COMBINED EFFECT OF PREBIOTICS & IRON FORTIFICANTS ON VARIOUS TYPES OF WHITE BLOOD CELLS IN IRON DEFICIENT FEMALE SPRAGUE DAWLEY RATS

Abdul Momin Rizwan Ahmad[1,*], Waqas Ahmed[2], Sanaullah Iqbal[2], Muhammad Hassan Mushtaq[2], Riffat Aysha Anis[3]

The current study was designed to determine the combined effect of prebiotics & iron fortificants on various types of White blood cells among Iron deficient female Sprague Dawley rats. For the present study, n = 126 female Sprague Dawley rats aged 6 to 8 weeks were obtained. Initially, anemia was induced among rats by feeding them with carbon tetrachloride which is an iron binder. After that, rats were orally fed with fortified feed daily for a period of three months. Five different types of white blood cells including Neutrophils, Lymphocytes, Basophils, Monocytes and Eosinophils were determined using the standard protocols. The results of the study showed that iron fortificants and prebiotics did not improve any of the white blood cells.

Background & Objectives

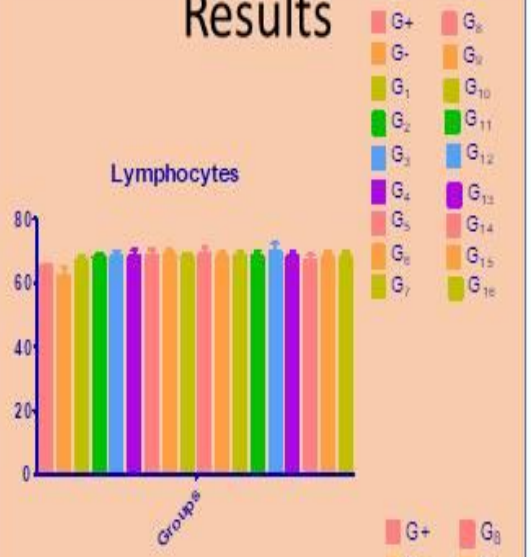
Iron deficiency is one of the biggest public health problems worldwide. Despite focusing on this menace for decades, this problem is still on rise and continues to affect millions across the globe.

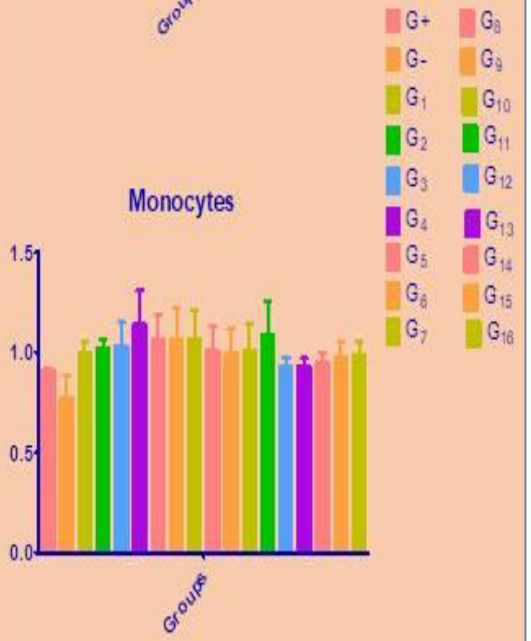
1. To probe the combined effect of Prebiotics [GOS & Inulin] & iron fortificants [NaFeEDTA & $FeSO_4$] on various types of White Blood Cells in Iron deficient female Sprague Dawley Rats

Methodology

For the present study, n = 126 female Sprague Dawley rats aged 6 to 8 weeks were obtained from the National Institute of Health, Islamabad. Two prebiotics namely Inulin and Galacto-oligosaccharides and two iron salts including Ferrous sulphate and Sodium Iron EDTA were used in varying dosages to prepare various types of feed to be fed to rats. Initially, anemia was induced among rats by feeding them with carbon tetrachloride which is an iron binder. After that, rats were orally fed with fortified feed daily for a period of three months. Blood samples of overnight fasted rats were collected at 0, 30th, 60th and 90th day of the trials. Five different types of white blood cells including Neutrophils, Lymphocytes, Basophils, Monocytes and Eosinophils were determined using the standard protocols.

Results





Conclusion

The current study indicated that white blood cells are not impacted by consumption of iron fortificants and/or prebiotics.

