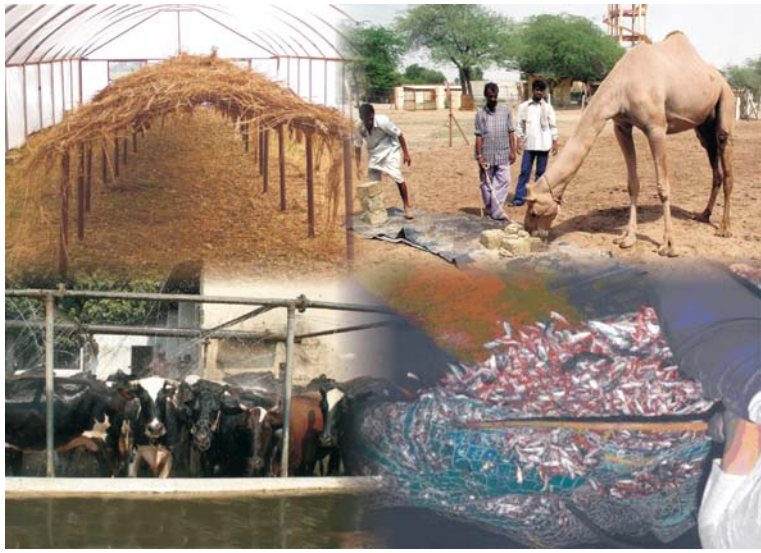

8. Livestock Management

ANIMAL MANAGEMENT

Cattle

CLA content in milk: The conjugated linoleic acid (CLA) content was significantly higher in milk of both cows and buffaloes fed high forage diet. Providing high forage diet increased milk yield in buffaloes as compared to high concentrate diet. CLA content was almost double in ghee prepared by indigenous method as compared to creamery method.

Sustainable milk production system

Annual milk productivity per cow, per labour and per acre land was sustained at 5,100, 14,450 and 8,640 kg, respectively, through application of integrated system approach comprising integrated breeding system (IBS), straw based feeding system, eco-friendly housing system and optimum herd management system. The sustainable milk production system was validated at a Gaushala.

Chelated zinc: Chelated zinc was prepared by enzymatic hydrolysis of soy protein in a single stage or double stage process using different enzymes to achieve maximum degree of hydrolysis (DH) for the preparation of zinc chelate. Per cent recovery of zinc declined as the level of zinc used for chelation increased. Absolute content of zinc in chelate increased as the addition of inorganic zinc increased from 5 to 20%. Protocols/technology for the preparation of chelated zinc by enzymatic hydrolysis of soy protein was developed and tested for its efficacy.

Energy expenditure: Energy expenditure (EE) of cattle was less when fed chaffed paddy straw *ad lib.* and at restricted level of intake, compared to that of un-chaffed paddy straw fed *ad lib*. Chaffing of maize stover resulted in 15–17% lower energy expenditure by animals, thus saved biological energy for productive purposes. Chaffing resulted in 11–12% reduction in CO_2 and 20–25% reduction in methane production by cattle.

Detoxification of aflatoxins: Plant extracts like olive oil, garlic and turmeric showed highest efficacy in reducing aflatoxin levels in feeds. Ethiglacetate extract of herbal products like neem-bark, annaa-seeds with witharia prevented mycotoxin contamination in stored feeds.

Pesticide residues: An analytical technique was developed for measuring the residue of imidacloprid in water and soil samples. A detection limit of 0.5 ppb was achieved.

Jatropha cake for animal feeding: Jatropha cake, a byproduct of jatropha, which is being promoted for bio-diesel production, contained phorbol esters an anti-nutritional factor responsible for toxic effects in animals. The seed-kernels were rich in crude fat and protein.

Microbial protein: Supplementation of finger millet straw based ration with rumen degradable nitrogen showed that digestibility of nutrients, PDC index and estimated microbial protein synthesis (spot as well as total urine) was comparable in all the supplemented groups. The optimum level of microbial protein synthesis was at 15 g RDN (rumen degradable nitrogen) level of supplementation with finger millet straw based diet.

Feed processing: Cooking significantly reduced IVDMD of maize, sorghum and finger millet grains while processing of wheat grains did not have any effect on the IVDMD. Treating maize, sorghum and rice grains with microwave significantly reduced IVDMD.

Evaluation of sperm quality of Frieswal bulls: Evaluation of frozen semen of Frieswal bulls showed that progressive motility ranged from 58.48 and 43.13% and acrosomal damage from 7.55 to

18.26%. The GOT and GPT enzyme activity in seminal plasma did not differ among bulls, whereas ACP and AKP differed. The total sperm abnormalities in Frieswal semen did not differ among bulls of different age groups. Proportion of free heads decreased with the advancement of age.

Embryo transfer: Superovulated Frieswal cows — 3 with folltropin V and 2 with folligon — produced 30 embryos, out of which 6 were of transferable quality. Transfer of these embryos to 5 recipients resulted in birth of two calves (one male and one female).

Augmenting reproductive efficiency: Additional energy supplementation through locally available ragi (finger millet) during the first month of postpartum (transient period) augmented reproductive efficiency in crossbred cattle in field condition. Ragi feeding during transient period avoided a steep fall in body condition score during first month postpartum. Blood plasma glucose and cholesterol showed significant negative correlation with the day of occurrence of first postpartum heat, but blood urea nitrogen, milk urea nitrogen and milk yield showed a positive correlation.

Stress related enzyme assay: Methodology for determination of stress related enzymes i.e. superoxide dismutase (SOD) and glutathione peroxidase (GSH-Px) was standardized both in blood plasma and haemolysate.It could be used as a tool to asses the effect of stress on animals.

Buffalo

Strategic supplementation for improving production: Feeding wheat straw/*bajra kadbi* based **complete feed blocks** improved milk yield in buffaloes. Incorporation of **limiting amino acid rich supplements**, like cottonseed cake and soybean cake in the diet of buffaloes resulted in 6–10% increase in milk yield. **Energy requirement** of pregnant buffaloes could be met with 55% TDN and 12% CP during ninth month, and with 60% TDN and 14% CP during the last month of gestation. **Higher plane of nutrition** did not improve conceptus growth, milk production or body condition score in early lactation. For optimum growth of Nili-Ravi buffalo heifers (647 g/day) the **concentrate level in diet** should not exceed 60–70% of DM intake as higher concentrate diet predisposes animals to ruminal acidosis. **Economic feeding regimen** was developed using urea impregnated sherry waste in combination with barley grain in place of conventional concentrate supplements for male buffalo calves for meat production. **Postpartum buffaloes** fed 120% of energy requirement had low reproductive performance in comparison to buffaloes fed either 80 or 100% of energy intake indicating that higher energy allowance than recommended levels may not be necessary.

Limiting amino acid in buffaloes: Buffaloes fed high bypass protein (UDP) rich in limiting amino acids diets showed an increase in milk production over that of controls. Milk fat percentage increased in all animals with progress of lactation indicating that ration for high yielding buffaloes need to be supplemented with bypass protein rich in limiting amino acid (lysine and methionine).

New fungus genus identified: *Cyllamyces icaris* p. nov., a new anaerobic gut fungus with nodular sporangiophores was isolated from the dung sample of *Bubalus bubalis*. It resembled the previously described *Cyllamyces aberensis*. These fungi hold special significance in the degradation of poor quality feed in ruminants.

Foot-and-mouth disease

Samples received from various states were processed and field samples revealed type Asia 1, type O and type A. Two-dimensional micro-neutralization test (2D-MNT), a modified form of SNT, was routinely used to test new field isolates to determine the appropriateness of the existing vaccine strains and to select new vaccine strains, if required. Serotype A isolates showed an r-value of <0.2 with current vaccine strain IND 490/97 implying the demand for new vaccine strain. Molecular epidemiological studies revealed endemic co-circulation of genotype VI and VII since 1990 in type A. But since 2001, dominance of genotype VII and disappearance of genotype VI from field was noted. FMDV type A recent field isolates were processed for molecular epidemiological analysis. In phylogenetic tree, all the isolates were clustered in genotype VII as with previous years indicating incessant supremacy of genotype VII in the field.

Genotype differentiating ELISA and RT PCR were developed to distinguish between genotype VI and VII. It gives a fast preliminary indication on the genotype prevalent before proceeding with thorough sequencing of 1D region, which definitely continues to be the confirmatory method to assign genotype by phylogenetic analysis. Similarly genotype-differentiating ELISA was also developed to find out the genotype of type A. Since this assay is very simple and technically not demanding, samples can be subjected first to ELISA and then ELISA—negative samples can be subjected to PCR. Both the assays were effective in molecular epidemiological investigation of FMDV type A in the country.

