Pakistan is capable of producing best food products in taste, aroma and yield due to its excellent environmental conditions (such as high variation in soil temperature and microflora, intense sunlight and elongated photoperiod etc.). However, to ensure consumer safety of such high-value food items from food contaminants (such as veterinary drug residues and other related chemical compounds) and also meet requirements of international trade, it is important to develop efficient, reliable and affordable analytical strategies for routine and large-scale food monitoring.

Food Safety Laboratories, NIAB (PAEC) are working on these challenges with specific objectives to:

- Enhance laboratory capabilities for chemical contaminants analysis in food of animal and plant origin complying with international standards using immunochemical and chromatographic analytical techniques.
- Enhance Pakistan’s competitiveness in food export markets through national/international networking.
- Create awareness among technical/non-technical stakeholders involved from “Farm to Fork” to produce safe/quality-assured foods.

Details of these activities are as follows:

**Accreditation of VDR Labs for ISO/IEC 17025:2005:** As part of Food Safety Laboratories at NIAB, Veterinary Drug Residues (VDR) Labs accredited for ISO/IEC 17025 standard on April 04, 2017 from Pakistan National Accreditation Council (PNAC). VDR Labs (PNAC Lab # 119) are the first accredited laboratories in Pakistan, offering accredited tests for seven types of major antibiotic groups and hormone in different food matrices to public and private sectors.

1. **R & D Activities:**

**Monitoring of Gentamicin residues in poultry tissue and bovine kidney in Faisalabad region.**

A highly sensitive Enzyme-Linked Immunosorbent Assay (ELISA) was standardized (0.1, 0.3, 0.9, 2.7, 8.1 ng/mL) for the determination of gentamicin in poultry tissue and beef kidney employing Commercial ELISA test kit (Green Spring Cat. # LSY-10023). Relative absorbance found inversely proportional to the concentration of the analyte. A linear regression was obtained (y = 88.81-10.0x) with correlation ($r^2$) of 0.888. Lowest detection limit (IC$_{20}$) and Middle of the test (IC$_{50}$) were found to be 0.74 ng/mL and 2.2 ng/mL, respectively, which is well below the maximum residue limit for muscle (50 µg/Kg) and beef kidney (750 µg/Kg)*. Test found suitable for gentamicin detection in poultry tissue and beef kidney samples. Poultry tissue and beef kidney (glomeruli) samples (20 each) were collected from local markets of Faisalabad city in an area of 25 Km with 5±1 Km East, West, North and South. Extraction protocols were optimized for tissues and kidney samples.
using phosphate buffer as extracting solvent. After due centrifugation, the supernatant was immunoassayed at lab temperature (25±1°C). From the results, it was observed that 10% (2 samples) of the poultry tissue samples found above the permissible limits. No gentamicin was found in beef kidney samples.

**Surveillance studies for nitrofuran metabolites in fish by using ELISA**

Fish samples (40 in number) were collected from different areas of Faisalabad region. Commercial ELISA kits (Cat. # W81117 and W881119 Quicking Biotech) for AMOZ and AOZ were used with sensitivity of 0.05 and 0.025 ng/mL & detection limit for aquatic product 0.1 ng/mL, respectively. Since AMOZ/AOZ is tissue bound metabolite; series of extraction steps were involved including addition of dilution-derivatization reagent, incubation (for 16 hr 37°C), addition of extraction solvents along with mixing & centrifugation. Supernatant was collected, dried in Romer’s Evap System and residue re-dissolved in n-hexane/washing buffer before finally collecting bottom layer for the assay.

**AMOZ Analysis in fish:** Standard curve was plotted with standards conc. (0, 0.05, 0.1, 0.25, 0.54 & 1.0 ng/mL) and evaluated by calculating inhibition concentrations (IC_{20} and IC_{50}) at 0.064 & 0.34 ng/mL respectively which is well below MRPL (minimum required performance limit) of AMOZ (1 µg/Kg)*. Results indicated that AMOZ residues in all fish samples were found below the MRPL (1.0 ng/mL) and declared negative.

**AOZ Analysis in fish:** Standard curve was plotted with standards conc. (0, 0.025, 0.05, 0.1, 0.2 & 0.5 ng/mL) and evaluated by calculating inhibition concentrations (IC_{20} and IC_{50}) at 0.035 & 0.12 ng/mL respectively which is well below MRPL (minimum required performance limit) of AOZ (1 µg/Kg)*. All fish samples found below the MRPL (1.0 ng/mL) for AOZ metabolite residues and declared negative.

From the results, it was observed that nitrofuran, being a banned drug, is not available in Pakistan and in practice for use in aquatic products. However, this developed analytical capability will be helpful to provide analytical services for AOZ/AMOZ analysis in local/exportable food samples.