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Acknowledgement

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INAPPROPRIATE USE OF HAND SANITIZER DURING THE COVID-19 OUTBREAK LED TO AN INCREASE IN MULTIPLE DRUGS RESISTANCE; A REVIEW

Faisal Siddique[1], Kashif Akram[2], Wasim Babar[3], Muhammad Saeed[4], Asif Javaid[5]

[1]Department of Microbiology, Cholistan University Of Veterinary and Animal Sciences, Bahawalpur, Pakistan
[2]Department Department of Food Science, Cholistan University Of Veterinary and Animal Sciences, Bahawalpur, Pakistan
[3]Department of Parasitology, Cholistan University Of Veterinary and Animal Sciences, Bahawalpur, Pakistan
[4]Department of Animal Nutrition, Cholistan University of Veterinary and Animal Sciences, Bahawalpur
[5]Department of Animal Nutrition, Cholistan University of Veterinary and Animal Sciences, Bahawalpur

Corresponding Author:
Email: faisalsiddique@cuvas.edu.pk

Abstract

Historical background: The global risk of multi-resistant bacteria is increasing day by day in both patients and healthy individuals, especially in children and the elderly who use commercially available hand sanitizers, especially in under developing and poor countries during the COVID-19 epidemic. There is no drug or vaccine available to treat COVID-19 infections up till now. The World health organization recommended the easiest way to control the transmission of infection in humans is to use a hand sanitizer.

Objective: The purpose of this review was to highlight the inappropriate, low quality and persistent use of various commercially available hand sanitizers in Pakistan, as well as their role in the development of antibiotic resistance in the normal human microflora.

Methodology: Data was collected from various organizational websites around the world, including the World Health Organization, Food and Agriculture Organization, and research articles, research reports, and review articles.

Results: Hand sanitizers are cheap and appear to be effective in combating SARS CoV infection. However, excessive use of these sanitizers can be fatal. Despite the lack of current data, the prediction of ethyl alcohol through its absorption of skin tumors and its carcinogenic effects is still the subject of scientific debate and controversy. Popular hand sanitizers are also known to reduce toxic effects. Isopropyl alcohols, including ethanol, have some adverse effects on human health and the ecosystem.

Conclusion: The excessive use and proliferation of substandard hand sanitizers has skyrocketed, causing significant harm to human lives as a result of COVID-19. Irrational and inappropriate use of these hand sanitizers leads to the development of gradual resistance in normal microbiota found in humans. Inappropriate or irrational use of hand sanitizer could pose a significant challenge in the near future.

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STUDY OF HISTOLOGICAL EXPRESSION OF INTESTINAL MUCOSAL HEALTH IN ROCK PIGEON IN RESPONSE TO THE DIETARY INTERVENTION OF MANNAN OLIGOSACCHARIDE

Sahar Ijaz*[1], Hafsa Zaneb[1], Saima Masood[1], Muhammad Shahbaz Yousaf[2], Saima Ashraf[1] and Mirza Muhammad Usman[1]

*1 Department of Anatomy and Histology, UVAS, LHR, Pakistan; 2 Department of Physiology, UVAS, LHR, Pakistan; *Presenter's Email: sahar.ijaz@uvas.edu.pk*

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ABSTRACT

Background:

Intestinal health of a food animal is of extreme significance for the animal's and for its consumer's health. The histology of the gut translates gut health. The Rock Pigeon (*Columba livia domestica*) is a bird thriving in our environment and this makes its intestine a worth studying subject. With the history of its meat usage in traditional medicine and as a delicacy, this bird could be an alternative meat source to make up for the increasing demand of animal protein. We recorded the dimensions of selected mucosal components in the small intestine of the Rock Pigeon and compared based on the feed given to different groups.

Objective: To evaluate the gut health through mucosal dimensional histological data from the Rock Pigeon's small intestine both with and without the dietary intervention.

Methodology:

Forty maternally isolated chicks were divided into four groups. Group A was fed the corn based basal diet (CBBD) while the groups B, C and D were fed CBBD with 0.1, 0.2 and 0.5% MOS respectively. At the end of the trial, the birds were sacrificed and samples from the small intestine were processed for the histological study of Villus height, Villus width, Crypt depth, Villus surface area, Mucosal absorptive surface area, Villus height to crypt depth ratio and thickness of lamina propria. The data were analyzed through one way ANOVA and Duncan's multiple range test. P<0.5 was considered significant.

Results:

The dimensions of the selected mucosal components were the greatest in the birds fed the CBBD only.

Significance:

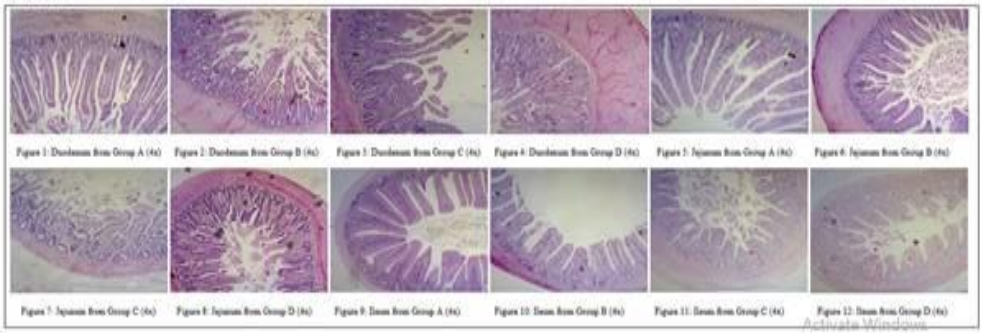
The results signify that the rock pigeon's intestinal mucosa has shown a good growth pattern in terms of enhanced dimensions when CBBD was fed while the dietary intervention of MOS failed to encourage any enhancement in the dimensions of the selected mucosal components. This calls for digging down deep into the rock pigeon's intestinal histology as well as the effect of MOS on it.

