

Proceedings of the 13th Annual Review Meeting of Niche Area of Excellence (NAE) Programme

The 13th Annual Review Meeting of Niche Area of Excellence Programme was held on 24th October, 2019 at Conference Facilities, NAS Complex, New Delhi from 9.00 AM onwards under the chairmanship of Dr. T. Mohapatra , Secretary DARE & DG, ICAR. The following experts and ICAR Officials participated:

- Dr R C Agarwal, DDG (Agril. Education)
- Dr. A.K. Singh, DDG (Horticultural Science/ Crop Science)
- Dr S P Ghosh, Former DDG, Horticultural Science
- Dr D Rama Rao, Former ND, NAIP
- Dr. S. Honaappagol, Ex Animal Husbandry Commissioner
- Dr Ch, B Gowda, EX PC, Millets
- Dr. C. V. Rao, Sr. Pri. Sci. NBRI, Lucknow
- Dr. Jyoti Misri, Pri. Sci., Animal Science, ICAR
- Dr G. Venkateshwarlu, ADG (EQR)
- Dr P.S Pandey, ADG (EP & HS)
- Dr M. K. Agnihotri, ADG (HRD)
- Dr Vanita Jain, Pri. Sci., Education Division

Dr Vanita Jain, Pri. Sci. EP&HS welcomed and gave a brief overview of the program. She briefed that Niche Area of Excellence (NAE) is one of the prestigious programmes of the Indian Council of Agricultural Research and was initiated with an overall objective to achieve educational excellence in teaching, research and capacity building in frontier areas of agriculture and its allied disciplines during 2006-07.

Dr PS Pandey, ADG (EP&HS) briefed about the achievements made under the programme by the different centres till date and raised various issues pertaining to Niche Area of Excellence. He underlined the importance of agricultural research, education and capacity building under this component and mentioned that till date 55 projects have been concluded and 16 are ongoing. He stressed that the purpose of the review meeting is not only for asking the deliverables but also to get feedback on constraints faced by the centres. The subject matter experts may look into these issues and clearly suggest action points for continuing, strengthening or changes in the technical programme if required. He appreciated the capacity building being done in specific niche areas and number of students conducting research in these centres, but emphasized that impact of such capacity building programmes needs to be assessed and students need to be tracked. He further told that about 387 publications were published, 170 of which are in journals with NAAS rating 5 and above during 2012-13 to 2018-19. During last two years, around 18 technologies generated by different centres have been transferred and many national and international linkages have been developed by the centres under this programme.

Dr. R C Agarwal, Deputy Director General, Agricultural Education, ICAR, emphasized that this was the programme with a mission and by the time the programme is concluded the centre should be in a position to give a technology/product for the stakeholders. He reiterated the criticality of capacity building as the core mandate for this programme. He informed that now the SMD would identify specific thematic areas and invite proposals in those areas as an approach to solve specific problems. This approach may also reduce the rejection rate. He also stressed upon the need of impact study of the programme as it is operational from 2006.

He further emphasized to check the duplication of the activities of similar nature, while scrutinizing the new project proposals being run under the National Agricultural Science Fund and National Higher Agricultural Education Project.

Dr. T Mohapatra, Secretary, DARE and DG, ICAR in his opening remarks mentioned that Niche Area of Excellence programme is one of the most prestigious programmes of the ICAR for building global competitiveness in agricultural education and research. He appreciated the important accomplishments of some centres. He emphasized that NAE is a brand name signifying recognition of the host institution in the identified areas. He appreciated that some of the centres have managed to attain visibility globally, attracting due recognition and collaboration, resulting in important linkages. He was of the view that since the proposals under this programme are selected through highly competitive process and are reviewed thoroughly the research should be of high class and impact in terms of quality research publications and technology is expected. He also asserted that since programmes under Niche Area of Excellence are supported to create a school for capacity building to address the challenges in frontier areas and therefore the niche and novelty in the project must be specified.

- He asserted the need to assess our own standing internationally and therefore emphasised that data on international rating of publications and their global impact needs to be evaluated.
- Defining indicators to assess the concluded programmes. He emphasized on the need for the quantifiable yardsticks to measure the visibility attained by the centre and publications in NAAS rated journals may be one criterion. Many products haven developed and commercialized however, the analysis of their present status in terms of coverage and volume may add value to the programme.
- He also expressed that action taken report must be presented to monitor the progress with respect to objectives and timeline so that a strategy can be redesigned to bridge up the gap.

During the inaugural session, two CDs of e-Courses developed by NAARM covering video lectures for UG students of veterinary and plant protection for UG (Agriculture) students and a diagnostic kit for detection of pesticide residues in milk developed by NDRI, Karnal under the Niche Area projects were released.

The inaugural session was followed by presentations of ongoing programmes wherein the programme-wise salient achievements for the year 2018-19 and technical programme for the year 2019-20 were presented. Invited experts as resource persons, offered valuable suggestions and inputs and discussed the way forward. The two concurrent sessions were conducted and session in Hall 'A', pertaining to Crop, Horticultural Sciences and Agricultural Education was felicitated by Dr P.S. Pandey, ADG, EPHS and session in Hall 'B' on Animal Sciences was coordinated by Dr. Vanita Jain, Principal Scientist, EP&HS.