Strains IND 40/00 and IND 81/00 were selected as candidates for FMDV type A vaccine. IND 81/00 showed marginally superior antigenic relatedness with the recent field viruses than IND 40/00, and hence both of these are effective vaccine candidates in present context. At present national repository contains 1,292 (818-O, 257-Asia 1,202-A, 15-C) field isolates.

Improvement of semen quality: Buffalo bull semen collected during different seasons, revealed significant seasonal variation in seminal plasma AKP, ACP, GOT and GPT in pre-freezing and post-freezing sperm motility, but not in ejaculate volume, mass activity, seminal plasma protein, cholesterol and testosterone level. Semen samples with higher values of sperm total motility, progressive motility, rapid motion and viability showed that these can be used as indicators for assessing semen quality and predicting fertilizing potential.

Zinc: Its supplementation significantly improved semen characteristics, viz. motility, sperm constriction and serum testosterone in buffalo bulls; organic form of Zn performed better in terms of per cent live spermatozoa and acrosomal integrity in addition to *in vitro* fertilizing ability.

Immunoblotting technique for detection of anti-oxidant enzymes: A sensitive technique for detection of anti-oxidant enzymes superoxide dismutase (SOD) and catalase (CAT) in serum, follicular fluid, oviduct, uterine secretions of pregnant and non-pregnant animals, was developed. The results confirmed that activities of both Cu-Zn SOD and catalase modulate according to the stage of estrous cycle and early pregnancy in buffaloes.

Vitrification of *in vitro* matured buffalo oocytes: For successful vitrification, BSA supplementation in maturation media showed positive influence on post-thaw survival and maintenance of developmental competence of *in vitro* matured buffalo oocytes, in comparison to FCS supplement. Post-thaw IVF-rate of *in vitro* matured buffalo-oocytes vitrified using ethylene glycol (EG) was high.

Blood profile: Plasma albumin levels were lowest in heifers and highest in late pregnant dry animals. Globulin levels varied nonsignificantly between different groups of diverse reproductive status. Blood urea was significantly higher in late pregnant animals as compared to heifers.

Environmental pollutants: High concentrations of heavy metals particularly cadmium caused cell death, lowered the growth potential of oocytes/ follicular somatic cells and sperm functions leading to infertility in buffaloes.

Sheep

Strategic supplementation for improving production: Supplementing 200 g of **concentrate mixture** during 3–6 months of age of lambs maintained on community grazing lands resulted in additional benefit of Rs 300/lamb. Supplementation of 300 g concentrate mixture during pregnancy in sheep maintained on grazing lands was adequate to meet the requirement of dam as well as for growing feotus**. Fallen tree leaves and stovers** of coriander cumin, *methi*, fennel, *ajwain* could be used as partial source of roughage in complete feed block. Utilization of abundantly available **damaged wheat** as a replacement of conventional and costly cereal supplement in lambs could economize cost of mutton production.

Standarised transcervical AI technique: Frozen-thawed semen using transcervical artificial insemination (TCAI) technique was standardized. A protocol for obtaining multiple births through precise control of ovulation using exogenous hormones was developed. The efficacy of indigenous vaginal sponges in inducing estrus among anoestrus ewes during non-breeding season was tested and found effective in regulating anoestrus cases.

Goat

Socio economic study

Black Bengal: Socio-economic condition, management practices and housing pattern were studied among adopted villages. This breed is highly prolific having 83.72% multiple births. The kidding rate was 1.80, the highest among all the goat breeds of the country. The age and weight at first kidding were 378±2.12 days and 13.52±0.22 kg. The overall mortality was 7.8% under field conditions. The socio-economic studies revealed that in Nadia district of West Bengal goat rearing proved more beneficial to the goat keepers having basic knowledge of animal husbandry. The annual income of a family was Rs 2,384.06 from goat rearing.

Black Bengal goat is highly prolific having 83.72% multiple births

Sangamneri: Baseline information on growth, production and reproduction were recorded. Economic gain through increase in body weight for 3 and 6 months of age, was Rs 42.40 and 30.40, respectively. Similarly economic gain through increase in milk production was Rs 176.40 for 90 days estimated milk yield.

SUCCESS STORY

Commercial Goat Farming

Goats play a significant role in ensuring livelihood security to the millions of small and marginal farmers, landless labourers and rural folk. Goat rearing under intensive and semi-intensive system for commercial production is gaining momentum. A progressive farmer, Shri Rohan Singh resident of Salempur village in Farah block of Mathura district of Uttar Pradesh, started a goat farm in his village with 68 Barbari does and 2 bucks, and after 6 months 33 does and 1 buck more were added to the flock. The objective of this goat-rearing project was to produce and market pure breed Barbari goats.

The initial investment made on purchasing of breeding stock and construction of sheds and equipments was Rs 1.62 lakh and another Rs 10,000 was used as working capital. The goats on this farm were maintained under semi-intensive system of management by the two unemployed youths of the family. Besides grazing, the animals were provided supplementary concentrate feeding, mineral mixture, fodder tree leaves lopping and guar straw.

Total annual expenditure incurred on supplementary feeding of goats worked out to be Rs 10,700, and on prophylaxis schedule (vaccinations against enterotoxaemia, FMD and PPR disease and twice medication against internal and external parasites) and treatment Rs 3,500. Thus the total recurring expenditure other than family labour for a flock of 104 goats was Rs 14,200 during one year. The returns from the sale of goats in one year were estimated to be Rs 75,000. Moreover the goat manure valuing Rs 4,000 was produced and used in the agricultural farm of the owner. Thus the annual net returns to the family from goat rearing was Rs 64,800. The farmer sold all the surplus animals (pure Barbari goats) for breeding purpose to the other goat farmers at the rate of Rs 100 per kg live body weight. Concurrently the other traditional farmers of this area maintaining non-descript goats could fetch a market price of Rs 60–65 per kg of live body weight for their improved goats sold mostly for meat purpose.

Ganjam: The information regarding production and reproduction parameters was collected from the adopted area. The kidding percentage on the basis of does tupped was 73.18. In the Ganjam district of Orissa the goat is a primary source of income of tribals (Gola). The goat rearing contributed 61.54% of their annual income.

Surti: The survey work on Surti goats was conducted in Bharuch district. The improvement of 15.10% was observed at 3-month body weight due to the use of elite bucks under field conditions. Females were registered and bucks were selected from the population.

Surti—Body weight improvement was observed with the use of elite rams for breeding

Malabari: In the three village centres females were registered and bucks were selected from the population. The average gestation length, age at first kidding and inter-kidding interval were 149.34±1.49, 381.36±18.33 and 263.13±15.62 days.

Sirohi: The population growth of the flock in field was 64.93%. The breeding efficiency on the basis of does kidded was 112.66, which was lower than previous year.

Low cost animal feed mixer developed: A low cost animal feed mixer was designed and developed to mix heterogenous feed ingredients before preparation of complete feed pellets and complete feed blocks. The feed ingredients comprising 50% straw (*Cajanus cajan*) and 50% concentrate with molasses could be uniformly mixed in 10–15 min with an output of 0.6 q/hr to 1.2 q/hr. The cost of mixer is Rs 20,000 and cost of mixing 100 kg feed is Rs 25 only.

Evaluation of embryos: *In-vitro*-produced embryos at 2- to 16-cell stage were cryopreserved using conventional freezing protocol. Approximately 50 IVRP embryos were used for freezing. After thawing none of the embryos had broken zona pellucida. However, blastomere membrane reptured and their cytoplasm were found disintegrated.

Early pregnancy diagnosis: The B-mode sono graphic imaging technique was standardized enabling screening of pregnant goats within 30 days of mating, thereby, allowing rebreeding of empty animals for increasing flock fertility.

Cardinal physiology response: Thermo-regulatory characteristics of Jakharna and Sirohi bucks at different temperatures revealed that they could adapt to hot-dry, hot humid and cool periods by increased heart and respiratory rate. Temperature around 9°C seems to be critical for bucks in cool period, and ambient temperature of 40°C and vapour pressure of 20.67 mmHg during hot humid conditions.

Pig

Economic rations were formulated for grower and finisher pigs by partial replacement of costly feed ingredients with the locally available feed resources, e.g. maize with tamarind seed and molasses, wheat bran with de- caffeinated waste, fish meal with dried cuttla fish waste, silage and concentrate mixture with green cabbage leaves. Supplementation of chelated minerals with dicalcium phosphate and incorporation of coconut oil in the grower diet improved growth and feed conversion efficiency.

Camel

Unique sperm depot: Semen evaluation revealed zero or low motility in freshly ejaculated semen from camel due to formation of a unique depot of spermatozoa from which spermatozoa are continually released over a prolonged period of time and does not exhibit mass motility and individual sperm motility as seen in the cattle, buffalo, sheep and goat. The spermatozoa develop motility only after liberation from this depot, and this delay was not due to use of rubber funnel as was speculated earlier.