Keywords:

Rock Pigeon, Small intestine, Mucosal histology.

Parameters	Groups*				
	A	B	C	D	p-value
Duodenum					
VH (µm)	339.86 ±57.86^{b}	232.99 ± 51.11^{a}	222.41 ± 63.39^{a}	201.02 ±76.54^{a}	0.001
VW(µm)	51.58 ± 8.6	49.84 ± 8.11	53.75 ± 10.53	44.12 ± 7.47	.217
CD(µm)	67.04 ±18.61	75.16 ± 14.23	89.27 ± 38.35	90.40 ± 45.32	.391
LPT(µm)	74.35 ± 20.07	77.59 ± 14.23	101.43 ± 41.79	98.29 ± 39.89	.206
MMT(µm)	14.29 ± 3.41^{b}	11.75 ± 2.42^{ab}	11.24 ± 2.49^{a}	11.23 ± 1.07^{a}	.073
VSA (um^2)	55417.74 ± 13391.74^{b}	36986.91 ±12390.06^{a}	36568.98 ± 9559.08^{a}	28466.48 ± 11744.70^{a}	.001
MSA(µm^2)	14.13 ± 3.94^{b}	10.30 ± 2.21^{a}	9.25 ± 3.58^{a}	8.73 ± 2.92^{a}	.011
VH:CD	5.28 ± 1.27^{b}	3.14 ± 0.62^{a}	2.65 ± 0.71^{a}	2.64 ±1.14^{a}	.000
Jejunum					
VH (µm)	285.60 ± 52.59^{b}	175.55 ± 45.08^{a}	203.86 ± 35.33^{a}	216.49 ± 1.66^{a}	.003
VW(µm)	48.88 ± 6.04^{ab}	40.16 ± 14.52^{a}	54.24 ± 11.26^{b}	45.86 ± 12.48^{ab}	.132
CD(µm)	58.69 ± 13.87^{ab}	49.31 ± 12.15^{a}	70.48 ± 20.6^{b}	64.06 ± 19.26^{ab}	.092
LPT(µm)	60.29 ± 13.6^{ab}	53.11 ± 11.19^{a}	78.15 ± 24.53^{b}	71.95 ±18.97^{ab}	.034
MMT(µm)	12.76 ± 3.01	10.84 ± 3.11	10.79 ± 2.31	11.46 ± 3.75	.557
VSA (um^2)	44306.06 ± 11383.67^{b}	22294.85 ± 10210.17^{a}	35526.38 ± 13531.13^{ab}	33432.80 ± 19316.24^{ab}	.024
MSA(um^2)	11.48 ± 3.34^{b}	08.34 ± 3.09^{a}	7.82 ± 1.60^{a}	8.54 ± 1.36^{a}	.040
VH:CD	5.10 ± 1.33^{b}	3.91 ± 1.95^{ab}	2.96 ± 0.35^{a}	3.43 ± 1.15^{a}	.033
Ileum					
VH (µm)	191.21 ± 26.11	136.51 ± 43.61	156.00 ± 90.94	145.46 ± 33.31	.199
VW (µm)	52.01 ± 5.98^{a}	46.30 ± 5.93^{ab}	44.44 ± 5.92^{b}	39.71 ± 7.00^{b}	.007
CD (um)	51.12 ± 6.34	59.10 ± 9.59	52.47 ± 12.03	53.35 ± 11.59	.381
LPT(µm)	51.08 ± 9.01	61.06 ± 10.56	52.82 ± 16.41	51.91 ± 6.75	.245
MMT(µm)	13.98 ± 3.49^{bc}	16.60 ± 4.81^{c}	12.14 ± 1.55^{ab}	9.50 ± 0.92^{a}	.002
VSA (um^2)	31381.22 ± 6310.80^{b}	20231.32 ± 8197.68^{a}	21125.82 ± 10702.43^{a}	18222.10 ± 5296.54^{a}	.012
MSA(um^2)	7.75 ± 1.13^{b}	4.99 ± 1.02^{a}	4.70 ± 1.29^{a}	5.78 ± 1.36^{a}	.000
VH:CD	3.81±0.80^{b}	2.39 ± 0.97^{a}	3.09 ± 1.87^{ab}	2.75 ± 0.55^{ab}	.096

*A= CBBD , B=CBBD +0.1% MOS, C= CBBD +0.2% MOS, D = CBBD +0.5% MOS, VH= