous and Pyrethroids.
- Veterinary drugs.
  - Banned Veterinary drugs – Chloramphenicol, Nitrofurans, Sulphanamides, Enrofloxacin, ciprofloxacin oxytetracycline and neomycin.
  - Drugs with permissible level – Tetracycline, Macrolide and Betalactam.
- Screening of Adulterants – Melamine/Urea.
- Screening for GMO in Feed/Livestock Products.
- Pharmacotoxicological diagnosis in disease outbreak (Nitrate/Nitrite, Zinc Phosphide, Urea toxicity, Hydrocyanic Acid toxicity).

**Facilities Available at PLAFFS as per European Union / FDA Norms**



High Performance Thin Layer chromatography (HPTLC) for Screening of Mycotoxins, Pesticides and Veterinary Drugs



High performance Liquid Chromatography (HPLC) for identifying Mycotoxins



Gas Chromatography (GC) for identifying Pesticides



Gas Chromatography Mass Spectrometer (GC MS) for molecular fragmentation and confirmation of Melamine and Pesticides



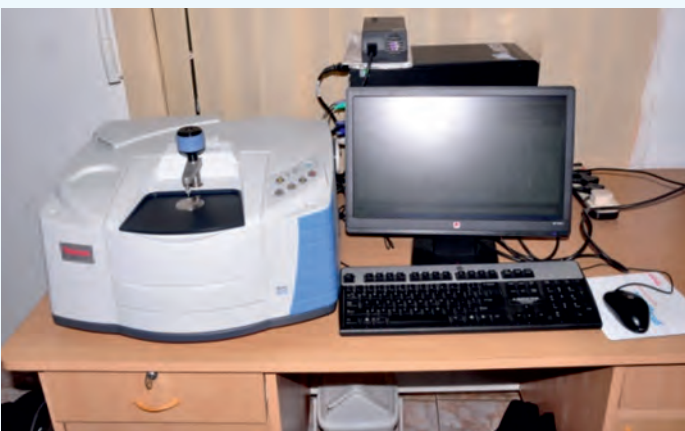
Liquid Chromatography Mass Spectrometer (LC MS MS) for molecular fragmentation and confirmation of Veterinary drugs



Automated Gel Permeation Chromatography (GPC) for cleanup of samples prior to LC MS MS/GC MS identification



Detection of GMO'S first of its kind in the country for screening livestock products



Fourier Transform Infrared Spectroscopy for identifying organic chemicals

The laboratory has to its credit completion of four research projects and has one ongoing project entitled "Establishment of Veterinary Forensic Sciences Laboratory".

#### Hallmark

- T2 toxicosis in ostriches - 2000
- Organophosphorous toxicity – 2001
- Nitrate/Nitrite toxicosis in Equines-2001

- Urea toxicity in heifers-2002
- Nitrate/Nitrite toxicosis in cattle-2004
- Zinc phosphide toxicity in monkeys-2005
- Hydrocyanic acid toxicity in cattle-2005
- Nitrate/Nitrite toxicity (Weeds) in ponies and cattle-2008
- Hydrocyanic acid toxicity (Tapioca) in cattle-2009
- Organocarbamate – Carbofuran toxicity in elephants-2009
- Tetradeconic acid toxicity in heifers-2010
- Diagnosed Malathion poisoning in ducks - 2016
- Diagnosed Dichlorovos poisoning in peacock -2016
- Diagnosed Dichlorovos poisoning in ducks -2017

#### PRESENT ANALYTICAL TARIFF

Name of the Parameter	Detection method	Students Charges/ Test (Rs.)	Tamil Nadu Charges/ Test (Rs.)	Outside state Charges/ Test (Rs.)
<b>PESTICIDES</b>				
Organocarbamates	Thin Layer Chromatography (TLC)	300	400	500
Organochlorine	Gas Chromatography (GC) per Compound	800	1200	1300
Organophosphates, Pyrethroids				
	Gas Chromatography Mass Spectrometer (GCMS) per Compound	750	1200	1300
<b>VETERINARY DRUGS</b>				
Chlortetracycline	Thin Layer Chromatography (TLC)	300	400	500
Ciprofloxacin				
Enrofloxacin	High Performance Liquid Chromatography (HPLC) per Compound	800	1200	1300
Salinomycin, Tiamulin, Tylosin				
Chloramphenicol, Furazolidone and metabolites	Liquid Chromatography Mass Spectrometer (LCMS/MS) per Compound	1000	1500	2000
<b>MYCOTOXINS</b>				
Aflatoxins	Thin Layer Chromatography (TLC)	250	300	350
Multimycotoxins				
Ochratoxin A, Citrinin, T2 toxin, Penicillic acid, Zearalenone, Patulin and Rubratoxin	High Performance Thin Layer Chromatography (HPTLC) per Compound	800	1200	1300