**Monitoring studies of Chloramphenicol (CAP) in Milk by using ELISA**

Commercial ELISA Kit (Cat # W81113 Quicking Biotech) was used to monitor CAP residues in bovine milk (40 in number) available in local market of Faisalabad region. Different dilution levels (0.025, 0.1, 0.2, 0.4, and 1.6 ng/mL) were standardized. Middle of the test (IC_{50}) found to be 0.31 ng/mL and lowest detection limit 0.07 ng/mL. From the results, it was observed that 4 samples (out of 40) contained CAP below MRPL (0.3 ppb) whereas only one sample contained 0.35 ng/mL and declared positive for CAP. Rest of the samples found negative.

*Official Journal of European Union (Regulations 2010/37)

**Aflatoxins (B_1, B_2, G_1, G_2) in wheat by HPLC and TLC.**

Studies to determine aflatoxin (B_1, B_2, G_1, G_2) in wheat were initiated to monitor the status of aflatoxin contamination in wheat samples available in local market and also to study aflatoxin production during storage. From the results, it was observed that no aflatoxin were present in wheat samples except 3 samples of Godown no. 2 set no. 3; Godown no. 5 set no. 3, and flour samples of a
Flour Mill. In these samples, TLC spots of aflatoxin B_1, B_2 and G_2 were present at the Rf values of 0.40, 0.45 and 0.30. However, these levels were below the maximum permissible limits of European Union regulatory limits (i.e. 2 ppb) in food. These positive samples were subjected to HPLC analysis which revealed that maximum concentration of aflatoxin B_2 (3.2 ppb) was present in the samples of Godown No. 2, Set No. 3 at the retention time of 4.7 min. In other samples, negligible amounts of Aflatoxins were present. Results of TLC and HPLC methods were comparable and reproducible.

Samples were stored under normal practices one year and analysed by TLC. From the results, it was observed that 5 categories of wheat samples (out of 11), were contaminated with Aflatoxin B_1, B_2 and G_1 after one year of storage; G_2 was absent. However, their concentrations were below the European Commission Regulatory limits (2 ppb) in food. After 2 years of storage, 10 categories of wheat samples, out of 11, were contaminated above the permissible limits for food.

**Deltamethrin analysis in milk by HPLC method:**

An HPLC method was standardized and validated for deltamethrin analysis in milk by Hitachi L-2000 Series HPLC System. Extraction/analytical protocol were optimized and applied to milk samples (n=10) collected from different dairy farms and local market areas. Keeping in view the EU permissible limits (50 ppb), 5 samples (out of 10) were positive in the range of 261-652 ppb. Samples of Dhuddiwala and Ghulam M. Abad areas had 530 and 652 ppb.

**Bovine Growth Hormone (BGH) analysis in Bovine Serum:**

Commercial ELISA Kit (Abnova GH Bovine ELISA Kit, Catalog # KA 2279, version: 3) was standardized for bovine growth hormone (BGH) analysis in serum of bovine and other related species. Reference standards of 0, 1.0, 2.5, 5, 10, 25, 50 ng/mL were used for standardization. Lowest detection limit (IC_{20}) and middle of the test (IC_{50}) were found to be 5.0 ppb and 26.0 ppb respectively. Test found suitable for analysis of Bovine growth hormone in serum. Bovine serum samples from 38 treated and one non-treated (negative control) buffaloes were collected from dairy farms at Theekriwala and Sammundari Road (near Faisalabad). After analysis, 7 samples (out of 38) were found negative for BGH. Whereas positive samples showed hormone concentration in the range from 0.3 to 9.5 ng/mL.

**Human Resource Development:**

- M.Phil (48) and PhD (8) Students were taught (M.Phil-Course BIO-512 on Analytical Techniques) and (PhD Course BIO-628 on Veterinary Drug Residues) under M.Phil/PhD degree program (PIEAS, Islamabad, NIAB Campus).

- One student from University of Agriculture, Faisalabad completed 4-Months Fellowship on Bovine Somatotropin analysis in bovine blood of control/treated animals.
In-House VDR Training Programs (M. Phil./Ph.D./Internship)

Dr. Uzma Maqbool, Head Food Safety Laboratories, NIAB participated as invited speaker at an International Conference entitled “Research and Enterprise on Food Safety in Developing World” sponsored by DAAD-HEC held at COMSAT Islamabad (December 5-8, 2016). She presented a talk on “Quality Control/Quality Assurance (QC/QA) of Food Safety Analysis”.

Receiving Certificate from Ms. Ina Lapel German Ambassador in Pakistan.

Ms. Mehwish Mumtaz, SE NIAB/Team member IAEA RAS 5078 and Mr. Muhammad Shahzad, JS NIAB attended Training course entitled, “Radio Receptor Assays, related screening and confirmatory methods for veterinary drug residues and associated chemical/natural food contaminants from 22nd May to 2nd June 2017 at BQCLP, Bangkok, Thailand.
Dr. Uzma Maqbool, Mr. Ismail Chughtai and Ms. Mehwish Mumtaz attended Seminar on Food Safety and Harmonization on 22nd Nov. 2017 at Serena hotel, Islamabad, Pakistan. Where Awilo Ochieng Pernet, outgoing-chairperson, codex Alimentarius Commission, Federal Food Safety and Veterinary Officer FSVO was also invited by Nestle group of Industries.