HALL A

Crop Sciences, Horticulture Science & Agricultural Education

Eight programmes were presented and discussed.

- 1. Development and validation of markers for novel alleles of candidate genes enhancing yield and low accumulation of arsenic from native rice germplasm**

(BCKV, Mohanpur).The achievements were presented by Dr. Somnath Bhattacharyya, PI (January 2017 to December 2019).

Salient Achievements:

- ✓ Allelic status of ten genes, Gnl1a, Spike1, NOG, DEP1, RCA, VDE1, ABCC1, AP37, PsbS1, ZEP was deciphered in 180 diverse genotypes.
- ✓ SNP derived PCR based co-dominant markers were designed from low arsenic accumulating ABCC1 gene and validated in a 190 RILs as well as in a set of 100 genotypes.
- ✓ Small-grain aromatic native rice germplasm of Bengal accumulates 5-6 times lower than most of the high yielding cultivars.High yielding photoperiod insensitive F6 lines with low accumulating ABCC1 allele selected following MAS.
- ✓ Purnendu, Swarnaprabha and Rudra registered with high NPQ values like Japonica genotypes under high light intensity (1600 μm photons/sq m/sec).
- ✓ IET25701 (BS) and Rudra exhibits quick recovery from photo-inhibition by five minutes during fluctuating light whereas most of the genotypes took more than 40 minutes.

Specific Comments:

- The experts appreciated the progress of the programme
- The experts suggested that the database of gene sequence may be submitted in our repository, ASHOKA

2. Risk assessment of metals and metalloids in water-soil-plant continuum under Basmati growing areas of Northern India. The achievements were presented by Dr.S.P. Datta, PI. (January 2019 to December 2021)

Salient Achievements:

- The program was sanctioned in January 2019.Standardization of protocol for sampling and analysis of soil, plant and water samples.

Specific Comments:

- The experts expressed concern about the use of “Basmati” in title
- The experts suggested to incorporate traceability of the varieties in the programme.

3. Plant source based environmentally safe crop protection and production technologies: development and capacity building: The achievements were presented by Dr Anupama Singh, PI (January 2019 to December 2021)

Salient Achievements:

- ✓ Air dried aerial parts (leaves and fine branches) of *Kalmegh* (*Andrographispeniculata*) and *Kulthi* (*Rhyncosia minima*) were fractionated by sequential extraction with four different solvents across different polarity starting from non- polar to polar solvents (hexane, dichloromethane, methanol and water) yielded 0.67, 1.3, 2.5, 9.3percent extract yields from *kalmegh* and 1.11, 1.46, 5.14 percent in hexane, dichloromethane and methanol from *kulthi*, respectively.
- ✓ Solvent extraction from seed kernel yielded neem oil in the range 19.98-29.7%. Methanolic extraction of azadirachtin from defatted neem seed kernel followed by two stage purification by partitioning and precipitation yielded 0.56 % azadirachtin.

Experimental design to standardize and optimize the extraction protocols of azadirachtin from neem seed kernel by minimizing the use of solvents with the aim to enhance the extraction efficiency, has been finalized.

- ✓ Eighteen isolates of *F. oxysporum* (18) were sub-cultured and assessed for their morphological characteristics. Highly virulent isolate was identified after testing pathogenicities under artificial inoculated conditions on a susceptible variety of tomato (Pusa Rohini) in a polyhouse. TOFU-IHBT has been selected as the most virulent strain, which will be pursued further for bioactivity studies.
- ✓ E-format has been developed as a precursor to the phyto-chemical database, taking tomato, maize and wheat as the target crops and root knot nematodes, mite, whitefly and *Fusarium* spp. as the pests.

Specific Comments:

- Volatile-Non Volatile part need to be care of all three
- Focus should be on comprehensive metabolic profile
- Contact with other national institutes such as Forest Research Institute, Dehradun to get quality plant material

- 4. Development of biochemical and physical processing technology to arrest oxidation of lipids/flavones and enhanced the pearl millet flour shelf life.** The achievements were presented by Dr. Shelly Praveen, PI (January 2019 to December 2021)

Salient achievements:

- ✓ Diverse accessions of pearl millet grown in different agro climatic zones were collected from AICRP on Pearl millet, Jodhpur and Division of Genetics, IARI, New Delhi.
- ✓ Standardization of protocols for rancidity measurement parameters in pearl millet flour.
- ✓ Standardization of protocols for activity assays of enzymes involved in lipid hydrolysis and off odour development in pearl millet flour.

Specific Comments:

- The experts suggested to evaluate the puffed product for rancidity/off-odour
- It was also suggested that brain storming workshop may be organized involving experts from the area viz. past directors of IIMR, personnel from processing industry to refine the technical programme regarding problems of working with pearl millet, value addition etc.