Semen sample after centrifugation

Gelatinous sperm pellet

Estradiol profiles: Estradiol profiles in unmated female camels are different from those of other species of livestock. These profiles monitored for 60 days in unmated female camels revealed that basal levels are relatively very high and peak levels of estradiol were observed at irregular intervals.

Mithun

Tree leaves based total mixed rations: *Lagerstroemia speciosa* (*thumero*), one of the important tree foliages having good nutritive value (8% DCP and 48% TDN), is relished by mithun under natural browsing condition. Inclusion of *Lagerstroemia speciosa* as green foliage in total mixed ration (TMR) showed higher daily body weight gain, dry matter (DM) intake, nutrient utilization and feed conversion efficiency in growing mithuns, suggesting that these tree leaves can be incorporated successfully in TMR for feeding of growing mithuns.

Vegetative propagation of selected foliages: The propagation study by using higher dosage of auxins on good quality fodder trees revealed encouraging trend with highest survival percentage, increased length and number of sprouts per stem cuttings of selected fodder tree species (*Ficus hirta*, *Ficus roxburghii*, *Lagerstroemia speciosa*, *Trema orientalis*, *Ficus hookeri*). Stem cuttings treated with IAA provide better option for multiplication in degraded pastureland of low carrying capacity.

Standardization of semen collection method: Collection of semen from mithun bulls is difficult, as they do not mount cows not-in-estrus. An innovative method of sprinkling urine from estrus

SUCCESS STORY

Successful pig farming

Shri Juby Mathew an agricultural graduate from Kodannur, Thrissur, Kerala, started a pig farm with technical assistance from the All India Coordinated Research Project on Pigs, with an initial investment of Rs 4.0 lakh in 2001. He currently maintains the parent stock of Large White Yorkshire pigs (34 adult female and 4 breeding males). More than 700 piglets per year were produced at the farm and parental stocks were also changed regularly. From each lot of piglets 50% of piglets are disposed after weaning and 50% are kept for fattening. The entire herd is fed with hotel swill, which he collects free of cost by his own tempo van and no feed additives are given to pigs. Using a locally available feed material, the hotel waste, he could achieve a litter size at birth and weaning ranging from 8–12 piglets and 7–10 piglets, respectively, and a body weight of 120 kg at 10 months of age. After 10 months, pigs were sold @ Rs 50–60/kg of body weight depending on demand of the market. Piglets are sold between Rs 800–1,000/piglet. Besides a piggery, he maintains fishery, poultry, duckery, and practices horticulture with spices and flowers. Alone from piggery, the turnover is about 2.5 to Rs 3 lakh/annum.

Ethno-veterianry medicine

Herbs used for common ailments and conditions of livestock were documented with information on their botanical and local names, active constituent, validation for use, etc, and 39 of them were identified by different centers for their evaluation in fluorosis, diarrhoea, mastitis, fever, helminthosis and ectoparasite infestation and as antipyretic, analgesics and hepatoprotectant. Experimental trial in rabbits established the ameliorative potential of tamarind fruit pulp (*Tamarindus indica*) in experimental fluoride toxicity. Methanolic extract of stone fruit (*Aegle marmelos*) powder given @ 400 mg/kg body weight in rats reduced 73.33% faecal output in castor oil induced diarrhoea. *In vitro* screening results showed that methanol extract of *ghila* (*Entada fascioloides*) seeds possessed activity against *Fasciola. In vitro* antibiogram of extracts of turmeric (*Curcuma longa*) and Indian gooseberry (*Phyllanthus emblica*) revealed antimicrobial efficacy against common mastitis pathogens, viz. *Staphylococcus aureus, Streptococcus agalactiae*, other *Streptococcus* spp. and coliform bacilli.

Utility of aqueous and alcoholic extracts of plants was determined in— *babrang* (*Emblia ribes*), *palash* (*Butea frondosa*), *kaliziri* (*Vernonia anhelmintic*a) against nematodes; Auatralian fever tree (*Ecalyptus globules*), *amrita giloy* (*Tinospora cordifolia*), *pippali*, *long*, *pipar* (*Piper nigrum*), *adulsa* (*Adhatoda vasica*), *henna* (*Lawsonia inermis*), *nirgundi* (*Vitex negundo*), *ashwagandha* (*Withania sominifera*), *punarnava* (*Boerhaavia diffusa*) as antipyretic/analgesics; and *kalmegh* (*Andographis paniculata*), *punarnava* (*Boerhaavia diffusa*), *bhrangraj* (*Eclipta alba*), *chirayata* (*Swertia chirata*) as hepatoprotectants.

cows over the perineal region of mithun cow not-in-estrus helped bulls donating the semen successfully. Urine collected from estrus cows stored at refrigerated temperature was effective till 7 days of post collection.

Estimating FSH in plasma: A simple and sensitive radioimmunoassay (RIA) procedure to estimate FSH in mithun plasma was developed. The biological validation of the assay was carried out in plasma samples that were collected during different stages of the estrous cycle.

Feed resources

District-wise database of feed and animal resources was prepared for 6 agro ecosystems of the country. The current requirement and dry matter availability from different feed resources and the projected demand and supply over the years showed that by 2020 the gap between requirement and availability should marginally increase from the current level of 19%. However, there would be a widening gap between demand and supply of concentrates.

Efficacy of AI in mithun established

The first mithun calf was born through artificial insemination using cryopreserved semen samples. The semen samples were collected from adult bulls through rectal massage method and

AI born mithun calf

cryopreserved in liquid nitrogen using tris-egg yolk-glycerol diluent. Three mithun cows inseminated using the cryopreserved semen conceived following insemination and gave birth to healthy calves.

Estrus synchronization using different protocols: Two injections of $PGF_2\alpha$ were given at 11 days apart in cyclic mithun cows for estrus synchronization, and found that all mithun cows responded to treatment. Cyclic mithun cows irrespective of any day of estrous cycle were subjected to ovsynch protocol of estrus synchronization and were inseminated following synchronization. Of the 16 animals inseminated artificially (AI) with the cryopreserved mithun semen, 12 conceived.

Yak

Complete feed block formulation: Complete feed formulation in the form of feed block was prepared utilizing the cost effective locally available feed resources for yak feeding.

Complete feed blocks

Mineral supplementation: Hair tissue mineral analysis (HTMA) proved an excellent tool for monitoring general health, nutritional status and toxic metal exposure to animals. Hair mineral profile of the yaks following supplementation of trace minerals zinc, copper, cobalt and manganese for six months revealed that Mn, Zn, Cu and Co levels of supplemented group were higher than that of the non-supplemented group. The study justified the requirement of the same.

Performance improvement in yaks through trace mineral supplementation: The supplementation of copper, cobalt, zinc and manganese at farm increased the milk production three-times and growth performance, and drastically decreased repeat breeding and anoestrus.

Estrus synchronization: Induction of estrus, synchronization of ovulation and timed artificial insemination (TAI) were attempted in yaks. The ovulation occurred usually within 50–72 hr of ECP injection. TAI was performed at 48 and 60 hr of ECP treatment. Pregnancy rate in yaks following TAI was 40%.

Endocrine status during different stages of growth: A direct simple and highly sensitive enzyme immuno assay (EIA) for GH (growth hormone) determination in yak plasma was developed and validated. Growth rate was positively correlated with mean GH and GH per unit 100 kg body weight. However, GH per unit 100 kg body weight was found to be a better indicator of growth rate. Both the concentrations decreased with advancement of age.

Artificial insemination in yak: Frozen semen technology in yak was standardized. With the use of frozen semen straws, pure yak and yak hybrid calves were born for the first time in India. Aforesaid technology is being successfully utilized in the farm and field conditions for improving yak germplasm.

Yak calf was born using frozen semen straws, for the first time in India

Wild Life

Supplementation of probiotic (mannanoli gosaccharides with or without added β-glucans) positively influenced gut health indices by altering microbial population and fermentative end products in the hindgut of dogs.

Rumen fungi: Out of 243 isolates of anaerobic rumen fungi isolated from domestic and wild ruminants anaeromyces and orpinomyces were predominant. *In vitro* gas production studies of the isolates showed that isolate CTS-110 outperformed all the isolates with respect to total gas production. Genomic library of the isolates was constructed for isolation of the cellulase gene.

Poultry

Supplementation of xylanase, produced from *Aspergillus* spp @ 3,500–4,000 U/kg improved the performance and digestibility of nutrients, reduced the NSP in intestinal digesta (21.7% in jejunum and 10% in ileum) and enhanced the mineral retention (Ca and P) in tibia and protein accretion in muscle (3.7%) over the non-xylanase supplemented DORB diet suggesting that deoiled rice bran can be included in broiler diets up to 15% with supplementation of xylanase @4,000 U/kg.