	High Performance Liquid Chromatography (HPLC) per Compound	800	1200	1300
Genetically Modified Foods Detection (GMO)	PCR Based Micro Array	5000 per sample	8000 per sample	10000 per sample
Nitrate/ Nitrite	-	300	600	1000
Urea	-	200	250	300

#### Completed / Assisted Research (M.V.Sc. / Ph.D.) work

- Estimating the Organochlorine pesticide residues in poultry feedstuff, feeds and products in 2007.
- Estimating Methionine and lysine in the Experimental feed in 2008.
- Assessment of residue levels of antibiotics in broiler chickens in 2009
- Residue profile of Chlortetracycline and tylosin in chicken eggs in 2010
- Incidence of Pesticide Residues and Assessment of Physico-Chemical Parameters in Grape and Its Products in 2012.
- Incidence of Aflatoxin Contamination and Assessment of Physico-Chemical Parameters in Breakfast Cereals in 2012.
- Ameliorating effects of Aflatoxicosis using toxin binders in dairy cattle in 2013.
- Exploration of fertility effect of siddha medicines (Nandhi mezugu and Kumatti kuzhambu) in experimentally induced cystic ovarian degeneration in rat model in 2015.
- Patulin prevalence and detoxification profile in apple juices marketed in Chennai, India in 2016.
- Assessment of different decontamination procedures for chlorpyrifos residue in cabbage and cauliflower in 2016.

#### Ongoing Research work

- Safety assessment of Distillers dried grains with solubles (DDGS) in Dairy cattle and Desi- Chicken.
- Development of molecular based diagnostic kit for the detection of antibiotic residues in meat and milk.

#### ACCOLADES received by the Laboratory

#### ILACMRA- OCI- NABL Accreditation

PLAFFS has been Accredited by NABL for chemical testing. Preassessment was conducted on 07.02.2017 by the NABL Lead Assessor. Final assessment was conducted on 18.03.2017 and 19.03.2017 to check both the management and technical competence of the laboratory

by the NABL Lead Assessor and Technical Assessor. PLAFFS has obtained grant of accreditation for chemical testing in accordance with ISO/IEC 17025:2005 on 31.03.2017 valid till 30.03.2019. The laboratory is first of its kind to get accredited for Mycotoxin analysis by High Performance Thin Layer Chromatography in the country. The first Veterinary University having obtained accreditation for Mycotoxin analysis in animal feed and Agricultural products.

#### The Laboratory has been accredited for following Recommended Scopes

S. No	Group	Specific Tests Performed	Test Method
1	Animal Food & Feeds Sub group -1 (Cattle Feed, Poultry Feeds, Swine Feed, Laboratory Animal Feed and Pet Foods	Aflatoxin B1 Aflatoxin B2, Aflatoxin G1, Aflatoxin G2, T2, Ochratoxin A and Citrinin	TLC and HPTLC
2	Food & Agricultural Products (Cereals, Pulses, Spices, Fruit and Fruit products)	Aflatoxin B1 Aflatoxin B2, Aflatoxin G1, Aflatoxin G2, T2, Ochratoxin A, Citrinin and Patulin	TLC and HPTLC
3	Animal Feed, Livestock drinking water, Milk	Adulterants – Urea	IS 14832:2000 FSSAI MANUAL 2015 –Milk - S.No:1.2.4

#### Human Resources Development activity of the Laboratory Course offered

- Food analysis courses offered for Final year B. Tech (FPT), M.Tech and PhD students, College of Food and Dairy Technology, Koduvalli.
- Courses offered for M.V.Sc students, Department of Veterinary Pharmacology and Toxicology, Madras Veterinary College, Chennai- 600 007.
- Post Graduate Diploma Course on Food Toxicology and Safety Assessment from current academic year.

#### Training offered

- Five days training programme in Chromatographic techniques.

#### Dr. G. Sarath Chandra

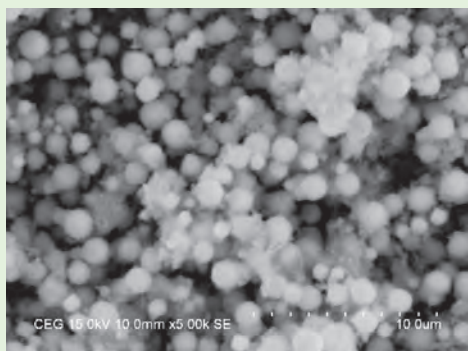
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## RESEARCH HIGHLIGHTS

(2015-16)

### Nanotechnology facility to augment farm animal production and health

- ▲ Copper (Cu), Zinc Oxide (ZnO), Iron (Fe) and Chitosan nanoparticles were synthesized and characterized. *In vitro* cytotoxicity assay results indicated that synthesized nanoparticles were non-toxic to Vero cells (Green monkey kidney cells), PK 15 cells (Pig kidney cells) and MDBK cells (Madin-Darby Bovine Kidney cells) at lower concentrations.
- ▲ Preliminary *in vivo* study results indicated that synthesized nanominerals used as feed additive were non-toxic to goats.
- ▲ Chitosan nanoparticles coupled Newcastle disease Virus (NDV) fusion protein was found to be protective when challenged with virulent NDV and chitosan coupled *Mycobacterium avium* subsp. *paratuberculosis* (MAP) in goats. Preliminary data generated indicated that MAP antigen gave better immunogenic response when delivered through chitosan nanoparticles.



SEM image of ZnO NPs

**Principal Investigator: Dr. K. Vijayarani**

Professor and Head,  
Department of Animal Biotechnology,  
Madras Veterinary College, Chennai

### Immunogenicity of recombinant fusion protein of Newcastle disease virus coupled with biodegradable nanoparticles

- ▲ Fusion protein of NDV-2K3 was cloned and expressed from *Pichia pastoris* and the size of the protein was 55 kDa

- ▲ The recombinant fusion protein was coupled with biodegradable nanoparticles such as PLGA and chitosan
- ▲ Indirect ELISA revealed that PLGA coupled fusion protein elicited a better immune response (0.640) compared to chitosan nanoparticles (0.608) coupled fusion protein.

**Research Scholar: Dr. S. Arul**

Department of Animal Biotechnology,  
Madras Veterinary College, Chennai

**Chairman: Dr. K. Vijayarani**

### Effect of computationally optimized broadly reacting spike1 protein polypeptide of innate immune response to avian infectious bronchitis virus

- ▲ For the first time, the broadly reacting epitopes of IBV of major serotypes prevalent globally have been designed, synthesized, cloned and expressed.
- ▲ *In silico* analysis showed that these epitopes have been present in all the IBV of major serotypes prevalent globally.

**Research Scholar: Dr. Bezina Arega Emeru,**

Department of Veterinary Microbiology,  
Veterinary College and Research Institute,  
Namakkal

**Chairman: Dr. A. Raja**

### Formulation of an inactivated vaccine for Infectious Laryngotracheitis

- ▲ Infectious Laryngotracheitis virus (ILT) isolated from field outbreaks were passaged in embryonated eggs, inactivated with binary ethyleneimine and vaccine formulated with Montanide adjuvant.
- ▲ A methodology for developing an inactivated ILT vaccine was established.

**Research Scholar: T. Shiny Jenifer**

Vaccine Research Centre - Viral Vaccines,  
Centre for Animal Health Studies,  
TANUVAS, Chennai

**Guide: Dr. K. Brinda**

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## Technology Developed at TANUVAS Innovations and Instrumentation Centre

### TANUVAS – ILFC - Guiding Pad for Pigs



- ✦ Designed to guide or direct pigs to desired destination/place/sty/pen with ease for relocation, weighing, etc.
- ✦ Can be used to transfer pig /group of pigs from one place to another.
- ✦ Made of thick board of 4 ft length and 3 ft height.
- ✦ Provided with two oval holes at the top for holding the guiding pad and one in the middle for pig vision.
- ✦ Minimize labour and time.
- ✦ Easy to handle.
- ✦ Available in two different models: one for guiding single pig and other for guiding group of pigs.
- ✦ Cost effective and user friendly for small, medium and large farmers.
- ✦ Single pad price Rs.600/-
- ✦ Double pad price Rs. 1,200/-

**Inventors :** Dr. P. Tensingh Gnanaraj , Dr.T. Muthuramalingam,  
Dr. E. Rachel Jemimah

### TANUVAS – ILFC - Calf Comfort Hutch



- ✦ TANUVAS – ILFC – Calf hutch is designed to improve health and growth of calves prior to weaning.
- ✦ Total length of calf hutch is 10.5 feet and width of the hutch is 4.5 feet. It has two division, pen and run. Length of pen is 4.5 feet, length of run is 6 feet.
- ✦ Rear height of pen is 5 feet, front height of pen is 6 feet and front height run is 5 feet.
- ✦ Calf enter into the calf hutch through upside down inlet door.
- ✦ Neck lock in calf hutch allows controlled feed and water to calf.
- ✦ Neck lock helps to provide clean feed and water to the calf at required time.
- ✦ Has installed detachable milk bottle.
- ✦ Unlocking neck lock at night allows the calf to move itself into the pen. So calf hutch provide night shelter to calf.
- ✦ Calf hutch helps to prevent the spread of contagious disease, reduce calf mortality.
- ✦ It is inexpensive. Properly designed calf hutch provides excellent natural ventilation which further reduce incidence of respiratory disease.
- ✦ It is light weight and portable, making them easy to clean.
- ✦ If calf becomes ill, you can move susceptible calves further away from other calves. It can also easily move to different areas of the lot or pasture.
- ✦ Calf hutch provide more space allowing more freedom for the calf to exercise and roam outside.
- ✦ Calf hutch allow greater control and management. A clean, dry, well ventilated hutch provides the best opportunity for a calf to stay health and growing.
- ✦ Cost Rs.15,000/-

**Inventors :** Dr. P. Tensingh Gnanaraj, Dr. T. Geetha and  
Er. M. Siddharth