- 5. Nutrient Composition, Value Addition and Commercialization of Lesser Exploited Millets.** The achievements were presented by Dr. Sarojani J. Karakannavar, PI (January 2019 to December 2021)

Salient Achievements:

- ✓ Questionnaires were framed and pretested for documentation of cultivation of millets, for documentation activities of the millet processing units and for utilization of millets.
- ✓ Visited millet processing unit “Krishi Sanskarana Ghataka” in Timmapur village of Haveri district owned by Sri. Manjunath Fakeerappa Baggadde.

Specific Comments

- ✓ A concern was expressed by the ADG about the programmes of the project. A meeting of the site committee is to be organized within three months.
- ✓ The experts suggested that colour sorting of seeds should be done
- ✓ PI should focus on ready to eat products.
- ✓ Work to estimate and increase shelf life of the products may be included in the technical program.

6. Genetic improvement of Kinnow mandarin for fruit quality, biotic and abiotic stress tolerance (PAU, Ludhiana). The achievements were presented by Dr. P.K. Arora, PI (December 2015 to March 2020)

Salient Achievements:

- ✓ A total of 840 'Kinnow' and 'Mukaku Kishu' hybrids developed and identified this year using polymorphic SSR markers: Ci06A05b and CiBE1500 and 273 from previously developed hybrids have been planted in the field; 60 of which were also top grafted on mature Jattikhatti trees for achieving early fruiting.
- ✓ Through flow cytometric analysis, five nucellar Kinnow seedlings were identified with ploidy level higher than the diploids.
- ✓ 160 'Jattikhatti' × 'X-639' hybrids and 120 Jattikhatti' × 'Sour orange' hybrids were developed and identified while 28 rootstock hybrids from different crosses multiplied.
- ✓ Sour orange exhibited higher tolerance to soil salinity than four other tested rootstocks (Rough lemon, Rangpur lime, Carrizo citrange and Cleopatra mandarin). Sour orange also displayed better tolerance to *Phytophthora* than 10 other tested rootstock genotypes [(Karan jambhir, two strains of Rangpur lime, three strains of Rough lemon, Volkamer lemon, Troyer and Carrizo citranges (*Poncirus trifoliata* × *C. sinensis*) and Karna Khatta].
- ✓ Nine thousand three hundred thirty one plants of the PAU Kinnow-1 were multiplied and 4,331 plants distributed among the growers.

Specific Comments:

- The progress of the programme needs to be accelerated.
- Further, experts suggested that the experiment should be done in pots instead of polythene bags. Pot have to be kept inside the polyhouse instead of net house.
- PI should focus on revenue generation too.
- Outcomes must be presented and defined in measurable terms.
- Number of plants produced and distributed is not satisfactory
- The plants developed in 2016 should have been taken to field.
- Screening must be done in the field along with polyhouse.
- For new hybrids multi locational testing under wide range of climatic conditions (Rainfall & Temperature) is must before release. Only duly released varieties/ hybrids to be distributed to the farmers
- Promising rootstocks to be short listed for desirable horticultural traits, multiplied and field tested in *Phytophthora* prone area. Innovative molecular breeding approach in rootstock breeding may be planned, as traditional breeding at inter specific level to achieve *Phytophthora* resistant rootstock has not been very successful
- There is need to review the project further at centre along with representative from

ICAR and site committee

- 7. Crop regulation for increasing productivity of Alphonso mango under climatic condition in Konkan region (Dr. BSKVV, Dapoli).** The achievements were presented by Dr. Dr. B. R. Salvi, PI (December 2015 to March 2020)

Salient Achievements:

- ✓ Application of KNO_3 and GA sprays after crop harvest are helping to induce vegetative flush (24%) in summer. Which ultimately gives early flowering and early harvest thereby getting premium price (Rs. 80/- per Kg compared to Rs. 30/- per Kg).
- ✓ Delayed rains cause emergence of vegetative flush during Oct.-Nov. thereby less flowering and harvest (May) so no price (Rs. 30/-per Kg). Application of plant growth retardant CCC @2500 ppm spray and basing exposure twice during Sept. and Oct. has helped to create stress thereby assured flowering (60%) during first and second flowering.
- ✓ Application of Ortho-phosphoric acid @ 1% as nutrient spray and PBZ @ 1000ppm spray as plant growth retardant has helped to hasten the post monsoon vegetative flush mature early by 14 days and 22% more flowering
- ✓ Under severe winter ($< 13^\circ C$ min. temperature period) conditions, sex ratio is changing (reduction of hermaphrodite flowers) so there was no pollination and fruit set. Spray of Tricontanol @15 ppm and application of fresh fish waste trap in orchard or 5% Jaggery spray or growing of mustard as intercrop in mango orchards are found to be promising to improve pollination and fruit set.
- ✓ Application of three sprays of 1% KNO_3 and 150 L of water/tree has helped to regulate crop load by way of 77% increase in productivity (2.43 t/ha) and also has helped to reduce spongy tissue incidence by 12%.

Specific Comments

- Concern was expressed about inappropriate use of drones and centre was advised to rectify the same.
- The centre must clarify the new developments and measurable outcomes.
- Mild summer (March-May) during fruit development stage reported as the ideal condition for Alphonso in Konkan region. Flower inductions by applying chemical/ growth regulators have been tried for long. Studies are now needed to understand the effects of climate change in vegetative flush linked flowering behaviour in Alphonso Germplasm screening for desirable characteristics and marker assisted breeding may lead to find a sustainable solution

Agricultural Education

- 8. Technology enhanced learning in agricultural education (NAARM, Hyderabad).** The achievements were presented by PI, Dr G R K Murthy, PI (January 2014- March 2020)

Salient Achievements:

- ✓ One digital course on “Veterinary Entomology and Acarology” was launched in September 28, 2018. One more course on “Pests of Crops and their Management is finished during this year
- ✓ MOOC is offered successfully fourth time during September-December 2018
- ✓ E-learning website registered 3 lakhs hits with highest monthly hit rate of 160921
- ✓ Following training/ policy workshops are organized for capacity building and policy support
- ✓ Training workshop “Digital Teaching Techniques, March 14-20,2019 (25 participants)
- ✓ MOOCon “Dynamics of teaching and Learning” November 1-30,2018(1192 participants)
- ✓ The eLearning platform was effectively used for running online activities of the Academy like conduction online evaluation of foundation programs of scientists and university staff FOCARS, FOCFAU programs of institute.

Specific Comments:

- The expert committee appreciated the progress of the programme
- The experts suggested that the e-Cources may be uploded on ICAR web site, announced in VC conferene and could be popularised via Kisan Channel

HALL B

Animal Sciences

Nine programmes were to be reviewed under Animal sciences, however, PI of the programme entitled “**Study of *Clostridium perfringens* and *Dichelobacternodosus* (SKUAST, Kashmir)**”, did not attend the Annual review.

- 9. Spore based sensor for monitoring pesticide residues in milk (NDRI, Karnal).** The programme achievements were presented by PI, Dr Naresh Kumar, PI. (May 2014 to March 2018)

Salient Achievements:

- ✓ The developed technology was refined for its working against more than 50 pesticides belonging to organophosphate, (OP), organochlorine (OC), carbamate, herbicides, fungicides and LODs were found well within MRL limits specified by codex (EU) / FSSAI for different food matrices like milk, cereal based foods, fruit juices, feed, fodder, manure, soil, fermented feed, water, turmeric / chili powder, tea leaves, mustard oil etc. These finding indicates that developed technology has scope for its application to almost all group of food matrix for detection of pesticide residues.
- ✓ **Establishment of pesticide LODs recommended by codex in dairy farm-** 12 New pesticides legally recommended for cattle feed / fodder were evaluated and LODs of 07 pesticides namely benomyl, dinotefuron, thiophanate-methyl, fenpropathrin, flubendiamide, indoxacarb, bitertanol was observed at 0.05 ppm while 5 pesticides namely ethion, chlorantraniliprole, pyraclostrobin, chlorothalonil and thiamethoxam showed LOD at 0.01 ppm complying MRL set by FSSAI / Codex.

- ✓ **Refinement of strip making process and its shelf stability study** -Strip making process was re-designed to achieve better color/differentiation when compared with negative sample and with extended shelf stability upto 8 months at 4 °C in vacuum packed condition.
- ✓ **Refinement of developed technology for its working with feed, fodder, soil, manure and water**-Primary secondary amines (PSA) was amalgamated into extraction protocol for removal of pigments from cattle feed to prevent the interference of pigment in enzyme activity during exposure step. PSA was supplemented with MgSO₄ in 1:2 proportion for effective removal of pigment and to avoid the interference of complex matrix and to reduce the cost of extraction protocol. Pesticide extraction protocol was developed successfully.
- ✓ **Screening of pesticides using optimized extraction protocol in selected dairy farm samples under field conditions**- Feed/food samples (340 Nos.) received from NDRI and other dairy farms which included fodder, dry feed, concentrated mix, fermented feed, soil, water, milk and manure, vegetables, commercial feed, market sample (alsi, gur, black mustard, yellow mustard, channa, channi and sugar molasses) etc. were evaluated using optimized extraction protocol for detection of pesticide residues and incidence of 5.0% was observed. Some entrepreneurs purchased the pesticide kit and tested these commodities for organic certification. Milk samples including raw and pasteurized milk (424 nos.) were also evaluated using strip based sensor and 3.06% were found contaminated with pesticide residues. The technology was transferred through Agrinnovate India Limited to a Delhi based entrepreneur i.e. M/s. Floreecer Services Private Limited for its mass production and distribution of kits in India.
- ✓ **To development the safety guidelines in dairy food chain:** Safety guidelines in terms of preventive measures were formulated for mitigation of pesticides through extension material i.e. technical leaflets, posters at dairy farm during various stages of milk production, processing and their handling under field conditions.
- ✓ **Surveillance data on presence of pesticide residues:** The surveillance data on presence of pesticide residues in milk, cereal based food, fruit juices, food, fodder, soil manure and other food matrix has been carried out extensively and will be shared with FSSAI in setting standards for pesticide keeping in view of Indian context.
- ✓ **Technology commercialization:** The developed technology was licensed to M/s. Floreecer Services Private Limited, New Delhi, through Agrinnovate India Ltd. New Delhi vide dated 01 July 2019 with Non Exclusive license fee of Rs. 5.90 Lakhs+ 2% royalty. Various stakeholders involved in primary production of fruits, vegetables, cereal based foods, milk and other foods have been trained on developed technology and these stakeholders have established mini laboratory in villages to ensure organic certification of their primary produce for pesticide residues under field conditions.
- ✓ **Capacity building programme:** 21 days CAFT course entitled “Rapid biosensors and micro techniques for monitoring contaminants and adulterants in dairy foods” was organized dated 4-24th January 2019. Need based training were conducted for 16 employees of PCDF Lucknow (28thMay upto June 2018) and 19 students from outside university were trained on biosensor based techniques in the area of food safety.

Specific Comments

- The concluded programme was appreciated by the experts
- The experts suggested that workshops/training's must be conducted to sensitize milk federations/farmers
- Guidelines may be developed for dairy farms especially commercial farms for better dairy practices

- Guidelines to be developed for dairy farms Refinement of developed technology to screen for specific adulterants/contaminants in food/ milk indicating quantitative estimations would be better and more specific.

10. Nutrition and gut health; probiotics, prebiotics and phytogetic as functional foods to augment gut health of dogs (IVRI, Izzatnagar). The achievements were presented by Dr. A. K. Pattnaik, PI (May 2014 to Sep 2019)

Salient Achievements:

- ✓ Successful development of three products based on canine-specific probiotic (*L. johnsonii* CPN23) and polyphenols from Jerusalem artichoke- (JAE) and pomegranate peel-extract (PPE) using chitosan-coated alginate based encapsulation. These 03 products (**Probiotic-PPE**, **Probiotic-JAE** and **Probiotic-PPE-JAE**) were tested for shelf-life
- ✓ The shelf-life study of products showed maintenance of appreciable viability even after 24 weeks of storage under deep-fridge (-20°C) in comparison to refrigeration (5 weeks) and room temperature (2 weeks) conditions
- ✓ The results from experiments in rats with chemotherapy-induced mucositis and in healthy dogs revealed that the product **Probiotic-PPE-JAE** could be used as a potential supplement for gut health promotion of dogs with inflammatory conditions of the intestine.
- ✓ Based on the results from the evaluation of the effects of dietary supplementation of JA-derived inulin and JA-derived polyphenolics along with a canine-origin probiotic, *Lactobacillus johnsonii* CPN23, the formulation containing '**Probiotic-JA inulin**' showing greater potential for gut health promotion was developed into a product from using the 'chitosan-coated alginate based encapsulation' technology.
- ✓ Shelf-life study of the product indicated that the viability of the probiotic could be maintained up to 45 days under room temperature and refrigerated conditions.
- ✓ Further evaluation of the product in rats with chemotherapy-induced mucositis and in health dogs confirmed its potential as an adjunct therapy in dogs undergoing chemotherapy for minimizing the adverse effects of the associated mucositis.

Specific Comments:

- The epidemiological data on incidence of canine neoplasms may be provided alongwith the prevalence of gastric neoplasms.
- Clinical trials and validation of the product in dogs as per the mandate.
- The project is already in extension period for carrying out the simple study on probiotics/prebiotics. The studies have so far being conducted on rats as model.
- PI to complete the studies within three-four months time with developed products and their impact on the survivability of the treated dogs with neoplasms. Extension may be considered till march 2020

11. Development of stem cell laden nanomaterial-scaffold for nerve, bone and cartilage tissue regeneration in animals. The achievements were presented by Dr. Sadan Bag, PI (January 2019 to December 2021)

Salient Achievements

- ✓ Isolated porcine mesenchymal stem cells (MSC) from umbilical cord tissue. Characterization is in process.
- ✓ Initiated isolation of caprine mesenchymal stem cells (MSC) from adipose tissue.
- ✓ Work initiated on studying behaviors of canine MSC on 3D scaffold made up of natural polymer.

Specific Comments:

- Comparison of usage of Stem Cell therapy with conventional therapy to be carried out, in terms of economic gains
- Equines to be included in the programme as horses are precious and prized animals

12. Development of alternate models and national standards for quality control of veterinary vaccines & diagnostics. The achievements were presented by Dr Ashok K. Tiwari, PI (March 2018 to March 2022)

Salient Achievements:

- ✓ The PK-15, BHK-21 and MDCK cell lines screened for mycoplasma and BVDV were found free from these extraneous agents
- ✓ Rabies, cell culture classical swine fever (CSF) and canine adenovirus-1 (CAV-1) were titrated in suitable cell line. The viruses had titre of rabies $10^{4.2}$ FFID₅₀ /ml, CSFV- $10^{9.5}$ TCID₅₀ /ml and CAV-1- $10^{6.3}$ TCID₅₀ /ml. CAV-1 and CSF viruses has been made in bulk, freeze-dried and kept at -20°C for further use.
- ✓ The IBD antiserum was raised in chicken using inactivated IBD vaccine and in rabbits and in guinea pigs using purified IBD virus. These anti-sera were titrated and characterized by ELISA and AGPT. The anti IBD chicken serum had AGPT titre of 1:32.
- ✓ The National Reference standard for rabies vaccine was developed by calibrating inactivated rabies vaccine against International Reference standard. The mean relative potency of the inactivated vaccine was found to be 6.09 IU/vial. The calibrated inactivated rabies vaccine will be used as a standard vaccine for potency determination of the inactivated rabies vaccine. Further, in-house rabies vaccine has also been developed and calibration is underway to develop National Reference Standard.
- ✓ Under Human Resource Development, a short term training on '**Sterility Testing of Veterinary Biologicals**' were organized from November 19-29, 2018 wherein 18 participants belonging to nine different biological units, Government institute and private industry were participated.