2. Outreach Programs:


3. Publications:


Remarks:

We are highly obliged for the technical as well as financial assistance (an amount of 24459.00 Euros) provided by the PMO and TO for the purchase of Liquid Scintillation Counter and Consumables. With the help of group trainings, procurements and expert support, it is hoped that Pakistan will be able to enhance national food safety and competitiveness in the export market.

Future Programs:
Data sharing and participation in IAEA TCP RAS5078 activity plan
Data sharing and participation in IAEA TCP INT5154 activity plan
Receptor Development and Isolation under IAEA Coordinated Research Program (CRP) entitled, “Radiotracer studies to develop/validate multi-class analytical methods for analysis of multiple food contaminants”. (IAEA R. C. No. 22144/R0).
Tetracycline Standardization and validation in tissue employing HPLC-UV-DAD.
Analytical services for CAP, Quinolone and Semicarbazide analysis in Sheep casings.
Farmer’s Day and Food Safety Training Course

4. News/Announcement:


Success Reported Reference

Understanding the context

Food safety is a global concern, especially in addressing food security. The Joint FAO/IAEA Programme of the Food and Agriculture Organization of the United Nations (FAO) and the International Atomic Energy Agency (IAEA) conducts research and supports capacity building on nuclear and isotopic techniques to enhance food safety and quality assurance. The programme is designed to address specific food safety challenges and improve food safety in countries with limited resources.

What FAO does

The Programme’s activities are coordinated and supported by FAO and IAEA, and implemented by IAEA, FAO, and its Member States in collaboration with other United Nations organizations and international, regional, and national agencies.

The Programme’s activities include:
- Conducting research and training on the application of nuclear techniques to improve food safety and quality assurance in developing countries.
- Developing and implementing guidelines, standards, and protocols for the safe and effective use of nuclear techniques in food safety.
- Providing technical assistance and capacity building to Member States in the use of nuclear techniques for food safety.
- Promoting international cooperation and information exchange on nuclear techniques for food safety.

Contact us

For more information, please visit the Joint FAO/IAEA Programme on Science and Technology for Better and Affordable Food website at www.fao.org/iaea.
Food Safety Labs, NIAB, Success Story - 2016

Agricultural and food security success stories

2014
- Strengthening food security: Establishing a laboratory for monitoring veterinary drug residues in food in Pakistan
- Implementing GMP (Good Manufacturing Practice) in animal feed production to ensure the quality and safety of animal feed.
- Food safety and quality assurance: Establishing a system for monitoring and control veterinary drug residues in food commodities.

2015
- Strengthening food security: Establishing a laboratory for monitoring veterinary drug residues in food in Pakistan
- Implementing GMP (Good Manufacturing Practice) in animal feed production to ensure the quality and safety of animal feed.
- Food safety and quality assurance: Establishing a system for monitoring and control veterinary drug residues in food commodities.

The challenges:
- Rising threats of antibiotic resistance and other drug residues in food.
- Limited access to quality food products.
- Lack of adequate food safety regulations and enforcement.

The impact:
- Improved food safety and quality assurance systems.
- Increased confidence in the food supply chain.
- Enhanced cooperation with international partners.
Brochure for Farmer's Day (in Urdu)

Farmer’s Day at Jaranwala
20th November 2017

Jaranwala 2017

Training Course on “Analytical Techniques for Food Safety Measures in Pakistan”
(27th September - 6th October, 2016)
ANALYTICAL SERVICES FOR VETERINARY DRUG RESIDUES
VDR Labs, Animal Sciences Division
NIAB (PAEC), FAISALABAD

Food safety and quality have become increasingly important worldwide in recent years, not only in terms of protecting the health of the consumer but also to meet requirements for international trade. Threats such as microbial contaminations, heavy metals, radioactivity, residues from pesticides, aflatoxins, veterinary drugs/hormones in food & their resulting health hazards have made the public quality conscious. It is of increasing concern for regulatory authorities of many developing countries including Pakistan that export foods to the major trading blocks of the developed world. To facilitate such trade and improve food quality nationally, it is necessary to develop awareness among stakeholders involved from ‘farm to fork’ directly or indirectly for the implementation of international standards for the production of quality assured foods. Not only productions, but also for testing of finished product fit for human consumption.

Nuclear Institute for Agriculture and Biology (NIAB), Faisalabad, is a research and development (R&D) centre functioning under the auspices of Pakistan Atomic Energy Commission (PAEC). NIAB mandate is to conduct research on applied problems in the field of agriculture and biology using nuclear and other related techniques. It also includes efforts to achieve food safety at the national level as well as for export purposes. Veterinary Drug Residues (VDR) Analytical Laboratories of NIAB are working on food and environment safety issues for the last 25 years with special reference to pesticides.