Se inclusion in diets of broilers improved antibody titers and **Zn supplementation** was needed for better immune response. Both Se and Zn complimented each other for improving immune response, particularly at low Se (0.15 ppm) and high Zn levels (80 ppm) in the diet.

Supplementing **selenium (Se)** or **chromium (Cr)** in organic form in the diet of broiler chicks decreased the activity of lipid peroxidase and increased the activities of glutathione peroxidase, glutathione reductase, RBC catalase and lymphocyte ratio thereby decreasing the holding loss during pre-slaughter fasting period.

Nutrient requirements of female parent line

Bluetongue (BT)

Bluetongue virus (BTV) isolates (15) of types 1, 2, 9, 15, 18 and 23 submitted till date, are being maintained and were characterized. No disease outbreak of bluetongue has been recorded in Uttar Pradesh, Uttarakhand, Haryana, Himachal Pradesh, Gujarat, Rajasthan, West Bengal, Orissa, Jharkhand and North Eastern states in last five years. BT outbreaks are being recorded every year from Andhra Pradesh, Karnataka and Tamil Nadu. Epidemiological maps for Tamil Nadu and Andhra Pradesh were prepared. Disease forecasting model was developed for Maharashtra centre.

Nine monoclonal antibodies clones produced against BTV reacted well with BTV r-Ag and purified BTV antigen in indirect–ELISA. Type specific primer designing, VP2 and VP7 gene cloning and expression, multiplex RT-PCR for BTV and PPRV, RNA-PAGE and nucleotide sequence were standardized. Confirmation of BT virus isolates was done by RT-PCR using VP7 gene specific primers. Inactivated BTV vaccines gave promising results in local and Bharat Merino sheep. Samples of midges from Uttar Pradesh, Uttarakhand, Gujarat, West Bengal, Haryana, and Rajasthan were identified as *C. oxystoma, C. clavipalpis, C. actoni, C. anophelis, C. orientalis*, *C. similis* and *C. imicola.*

of Vanaraja chicken during first 6 weeks of age was estimated to be 2,600 kcal/kg metabolizable energy, 20% crude protein, 1% lysine, 0.4% methionine, 0.8% Ca and 0.4% non-phytate phosphorus, respectively, for optimum performance.

Protein and energy requirements of Krishibro, a multicoloured broiler, showed that high dietary energy (3,000 kcal ME/kg in starter diets and 3,100 kcal ME/kg in finisher diets)) and low dietary protein levels (20% in starter and 18% in finisher phases) could support optimum performance and immune response. **Feed deprivation** up to 48 hr post-hatch reduced the yolk-sac utilization, gastrointestinal tract development, body weight gain, dressed weight and breast meat yield in broiler chickens. Feed deprivation for initial 24 hr after hatch had no adverse effect on performance of broiler chickens. Feeding crude gum from rice bran oil was a useful **energy supplement** in broiler diets and could be safely used in commercial broiler chicken diet with some positive effects on performance. **Crude lysolecithin** (LL) improved broiler performance.

Hormonal modulation of egg production: Prolactin levels above the normal physiological ranges in the circulation showed negative effects on egg production and increased the pause days in PB3 birds. Through endocrinological manipulation of prolactin hormone and its secretory hormone vasoactive intestinal peptide (VIP), higher levels of egg production was obtained in birds immunized against prolactin or VIP or birds fed bromocriptine by 8.9, 10.98, and 5.38, respectively, in dual purpose birds (PB3). Field testing of the protocol developed for improving egg production indicated that egg production could be improved with available resources under normal feeding practices.

Cloacal gland foam: The effect of surgical removal of cloacal gland showed that this gland and its secretion do not play any role in the fertility of quails. The actual function of this gland needs to be explored.

Rural poultry production: Vanaraja, a dual type chicken is the most popular rural variety and is a cross of two pure coloured lines VML and VFL, improved genetically through selective breeding. In VML, the shank length showed an improvement of 2.81 mm over previous generation. The genetic and phenotypic correlations between shank length (primary trait) and body weight at 6 weeks were high and positive. The 40-week egg production showed an improvement of 1.87 eggs in the present generation over the last generation. In VFL, the age at sexual maturity decreased by 3.2 days and 40 week egg production showed an improvement of 4.18 eggs over previous generation. The shank length and antibody response to SRBC improved, which is desirable for better survival in the harsh environment of rural areas. In addition, genetic improvement and evaluation of a tinted egg female line of **Gramapriya** was undertaken. In S3 generation, age at sexual maturity, body weight at 20 weeks of age and egg weight at 28 weeks of age were 164 days, 1,189 g, 49.2 eggs, respectively.

The Vanaraja bird is popular among rural masses as it is better adapted to harsh conditions of rural areas

SUCCESS STORY

Vanaraja birds in Kashmir Valley

Vanaraja chicks, 5-week-old were distributed to farmers in Kashmir valley. The performance of birds under backyard condition was highly satisfactory. Body weight of birds at 12 and 27 weeks of age was 726 and 2,850 g respectively. The hens matured between 177 and 182 days of age and the mortality between 5 and 11 weeks of age ranged from 3 to 7%. The Vanaraja chicken was very well accepted in the valley. Vanaraja chicken, which is being successfully reared in most parts of the country was also found suitable for temperate climate of Kashmir valley.

A proud Kashmiri woman farmer with a flock of Vanaraja chicken

LIVESTOCK PROTECTION

Cattle

Isolation of *Campylobacter jejuni* from semen and preputial washings of breeding cattle and buffalo bulls as well as vaginal secretions of infertile and repeat breeder cows and buffaloes, and *Arcobacter* (aerotolerant *Campylobacter*) from cattle faeces were attempted. A recombinant 41

Bacteriocin based preparation for treatment of bovine mastitis

Attempts were made to develop a formulation containing natural food-grade antimicrobials of lactic acid bacteria, i.e. bacteriocins for the treatment of mastitis in lactating animals. Biochemical characterization of the pathogens revealed the predominance of coagulase positive *Staphylococcus aureus* (35%), followed by *Streptococcus agalactiae* (12%), coagulase negative staphylococci (7%), *S. dysgalactiae* (3%), *S. uberis* (4%), *B. cereus* (5%), Gram-negative organisms such as *E. coli* (11%), *Ent. aerogenes* (4%), *Klebsiella* sp. (4%), citrobacter (1%), pseudomonas (4%), proteus (1%) and yeast (1%). Incidence of mastitis was highest in the crossbred cows (57%), followed by indigenous cows (27%) and least in buffaloes (17%). Antibiogram of selected gram-positive pathogens revealed that 95% of the isolates were MAR (multiple antibiotic resistant) variants.

Among the 53 bacteriocinogenic strains, 11 had broad spectrum of activity against several spoilage and pathogenic organisms including mastitis pathogens. The bacteriocin of potent *Lactococcus lactis* M386 was optimally produced. The bacteriocin was extremely thermostable (100°C/60 min) and active over a wide pH range (1–12).

Bacteriocin based preparation was quite effective against both gram-positive and gram-negative pathogens under *in vitro* and *in vivo* conditions. *In vivo* application of formulation through intra-mammary infusions for 6 consecutive days in clinical mastitis cases resulted in cure rate of 66.6%. The formulation was well tolerated in udder and was non-irritating to animals. It was stable for more than 3 months at refrigeration temperature.

kDa OMP protein of *Leptospira interrogans* was identified as an important antigen for serodiagnosis of leptospirosis in animals. Aqueous extract of *Phyllanthus emblica* showed *in vitro* antimicrobial activity against *Staphylococcus aureus* and *Streptococcus agalactiae* isolated from clinical cases of mastitis.

Buffalo

Bacteriological examination of fecal samples from neonatal buffalo calves led to isolation of *Escherichia coli* belonging to serogroups, viz. O139, O37, O9, O106 and O143, while parasitological screening of samples was suggestive of coccidiosis with single/mixed infection with *Eimeria* spp.

Ultrasonography of mastitis affected udder indicated fibrosis, accumulation of particulate matter, calcification and abscess in lactiferous ducts, gland cistern, glandular parenchyma and teat cistern. Teat cistern was almost obliterated with fibrosis and teat wall thickness also increased in comparison to normal teat of the same animal.

Goat

A saponified sonicated vaccine against caprine pleuropneumonia proved safe and conferred immunity in goats. Enrofloxacin could effectively be used in combination with nimesulide for treating

SUCCESS STORY

Goat-pox vaccine

Research for development of a live attenuated goat-pox vaccine was initiated in 2001. Initially, a controlled experimental trial was carried out in experimental goats to determine the virus safety, potency and protection against virulent virus challenge. The experimental vaccine provided complete protection against high dose of virulent challenge virus. It produced no adverse reaction even at 10^5 $TCID_{50}$, while it conferred protection at a dose as low as 10^1 $TCID_{50}$. Upon vaccination, goats react initially by local hyperemia at the site of inoculation with a marginal rise in temperature from 5–7 days post vaccination. These reactions are generally transient.

The protection was confirmed by virulent challenge and by determining of humoral immune response. The longevity of protection was for more than 2 years, the maximum period studied so for. Preliminary studies carried out in pregnant goats that were in different stages of gestation showed that the vaccine is safe.

The goat-pox vaccine has been widely used across the country with no adverse reports

The experimental goat-pox vaccine has passed all in-house quality control studies. A large-scale field validation of the vaccine has been going on for the past 24 months. Till date, more than a lakh doses of vaccine have been used across the country with no adverse reports. The performance of the vaccine has been satisfactory as evident from the growing demand for supply of the vaccine and also the feedback received from the end-users.

Gastrointestinal parasitism

Epizootiological studies were conducted in different agro-climatic zones of country. In Madhya Pradesh *Haemonchus* was the most predominant (44.4%) species followed by *Oesophagostomum* spp. (22.4%) and *Cooperia* spp. (6%). Amphistome infection was more common than *Fasciola gigantica* and *Schistosoma indicum*. In West Bengal strongyles constituted the major nematode of which haemonchus was the predominant species in all the animal species surveyed. In Uttarakhand, prevalence of infection was higher in sheep and goats compared to cattle and buffaloes. In Tamil Nadu, overall prevalence of 47.45% of helminth infection was recorded. In Rajasthan bioclimatograph was prepared for semi-arid and arid zones of Rajasthan for future prediction of haemonchosis and trichostrongylosis in farms. Forecasting system FROGIN was evaluated for performance in semi-arid and arid zones. In Sikkim, goats were highly susceptible to G–I helminths with 62% prevalence as compared to yak and cattle having 22 and 25% prevalence respectively. Higher helminthic infection was recorded during autumn (September–November) with 78.57% infection in goats and 33.33% in cattle, whereas, in yaks it was 25.75% in summer. Least infection could be recorded during winter. In Sikkim, *H. contortus* was predominant followed by *Oesophagostomum* spp, *Bunostomum* spp and *Nematodirus* spp. These infections were prevalent in yaks also.

ES antigen of *H. contortus* was purified by immunoaffinity chromatography. In the bound fraction 26, 32, 60 and 120 kDa polypeptides were fractionated by SDS-PAGE; and 120 kDa polypeptide was recognized as early as 4 day PI, showing promise for diagnosis of preclinical haemonchosis in western blotting. In 60 and 120 kDa polypeptide L3 larvae, were also recognized in western blotting by 4-day preclinical sera of sheep. In dot-ELISA affinity purified fraction of ES antigen could detect anti-*H. contortus* antibodies in 4-day experimental sera of sheep. In western blotting 120 kDa polypeptide of ES antigen did not show cross-antigenicity to *Oesophagostomum columbianum, Paramphistomum epiclitum* and *Fasciola gigantica*. Revalidation of dot-ELISA based diagnostic kit for *Oesophagostomum* and *Bunostomum* was initiated in goats, cattle, yaks and mithuns. In a sequel of immunoprophylactic studies H-gal-GP antigen of *H. contortus* has given promising results. Egg count reduction was 54.15% and worm count reduction 92.15% for female worms and 87.1% for male worms with absence of lesions in immunized animals. H-gal-GP antigen was purified through peanut lectin agrose column chromatography. Molecular biology technique for detection of benzimidazole resistance allele-specific PCR was standardized. Application of the technique has revealed higher proportion of homozygous resistant larvae in farmer's flock as compared to farm flock. Selected isolates of nematophagous fungi *Arthrobotyrus oligospora* from field flocks from arid and semi-arid zones reduced the number of GI nematodes. Attempts to get wild isolates of hypomycete on trematode egg from organic environment of Durg, Chhattisgarh, were successful.

susceptible bacterial infections associated with inflammatory disease conditions in goats.

Surgical and clinical interventions: Techniques for application of acrylic external skeletal fixators for management of compound fractures of small animals and circular external skeletal fixation for large animals were standardized.

Technique for dissolution of cystic and urethral calculi was developed using oral and (or) cystic irrigation of ammonium chloride or hemiacidrin in goats and buffalo calves suffering from obstructive urolithiasis/uraemia. Calcium carbonate (80%) and potassium iodide (15%) were cost-effective alternative radiographic contrast to barium sulphate to delineate stomach and large intestine and gastrointestinal and urinary tract in small animals.

Diagnostics: *Toxoplasma gondii* SAG1 gene was cloned and expressed in eukaryotic system, and the recombinant protein was successfully tested with good immunoreactivity for detection of antibodies in goats.

Field validation of cathepsin-L cysteine proteinase of *Fasciola gigantica* confirmed that this protein was specific with no cross-reactivity with *Paramphistomum epiclitum*, *Gastrothylax crumenifer*, *Gigantocotyle explanatum*, hydatid (metacestode) and *Strongyloides stercoralis*. The ELISA sensitivity recorded with this protein was 95%.

The Bm86 homologue of *Hyalomma anatolicum anatolicum* was cloned and expressed in both prokaryotic and eukaryotic systems.

Sheep

A sterile and safe inactivated pentavalent vaccine against bluetongue in sheep was developed. The combined HS+FMD vaccine passed the safety tests with no adverse reactions. The protective titre 1.5 (SN titre) indicated the need for boosting of the vaccine every 6 months. An indirect immuno-peroxidase monolayer assay (IPMA) was developed for detecting pestivirus antigen and neutralizing antibodies in small ruminants.

Pig

A PK-15 adapted cell culture swine fever virus vaccine developed at sixth passage having $10^5 TCID_{50}$ titre was found potent in pigs. Twenty-one days vaccinated pigs were protected against

Diagnostic kits developed by PD_ADMAS

virulent challenge infection. PD50 of this vaccine contained ≥ 200 PD50/dose.

RT-PCR was optimized to detect classical swine fever virus (CSFV) in 1:10,000 diluted infected cell culture fluids and to screen clinical samples.

Equines

Surveillance and monitoring of equine diseases: Unique programme of nation-wide active equine disease surveillance facilitated poor equine owners in saving the precious indigenous equine germplasm through timely diagnosis and management of diseases. An outbreak of glanders was reported during July-August 2006 in Maharashtra. Subsequently, cases of glanders were also reported from Punjab, Uttar Pradesh and Uttarakhand. Nation-wide testing on 4,395 serum samples revealed 97 samples to be positive for glanders during the year. The disease was confirmed to be glanders serologically and subsequently by isolation of the causative agent, *Burkholderia mallei* from the suspected cases. The follow-up surveillance of the area revealed no new cases from Maharashtra, indicating control of the outbreak in that state. It is being monitored continuously to assess the situation in the entire country, with particular emphasis on the equines of Uttar Pradesh and Uttarakhand. Equine infectious anaemia, African horse sickness, equine influenza and *Salmonella* Abortus equi, were not detected in the samples. Equine rotavirus strains exhibited G10, G3, G6 and G1 type specificities that accounted for 19.0, 42.9, 14.3 and 9.5% of the isolates, respectively.

Diagnostics: Haemagglutination inhibition (HAI) assay was standardized for serodiagnosis of Japanese encephalitis (JE). On testing equine serum samples, from different states, for antibodies to JE serocomplex by HAI, 14.32% samples were detected positive. An RT-PCR was also developed for detecting the virus in infected tissues. A recombinant protein-based ELISA was standardized for differentiation of EHV-1 and EHV-4. A recombinant-protein based ELISA was developed for detection of *Babesia equi* antibodies in equine sera. This assay could detect *B. equi* antibodies in equines as early as 6 days post-infection till 90 days of observation period.

Validation of EHV-1 vaccine: An equine herpes virus-1 (EHV-1) killed vaccine incorporating indigenous strain of EHV-1 was evaluated in experimental pregnant indigenous mares. Challenge studies did not reveal any fever or virus shedding. Three of the vaccinated mares gave birth to healthy foals and only one mare aborted due to EHV-1 infection after challenge. Hyperthermia and virus shedding was observed in non-vaccinated mares after challenge. On challenge, one non-vaccinated mare aborted due to EHV-1, while one gave birth to a weak foal that died within 24 hr of birth. Virus could be isolated from the lungs and liver tissues of both the foals. The vaccine provided satisfactory level of protection against EHV-1 associated abortion but before commercialization further immune response studies need to be undertaken in selected organized equine farms.

Equine health camps and farmer meets (*Ashwa Palak Goshties*)

The equine owners were enlightened on various aspects of disease control and management. In addition to the treatment of major equine ailments in these camps, deworming and tetanus vaccination were done in equines. Feedback from farmers was obtained for further research and development in equine health and production.