Specific Comments:

- The progress of the programme was appreciated.
- The centre was suggested to involve third party/agency or accredited institute for quality testing.

13. Centre for Zoonoses (MAFSU, Nagpur). The presentation was by Dr. Sandeep P. Chaudhary, PI (March 2015 to March 2019)

Salient Achievements:

- ✓ The **human epidemic of Scrub Typhus** in Nagpur region in the months of August-Sept., 2019 was successfully diagnosed which prevented further casualties.
- ✓ **Karp strain of *Orientiatsutsugamushias*** a major circulating genotype among rodents of the region has been identified for the first time.
- ✓ The epidemiological investigation of the **Scrub Typhus epidemic** has been studied along with NCDC, New Delhi. The route cause has been identified as the Chigger mites harboured by the field rodents.
- ✓ The cases of **TB in elephants** have been detected.
- ✓ **Molecular epidemiological study** and distribution of *Listeria monocytogenes* isolates was studied by PFGE confirming the 4b as major circulating serotype among animals, vectors and environment.

Specific Comments:

- The programme concluded in March 2019 and its objectives are fulfilled
- The House appreciated the achievements and requested the PI of the project to organize Short term training /workshop for enhanced capacity building in the area of Zoonoses.

14. Study of *Clostridium perfringens* and *Dichelobacter nodosus* (SKUAST, Kashmir). The PI (Dr. S.A. Wani) /Co-PI did not attend the meeting. (August 2015 to March 2020)

Salient Achievements:

- ✓ Whole Genome Sequence of *D. nodosus* serogroup B isolate deposited in GenBank (Acc. No. **NZ_SRJB00000000**). The genome content of serogroup B of *D. nodosus* is 44.38% and a total of 1,272 protein-coding genes, 44 tRNA genes, four ncRNAs, nine pseudogenes and seven rRNAs genes were identified. Genome sequence of *D. nodosus* serogroup B revealed presence of 21 genes which are unique to *D. nodosus*, serogroup B and were not found in the reference strain. The genes comprise mainly of oligopeptide transport system permease protein OppB (TC 3.A.1.5.1), Oligopeptide transport system permease protein OppC (TC 3.A.1.5.1), Type IV pilus biogenesis protein PilO, intracellular septation protein IspA etc.
- ✓ Whole Genome Sequence of *C. perfringens* Type D isolate submitted to GenBank (Acc No. **VFIZ00000000**).
- ✓ A 43 kDa immunogenic protein of *C. perfringens* Type D, identified by MALDI-TOF/MS, has been found to have homology to ABC transporter protein (ATP Binding protein) of *C. perfringens* Type D.
- ✓ Quantitative analysis of *C. perfringens* and *C. difficile* was carried out in chicken with enteritis by Real Time-PCR. The *C. perfringens* count was significantly higher (Log 6.12+0.44) in diseased than in healthy birds (Log 2.29 +0.19)
- ✓ Gene deletion mutants: Primers were designed for the amplification of truncated epsilon (*etx*) toxin gene (757 bp) of *C. perfringens* Type D, lacking 80 amino acids at N-terminal of Domain I with suitable restriction sites for cloning and expression into a suitable expression vector (peT28a). The *etx* gene has been amplified and gel purified.
- ✓ Under continuous surveillance programme, 61 unorganized sheep flocks comprising 2807 sheep were inspected and footrot was recorded in 188 (6.7%) sheep. Serogroup

B of *D. nodosus* continued to be predominant (83.17%) followed by serogroup E (13.8%) and mixed infection of both B & E in 3.7% animals.

- ✓ In organized sector, 6455 sheep were inspected. Footrot was recorded in 1.08% sheep. Serogroup B and E were detected in 92.85 % and 4.76 % animals, respectively, while both the serogroups were found in 2.56% animals.
- ✓ A total of 711 faecal samples from sheep (adult=220, lamb=195) and goats (adult =156, kids=140) were screened from organized and unorganized farms for *C. perfringens* and 458 isolates (sheep= 112, lambs=133, goat= 98, kids= 115) were obtained. Toxinotyping of the isolates by PCR revealed all the isolates as type A, 266 (58%) isolates were also positive for $\beta 2$ gene (cpb2).
- ✓ A total of 120 fish samples (60 *Cyprinus carpio* and 60 *Schizothorax niger*) and 45 water samples at five identified points across Dal lake were analyzed for the presence of *C. perfringens*. *C. perfringens* Type A (confirmed by 16SRNA based PCR) was isolated from 37 samples (21 *Cyprinus carpio* and 16 *Schizothorax niger*). The average CFU of *C. perfringens* per milliliter of water was found to be 1.4×10^{-2} .

15. Development of Production and Protection Technologies for Organic Poultry Farming. The presentation was by Dr. S.K. Sharma, PI (January 2019 to December 2021)

Salient Achievements:

- ✓ Layout and designs for different poultry production housing and rearing units along with analysis unit have been developed and submitted to ICAR for approval.
- ✓ Sowing of organic maize for feeding of birds.

Specific Comments:

- Technical programme needs to be reviewed thoroughly as it appears to be poultry production oriented developmental programme with no research component.
- Technical programme to be submitted for review and the same to be forwarded to the experts
- The Centre is advised to visit large commercial poultry farms
- Benefits of organic maize and its usage vis-a-vis normal conventional maize diet pertaining to economic gains

16. Phage display techniques for production of veterinary immunobiologicals without sacrificing animals. The presentation was by Dr. Sanjay Kapoor, PI (January 2019 to December 2021)

Salient Achievements:

- ✓ Identification and characterization of different antigens of *Pasteurella multocida* (B:2), *Staphylococcus aureus* exotoxins, *Brucella abortus* and Foot and Mouth disease virus to be used in the research was carried out.
- ✓ *Pasteurella multocida* B:2 (Vaccine strain P⁵²) was characterized by conventional PCR using OIE recommended *P. multocida*-specific primer; HS-causing type-B-specific primer and *P. multocida*- Type A specific primer. The strain was found to be positive with *P. multocida*-specific primer and HS-causing type-B-specific primers as exhibited

by product size of 460bp and 620bp respectively(Fig. 1, Annexure I).The strain was found negative with *P. multocida*- Type A specific primer which is desired.

- ✓ Lipopolysaccharide (LPS)of *Pasteurellamultocida* (B:2) was extracted by hot phenol-water method as described by Westphal and Jann (1965), characterized by silver staining (Fig. 2, Annexure I) and was found to be pure. The extracted LPS was stored at -20°C for further panning procedure.
- ✓ In *Staphylococcus aureus*, beta haemolysin has been identified as an antigen of interest. The Clone “3-2 SA-Hlb-pQE” (CL 3-2) containing beta haemolysin gene was characterized by PCR and resolved in a midigel horizontal apparatus by 2% agarose gel electrophoresis and was found to be positive for ~420bp product (Fig 3, Annexure I).
- ✓ The beta haemolysin gene was expressed and found to be ~ 37 kDa product when resolved by SDS-PAGE.
- ✓ PCR was standardized for molecular identification of *Brucella spp.* (bcsp-31 gene) and *Brucellaabortus*(IS 711 gene). The PCR product was resolved in 2% AGE and was found positive for 498 bp and 223 bp product which is specific for *B. abortus* and *Brucella* genus, respectively (Fig. 4, Annexure I).
- ✓ FMD suspected tissue specimens (heart tissue from post-mortem cases, saliva and skin epithelium) collected from affected animals in Haryana were processed by multiplex RT-PCR assay (Giridharan *et al.*, 2005) for detection and typing of FMDV serotypes O, A and Asia-1. All the samples were found positive for FMD virus serotype O as exhibited by PCR product of 249 bp(Fig.5, Annexure I).

Specific Comments:

- Economics / cost benefit ratio to be assessed in terms of input provided and the output
- The Centre must assess the safety and efficacy of the Immunobiologicals to be developed without animal models

17. Antibiotic resistance: Animal-Human interface (March 2018 to March 2022)

Salient Achievements:

- ✓ **Meta-analysis of the data for prevalence of foodborne pathogens:** A total of 611 studies out of 10054 (from online database and offline database) for foodborne pathogen prevalence were obtained from 25 states and three union territories of India after applying inclusion and exclusion criteria for systematic review and meta-analysis. Information of 1,14676 samples (food of animal origin) from 611 study was extracted to know the prevalence of food borne bacterial pathogens/microorganisms in various type of animal origin food (such as meat, dairy products, fish and egg).
- ✓ **Meta-analysis of antimicrobial resistance profile of food-borne microorganisms:** A total of 171 studies out of 1200 (from online database and offline database) for antimicrobial resistance prevalence were obtained from 21 states and one union territories of India after applying inclusion and exclusion criteria for systematic review and meta-analysis. Information on a total of 31,636 study samples was extracted in which 6676 bacterial strains from 14 genera were used for antimicrobial sensitivity testing by disc diffusion method. Analysis indicated high prevalence of antibiotic resistant (79.71%), and multidrug resistant bacteria (83.62%) in food of animal origin.

- ✓ **Occurrence of antibiotic resistance in organisms at animal human interface:** 663 samples comprising pooled farm milk, slurry waste, drinking animal water and human water were analyzed *E.coli* and *Klebsiella pneumoniae*.
- ✓ prevalence of *E. coli* in raw pooled milk samples, animal drinking water and human drinking water samples was 24.8%, 60%, 26.7% and 16.3% respectively.
- ✓ For *K. pneumoniae* overall prevalence of 19.6%, 51%, 20.2% and 5.8% was observed from slurry, raw pooled milk samples, animal drinking water and human drinking water respectively.
- ✓ 32.9 % of *E. coli* and 29.4 % *K. pneumoniae* isolates in slurry samples were resistant to tetracyclines, enrofloxacin and sulfonamide. From milk, 21% of *E. coli* and 6.7% of *K. pneumoniae* isolates were resistant to the three antibiotics.
- ✓ Comparative frequency of resistance genes in positive isolates of *E. coli* was: *tetA*>*tetB*>*tetC*(53.5%>24.7%>0.59);*qnrS*>*qnrB*>*qnrA*(35.2%>28.8%>2.9%);*sulII*>*sulI*>*sulIII*(34.1%>14.1%>0%).
- ✓ Comparative frequency of resistance genes in positive isolates of *K. pneumoniae* was:*tetA*>*tetB*>*tetC*(53.2%>20.1%>2.9);*qnrS*>*qnrB*>*qnrA*(40.3%>26.6%>5.0%),*sulII*>*sulI*>*sulIII*(31.7%>16.6%>0%).
 - **Antibiotic resistance in fisheries:** Out of 45 *Vibrio* spp. isolates from 20 samples of water/shrimp collected from seven villages located in two inland salt affected districts of Punjab, 32 isolates were identified as *V. parahaemolyticus* by PCR. Antimicrobial susceptibility of 20 *Vibrio*(including 15 isolates of *V. parahaemolyticus*)isolates indicated that all the isolates were resistant to ampicillin. Other drugs gentamicin, tetracycline, chloramphenicol, ciprofloxacin, sulfisoxazole and trimethoprim were found highly effective against *Vibrio* isolates.
- ✓ **Antimicrobial Resistance in dairy environment, milk and milk products:** To understand the dynamics of antimicrobial resistance in dairy environment, raw milk (n=50), milk products (Curd etc.) (n=45), collected from local households and bulk tank milk samples (n=10) collected from milk plant; and faecal samples collected from dairy farm were processed for isolation of *E. coli*, *Staphylococcus* spp., *Salmonella* spp., and *Lactobacillus* spp. isolates.
- ✓ A total of 150 tentative *E. coli*, 40 *S. aureus*, 10 *Salmonella* spp. and 60 *Lactobacillus* spp. isolates were identified by biochemical as well as molecular methods. More than 80% *E. coli* isolates from raw milk samples displayed resistance to methicillin. Over 30% isolates showed resistance to amoxycylav, ceftazidime, clindamycin and fusidic acid. Two antibiotics from carbapenem family, imipenem and meropenem showed remarkable antagonistic activity against all the tested isolates.

Specific Comments:

- Meta-analysis studies projected in the meeting should not be projected
- The Centre should work on surveillance of AMR especially in Livestock, poultry and aquaculture with WHO identified pathogens viz., *Staphylococcus*, *E. coli*, *Pseudomonas*, *klebsiella*, *Salmonella* and for Aquaculture *Aeromonas*, *Vibrio* etc.,
- AMR surveillance in environment is also very crucial

Concluding Session

The following were the major suggestions/recommendations:

- Every centre especially, the concluding centres may submit a write up of conclusions, knowledge generation, basic /applied research conducted and technologies generated, benefit to the stakeholders along with success story if any.
- The monitorable indicators to be defined with base line in quantifiable terms.
- Way forward and sustainability of the centre after the conclusion of the programme to be indicated. The centres must ensure to further the mandate of NAE in terms of capacity building
- It was decided that date for the internal review may be communicated to the Council well in advance so that official from Education Division may also be deputed for the same.
- No deviation from objectives and technical programme be allowed by the Internal Review Committee.
- Every centre must do SWOT analysis.
- All the centres must develop a website for increased visibility.
- The funding from the Council must be acknowledged in all publications, technologies, products and patents arising out of these programmes.
- The products registration and patenting need to be taken up through IP&TM Unit of the ICAR.
- The Principal Investigator must state action taken, if any, or his comments on observations of Review Meeting. As well as action taken on the recommendations/suggestions of Internal Review committee suggestion in annual as well as final reports.
- All the centres where NAE has been concluded may submit the printed final report as per the format immediately. Changes /modifications if any, as suggested by experts may be incorporated along with ATR.
- Final report to be presented objective wise and must specify the way forward and sustainability.
- First slide during each review must always be of the suggestions/ATR of the last review meeting as well as comments /ATR of the Internal Review Committee.
- The PIs were advised to keep the Nodal Officer identified by the university in loop regarding progress under NAE.
- It is expected that university should not shift or transfer the PI without the prior permission/information to Agricultural Education Division, ICAR.
- To assess the impact of the programme vis a vis support provided every centre require to submit the following:
 - ✓ NAAS rating of the research publications
 - ✓ Citation of these publication
 - ✓ Viewership of the publications if recently published
 - ✓ Technologies commercialized
 - ✓ Revenue generated
 - ✓ Total number of faculty and other stakeholders trained
 - ✓ Data on how they are presently engaged including students
- All the centres must ensure that any major equipments supported under the programme needs to be shared within the institute/ university and among other institutes as well, thereby generating revenue.
- License for any technology generated under NAE should be non exclusive.

The meeting ended with the vote of thanks.