Camel

To popularize the knowledge of our age-old scholarly system of medicine against one of the most common skin disease sarcopticosis in camel, an indigenous medicine, M-Cure has been developed, evaluated and made available for use at the NRCC. Five to seven local applications are required to cure the disease without any sign of relapse.

In camels intra-mammary infections cause health problems of diarrhoea etc. in suckling calves. *Staphylococcus*, Streptococcus, *Corynebacterium* and *Bacillus* were the commonly encountered intramammary infections in camels. Characterization of 55 isolates of staphylococci revealed that *Staphylococcus aureus, Staph. hyicus, Staph. intermedius, Staph. haemolyticus, Staph. auricularis, Staph. sciuri, Staph. hominis, Staph. epidermidis, Staph. capitis, Staph. warneri* were associated with camels. Mean pH of quarter milk samples was within the normal range in all the non-clinically infected quarters whereas in clinically infected quarters there was a significant rise in mean pH. Increase in sodium concentration in milk of clinically infected quarters compared to

milk of healthy quarters showed increased permeability of sodium due to inflammation of the secretary tissue. There was significant variation in mean Co concentrations among negative, sub clinical, non-specific and clinical groups. However Cu varied significantly with somatic cell count of milk. Daily feeding of Cu, Zn and Se for 30 days resulted in almost 40% lower infections.

Mithun

The overall seroprevalence of antibodies to BTV in mithun was 86%. Highest seroprevalence was observed in mithuns above 4 years of age and the lowest in 1–2-year-old. The likelihood of BTV infection was almost six-times more in mithuns in the age group of 2–4 years than that of > 4 years. Seroprevalence was higher in mithuns from free-ranging system in comparison to mithuns kept in semi-intensive system of management. Likewise mithuns from free-ranging system were almost seven -times more seropositive to BTV in comparison to mithuns kept in semi-intensive system of management.

The overall prevalence of antibodies to *Neospora caninum* in mithun was 10%. Highest seroprevalence was in mithuns above 3 years of age and lowest in mithuns of 2- to 12-month-old. No statistically significant difference was observed in seroprevalence between males and females. An overall seroprevalence of antibodies to *A. marginale* in mithuns was 63% and it increased with the increase in age of animals. In both the cases the seroprevalence was higher in mithuns in free-ranging condition in comparison to mithuns kept in semi-intensive system.

The distribution of *Escherichia coli* serogroups isolated from faecal samples of mithun calves with diarrhoea revealed 72 *E. coli* strains belonging to 38 different serogroups (based on "O" antigen). Of the 72 strains isolated, 10 were not typeable and four were rough. The virulence gene profile of different *E. coli* serogroup **(**isolated from faecal samples of mithun calves with diarrhoea) was studied — *stx1* genotype was prevalent in serogroups O4 (4 strains) and O32 (3 strains); *stx2* genotype in serogroups O55 (2 strains), O2 (2 strains) and O49 (2 strains); and *hlyA* genotype in serogroup O55, O2 and O49 (2 strains in each serogroup).

Yak

Epidemiology: Infectious diseases like foot-and-mouth disease, brucellosis, infectious bovine rhinotracheitis, haemorrhagic septicaemia, tuberculosis, babesiosis and non-infectious diseases (like keratoconjunctivitis, diarrhoea, tympany) were prevalent in different yak tracts. *Staphylococci*, *Pseudomonas aeruginosa* and *Escherichia coli*, *Candida albicans* were found associated with the chronic haemorrhagic diarrhoea in yaks with prolonged antibiotic therapy. *Mannheimia haemolytica* was found in yaks for the first time in respiratory complications. Survey at Nyukmadung revealed that 19% of the yaks (27 out of 140) bear this opportunistic organism in respiratory tract. *Haemophilus somus* and *P. aeruginosa* were the common associates with this pathogen. Most of the isolates of these pathogens were muti-drug resistant. Survey revealed that 12.42% yaks were positive reactors. Incidence of gastrointestinal disorders in yaks was highly correlated with rainfall and temperature. A fast and reliable PCR was standardized for molecular diagnosis of *Babesia bigemina* in yak and its hybrids.

FMD: The antibody titre level in yaks following application of trivalent oil adjuvant FMD vaccine, sharply increased to the protective titre within one month but tended to fall from third month onwards. Protective antibody level was present up to 90 days of vaccination only. However, following booster administration, protective antibody response was rejuvenated within a short period of time.

***Senecio* induced oxidative injury in murine macrophage cell line J774:** *Senecio* poisoning in yaks is very common in Arunachal Pradesh and other

Haemorrhagic septicaemia

A low volume saponified HS vaccine was prepared and tested in dairy animals and 100% protection was observed on direct challenge at one year duration. The vaccine was also inoculated in animals at Izatnagar, Himachal Pradesh, Tamil Nadu and Orissa. No untoward reactions or any clinical signs were observed in any vaccinated animal. Apart from that, a vaccine trial of combined HS and FMD vaccine was also carried out and a booster dose was given at 6 months post vaccination. At 7 months challenge, 100, 75 and 75% protection were observed, respectively, against FMD virus type A, O and Asia 1.

Major outer membrane proteins (OMPs), OmpA and OmpH, were identified and characterized in *Pasteuralla multocida* serotype B:2 that originated from sheep, goats, cattle and buffaloes. The tbpA gene of *P. multocida* serotypes D and F were cloned and sequenced. The nucleotide amino acid sequences of *P. multocida* D:1 showed 98.5 and 96.9% homology, whereas F:3 serotype showed 97.7 and 95.2% similarities with tbpA of *P. multocida* serotype A. Apart from that 77 isolates of *P. multocida* from various animal species and birds were identified and characterized using morphological, cultural and molecular techniques. A patent for low volume saponified haemorrhagic septicaemia (HS) vaccine was submitted.

sub-Himalayan areas of India. The plant contains seneciophylline that induces severe hepato-toxicity. Ethanol extract of *Senecio* increased severe oxidative burden in murine macrophage cell line J774. Following incubation with *Senecio* DNA laddering and single stranded DNA breakage were evident.

Herbs: Indigenous medicinal plants were collected from the West Kameng and Tawang districts of Arunachal Pradesh especially in the high altitude areas of Indo-Tibetan borders. The indigenous knowledge of the local tribal people was taken into account in many cases for collection of the plants. The anti-bacterial, anti-fungal, anti-inflammatory and wound healing properties of *Saussurea costus* and *Rubus idaeus* were found out through experimental trial.

Wild life

A bicistronic DNA vaccine was developed to induce virus neutralizing antibody response against rabies and canine parvovirus in dogs. A recombinant RNA replicon – based vaccine vector with immunogenic gene from canine parvovirus was constructed.

Local application of combination of oils of *Jatropha curcas* + *Phyllanthus emblica* + *Commiphora mukul* was highly effective in mitigating immuno suppression and oxidative stress in canine demodicosis.

Poultry

A PCR was developed and applied for detection and differentiation of avian tumours and determining the prevalence of MD and ALVs in various flocks. The results were confirmed by using real time PCR, which proved that the PCR is a useful technique for detection and differentiation of tumours caused by multiple viral infections such as MDV and ALVs. The incidence of MD detected in pureline layers was 7.5, 4.1, 2.6 and 3.7% in IWK, Anand, IWH and IWI respectively. Sporadic cases of MD were recorded in PB-2, Dahlem Red and naked neck populations.

The inactivated chicken infectious anaemia virus (CIAV) vaccine protected the SPF chicks against CIAV when vaccinated at day-old age, whereas layer *Gallus* chicken vaccinated at 18 weeks of age showed induction of antibodies up to 20 weeks post-vaccination and transferred protective maternal antibodies to their progeny. The chicken embryo origin turkey pox vaccine provided 90–95% protection under field trial, whereas cell culture origin turkey pox vaccine provided 72–72% protection. The formalin inactivated Newcastle disease (ND) virus (field isolate), adjuvanted and emulsified induced humoral and cell-mediated immunity, which was comparable to that induced by inactivated commercial ND vaccine. The experimental vaccine afforded protection up to 66.6% against challenge infection.

Quality control and production of Veterinary biologicals

RD 'F' strain vaccine, lapinized swine fever vaccine, tissue culture sheep pox vaccine, HS oil adjuvant vaccine, *Brucella abortus* strain-19 (live) vaccine, enterotoxaemia vaccine, Johnin PPD, Mallein PPD, *Brucella* agglutination test antigen, *Brucella abortus* Bang Ring antigen, Rose Bengal Plate Test antigen, *Brucella* positive serum, *Salmonella* Abortus-equi 'H' antigen, *S.* Pullorum plain antigen, *S.* Pullorum coloured antigen, *S.* Pullorum positive serum, and *Salmonella* poly 'O' sera were produced and quality tested. PPR vaccine, PPR c-ELISA kits and PPR s-ELISA kits were produced and supplied by Mukteswar campus, as per demand. Monovalent FMD vaccine comprising doses of type 'O', type 'A' and type 'Asia 1' were produced at the IVRI, Bangalore campus.

A rapid and simple recombinant antigen based user-friendly ELISA test kit was developed for serodiagnosis of infectious bursal disease. A sensitive, specific and accurate recombinant haemagglutinin neuraminidase antigen-based ELISA was developed to measure the specific antibody in sera of chickens against Newcastle disease virus.

Six strains of *B. stearothermophilus* were used for antibiotic disc assay. In all, 17 antibiotics from different groups were used for disc assay. Pencillin G, ampicillin and amoxicillin depicted sensitivity at MRL dose of 0.004 µg/ml. Cloxacillin, cefalexin, tetracycline, doxycyclin, gentamycin, streptomycin and lincomycin exhibited sensitivity close to MRL concentrations, i.e 0.5, 0.5, 1.0, 10.0, 0.1, 1.0,10.0 and 1.0 µg/ml, respectively.

Animal disease monitoring and surveillance

The databank was developed on animal disease trends, disease prevalence, meteorological data, land use data, animal and human demography, soil pattern data and crop production data. Integrating these factors an interactive website—National Animal Disease Referral Expert System (NADRES) — was developed, which can predict the probable occurrence of 10 major livestock diseases in any particular district of the country. This is a user friendly programme for general public, and can be accessed through internet (www.nadres.org).

An offline version ***India.AdmasEpitrak*** was also developed for collaborating units of PD_ADMAS. This programme provides data on ecopathozones, disease prevalence studies and

frequency of disease occurrence. It helps to estimate the spatial and temporal specific disease prevalence profiles of economically important diseases.

Latex based agglutination test for detection of *Brucella* antibodies in cattle, sheep and goat was standardized using heat killed sLPS antigen. An indirect ELISA protocol for *Brucella* screening in small ruminants was standardized. Seropositivity for *Brucella* antibodies in sheep and goat was found highest by RBPT (9.95%) followed by iELISA (7.36%) and least in STAT (5.67%). The PCR protocol to detect *B. abortus* and *B. melitensis* from the aborted material, foetal contents and vaginal discharges, was standardized. It is advantageous over serological tests, and avoids risk of laboratory-acquired infections. This protocol can be used to screen suspected herds for confirmatory diagnosis. Leptospiral abortions were scarcely reported and no systematic study has been carried out in our country. Clinical cases from various species of livestock and human samples were screened for the leptospira organisms, and in 119 cases out of 1,120 samples the organism could be isolated. The advantage of this study is that it would be possible to establish not only leptospiral etiology, but also *Brucella*, IBR and other infectious causes.

A study on molecular epidemiology of BHV-1 was initiated as the long term serological studies have shown serological evidence of the disease (35–40% sero-positive) all over the country, although there are limited reports of isolation of virus from Karnataka, Punjab and Uttar Pradesh. BHV-1 infection in bulls is epidemiologically important as the virus excreted intermittently in the semen may spread infection to cattle receiving semen from them. Identification and elimination of the bulls positive for BHV-1 antibodies and genome sequences by serological tests and PCR assays, respectively, would help in formulating the control strategies. Hence, the Directorate has taken up detection of BHV-1 genome/antigen in the semen samples of all bulls in the breeding farms on regular basis. Semen samples from bull mother centers were tested for the presence of the BHV-1 antigen/genome and all were found negative.Genome sequencing of IBR-1-ADMAS isolate was compared with those of reference sequences (one from Switzerland and two from Brazil). There was 100% homology with the sequence of AJ 004801 (Switzerland), 98.3% with AY 58382, and 98.7% with AY 330349 (both from Brazil). In clustering, the Indian isolate was closer to the sequence from Switzerland.

A survey showed the apparent prevalence of *Neospora caninum*—a cyst forming coccidian parasite—infection in dairy cattle which is now recognized as a major cause of abortion and economic losses to producers worldwide. Screening of sera samples from apparently healthy goats from different geographic zones of India suggested an overall countrywide seroprevalence of 7.93%.The carrier animals of trypanosomiasis become nuclei for its propagation in a particular area, and its detection is very important to control and eradicate the disease. The PCR technique was standardized and its field validation is in progress.Serum Bank facility, **the first of its kind in India** having more than 170,000 serum samples from all over the country was developed, and is being used for long-term national surveys on IBR, brucellosis, yersiniosis, RP, PPR,BT and other diseases. **Diagnostic kits for bovine brucellosis** were developed and standardized, and the state-of-the-art software based Avidin-biotin enzyme linked immunosorbent assay (Ab-ELISA) for bovine brucellosis can be used on serum samples and milk samples containing too little cream, colostrum, clotted milk or frozen milk.

Computer interface based BHV-1 whole antigen Ab-ELISA was developed as per the standards of IAEA, and standardized and validated.

CAPTURE FISHERIES

Marine fisheries

Marine fish landings and catch structure: Analysis of the marine fish landings in India during the year 2006 indicated an increase in total fish landings and a change in catch structure. The total marine fish catch, estimated at 2.71 million tonnes,

Successful pen culture in Bihar

Pen culture of fish in pens was demonstrated in the *maun*s (floodplain lakes) in Bihar for augmenting fish production and creating opportunities for employment and income generation. Pens of 0.1 ha size, installed in 3 *maun*s, viz. Kaithkola, Rajoura and Bahuara, were stocked with fish @ 15,000–20,000/ha. The grass carp *Ctenopharyngodon idella* recorded the highest survival rate followed by *Catla catla*. Overall survival rate ranged between 40 and 65% at an average of 50%. Highest fish growth both in terms of length and weight was recorded in Bahuara followed by Kaithkola and Rajoura. Among the fish species stocked, average maximum growth in length was recorded for *C. mrigala* (72%) and in weight for *C. catla* (567%). The maximum fish harvest was from the Bahuara (5,047 kg/ha). With a total cost of inputs of Rs 164,080, the gross and net returns per pen area were estimated at Rs 227,510 and net at Rs 63,430/ha respectively. The average B:C ratio for the *maun*s was 1.39. The demonstrations motivated the local communities to adopt pen culture technology.

registered an increase of about 4.1 lakh tonnes compared to the previous year. Pelagic finfishes constituted the majority (55%), followed by demersal fishes (24%), crustaceans (16%) and molluscs (5%). While mechanized sector accounted for 71% of marine fish landings in the country, motorized and artisanal landings contributed 24 and 5% respectively. Oil sardine, ribbon fishes, lesser sardines, cephalopods, seer fishes and croakers recorded increase, while there was a marginal decrease in the landings of Bombay duck and penaeid prawns. West coast accounted for more than 68% of marine fish landings. While the south-west region comprising Kerala, Karnataka and Goa coasts recorded 34.6%, the north-west region comprising Maharashtra and Gujarat coasts accounted for 33.5% of the national marine fish production. In the east coast, the south-east region consisting of Andhra Pradesh, Tamil Nadu and Pondicherry coasts contributed 21.8%, while the north-east region, comprising the West Bengal and Orissa coasts contributed 10% to the total marine catches.

Impact of climate change on marine fisheries: Oil sardine, the most abundant marine fish in India, showed signs of adaptation to climate change as per the interim findings of the National Network Project on Impact, Adaptation and Vulnerability of Indian Fisheries to Climate Change.

Inland fisheries

Changing catch structure in Ganga fisheries: Decline in catches and a shift in the species spectrum, observed in 3 different stretches of the river Ganga between Deoprayag and Farakka, were attributable to deteriorating fish habitats. Small and economically unimportant fish species dominated upper (Rishikesh to Haridwar), middle (Kannauj to Varanasi) and lower (Varanasi to Farakka) Ganga contributing 42–100% to the total catch. In the upper stretch, the contribution of major carps was negligible, but a good fishery of *Tor* spp. was recorded below Haridwar. In the middle stretch (Kannauj - Varanasi) contribution of major carps ranged from 7.5 to 15.5%, while in the lower stretch landing of major carps *Labeo rohita*, *Catla catla*, *Cirrhinus mrigala*, was still lower, with the exception of Farakka, where these fishes formed up to 14.1% of the catch. Presence of exotic species *Cyprinus carpio* in the middle stretch (maximum recorded at Allahabad) suggested an impact on natural food web and ecological balance in the river ecosystem. Decline of major carps in the middle stretch is attributable primarily to man-made environmental changes and the resultant loss of fish habitats for breeding and feeding. Based on these findings, necessary packages for restoration of habitat and fishery are being formulated.

Impact of regulated river flows on the estuarine fisheries: A study was undertaken at Devi estuary in Orissa to assess the impact of Narauj barrage on Devi river, 70 km upstream of its confluence with the Bay of Bengal. At present, 4–5% of the total regulated river discharge at the barrage is released during the lean season, which was reasonably adequate to sustain the biotic communities. The salinity amplitude in the estuary was in the range of traces to >18.0 ppt and tidal regimes comprised marine (12%), gradient (55%) and freshwater (23%), which were favorable for fish productivity. During monsoons, estuarine fish fauna was dominated by freshwater forms like Indian major carps, minor carps and large catfishes, which migrated downstream of the estuary for breeding and nursing. During post-monsoon, as salinity increased, brackishwater fishes such as mullets, sciaenid, perches, threadfins, anchovies and shellfishes moved upwards in the estuary. In commercial landings of Devi estuary, 97 species of fishes were identified. The freshwater fishes contributed significantly (> 20%) to the fisheries of estuarine zone.

In contrast, drastic control of river water discharge in upstream of the river Krishna negatively impacted the estuarine salinity regime turning it into hypersaline. As a result, oligohaline and freshwater species such as carps, catfishes, murrels adapted to riverine environment, and featherbacks disappeared from the estuarine zone. This changed salinity regime contributed to domination of mullets mainly represented by *Mugil cephalus* and *Liza parsia*. Results emanating from these two case studies paved the way for setting the key parameters for assessing the environmental flows in rivers and estuaries.

Culture fisheries

Freshwater aquaculture

Cage culture: As part of developing a cage culture technology for the upland lakes,

Cages installed in lake Bhimtal registered an increase in biomass

experiments were conducted in Bhimtal lake, Uttarakhand. Four sets of rectangular cages made of HDPE (3 m × 3 m × 3 m) with 4–15 mm mesh size were installed at Bhimtal lake. Golden mahseer (*Tor putitora*) and snow-trout (*Schizothorax richardsonii*) of varied sizes were stocked in the cages. Fry of golden mahseer, reared in cages registered an increase in biomass of about 1.73 kg/cage in 3 months and the growth was 2.4 kg/cage in fingerlings for the same period. Growth of snow-trout, was slower –- the increase in fry biomass being about 300 g/cage in 3 months; and fingerlings registered a growth of 334 g/cage in 3 months, suggesting the potential of cage culture in upland lakes.

Pen culture: An alternative material to replace split bamboo screens was developed to construct pens in wetlands to raise stocking material. HDPE net material was cost effective and could be used as a substitute for split bamboo screens. Initial estimates indicate that HDPE net material reduces pen construction cost by one-third.

A haul of fish after 136 days rearing in pen

Coastal aquaculture

Organic shrimp farming: Success has been achieved in producing shrimp, *Penaeus monodon* using organic inputs. The organic shrimp farming being developed is based on use of natural nutrients, probiotics and bioremedial measures and avoiding artificial chemical fertilizers and pesticides, chemotherapeutic medicines including antibiotics in the culture process. Yeast (*Sachharomyces cerevisae*)-based organic preparations induced significantly higher growth in tiger shrimp *P. monodon*. These organic extracts have a greater role to play in organic farming when use of chemicals and antibiotics are to be restricted or prohibited. Organic manures including vermicompost were used for fertilization in organic shrimp culture. Under organic shrimp farming, stocking density of 6.5 nos./sq. m. yielded a higher growth rate of quality shrimps of more than 30 g in 110 days of culture with an average production level of 1,305 kg/ha/crop with a feed conversion ratio (FCR) of 0.95. The organically grown shrimps maintained a higher growth rate and health status (including coloration and texture) as compared to the other shrimps during the culture.

Culture of banana shrimp *Fenneropenaeus merguiensis*: Culture practice for the banana shrimp, *Fenneropenaeus merguiensis* was successfully developed through a public-private partnership in Bilimora, Navasari, Gujarat. The shrimps attained on average weight of 22 g in 120 days of culture. The pond was harvested after 166 days with the final average size of shrimp being 26.5 g with 78% survival and a production of 806 kg/ha. Apart from mitigating disease and price risks, diversification in shrimp culture is needed to accelerate the growth of *P. monodon* during winter in Gujarat, where the pond water temperatures falls below 20°C. Successful demonstration of the culture of banana shrimp indicated high potential of this species as an alternative to tiger shrimp especially during winter in Gujarat.

Indigenous shrimp feed technology: A shrimp feed was developed using indigenous ingredients to meet the dietary requirements of tiger shrimp, *Penaeus monodon* and Indian white shrimp, *Fenneropenaeus indicus*. The feed technology has been transferred to two entrepreneurs.

Fish harvest

Copper chrome arsenic (CCA) wood preservative for protection against borers: Painted and FRP sheathed panels accorded total protection against woodborers. Biodeterioration of rubber wood panels treated with CCA to three retentions, viz. 16 kg/m^3, 29 kg/m^3, 42 kg/m^3, dual preservative (16 kg/m^3 CCA followed by

Voluntary adoption of better management practices (BMPs)

Studies conducted to assess the 'adoption gaps' in BMPs of shrimp farming in Andhra Pradesh (AP) and Tamil Nadu (TN) indicated average adoption gaps of 32 and 31% respectively. About 68 and 69% shrimp farmers, respectively, in AP and TN had voluntarily adopted the BMPs. However, BMPs that address environmental and food safety issues like proper site selection, conversion of other land uses, overcrowding of farms and low stocking density were not fully adopted. Though these BMPs are not directly concerned with the productivity, they are essential for the long-term sustainability and marketability of the produce. This result highlighted the need for taking up suitable awareness programmes on BMPs to achieve in long-term sustainability of culture systems, food safety and marketability.

Construction of improved wooden canoes in Asom and Arunachal Pradesh

Improved fishing canoes were designed and fabricated at Guwahati, using locally available low cost wood such as *poma*, mango, *moje* and pine. These canoes were sheathed with fibre reinforced plastic (FRP) to increase the strength and durability. Canoes were handed over to the Department of Fisheries, Asom, which were eventually distributed to the Fishermen Cooperative Societies for use in rivers and *beel*s of Asom. The second set of canoes was handed over to the Department of Fisheries, Arunachal Pradesh, which will be used for fishing in Ganga lake, Itanagar, and Renganadi reservoir in Lower Subansiri district.

150 kg/m^3 creosote) and panels coated with paint and sheathed with FRP (fiberglass reinforced plastic) was studied under the estuarine conditions for 6, 12 and 18 months. On prolonged exposure for 12 to 18 months, the CCA treated panels at lower retention of CCA, showed susceptibility to borer attack especially to *Teredo* spp. Higher retentions of CCA preservative in wood imparted higher degree of protection. Dual treated panels performed equally well as CCA treated panels of 42 kg/m^3 retention. Painted and FRP sheathed panels accorded 100% protection against woodborers. The FRP sheath also ensured that there was no leaching of the constituent chemicals into the aquatic environment.

Development of monoclonal antibody: Production and characterization of monoclonal antibodies (MAbs) to serum immunoglobulins (Ig) from rohu,*Labeo rohita*, was achieved for the first time. Purified r-Ig was used as an antigen to develop MAbs to r-Ig. These MAbs were conjugated with horse radish peroxidase to produce anti-rohu Ig conjugate. This conjugate was used as a powerful tool for monitoring primary and secondary humoral responses by ELISA-based analysis.

Molecular fingerprinting of *Salmonella:* Enterobacterial repetitive intergenic consensus sequence (ERIC) PCR and PCR-ribotyping profile of two most prevalent serotypes in seafood (*Salmonella* Rissen and *Salmonell*a Weltevreden) were carried out to fingerprint the serotypes based on origin. PCR-ribotyping results showed 3 fingerprint pattern in *Salmonella* Rissen (n, 20) whereas, 4 different fingerprint profiles were observed in *Salmonella* Weltevreden (n, 24). ERIC-PCR fingerprinting pattern was more diverse as compared to PCR-ribotype as *S*. Rissen and *S*. Weltevreden serotypes have showed 14 and 16 different fingerprinting patterns respectively. ❑

Aquaculture development in NEH region

Fish culture technology packages was developed for carps and exotic trouts suitable for the northeastern region of the country. In Arunachal Pradesh, about 60% of culturable area is suitable for carp, where Chinese carps and Mahseer can be cultured together in ponds located at 600–1,800 m above sea level. Earthen/RCC ponds can be stocked at a density of 4–5 fish/m² during early March when water temperature is above 15°C. Common carp (25%), grass carp (35%), silver carp (20%), rohu (10%)

Carp farming in a fish pond of NEH region

and/or chocolate mahseer (10%) can be stocked with provisions of supplementary feed @ 2–4% of body weight. Organic fertilizers up to 9,000 kg/ha/year and liming 500–600 kg/ha need to be provided at suitable intervals. This technology has enabled the farmers to fish production of about 0.4–0.8 kg/m² in 8 months. Staggered stocking and harvesting can also be adopted.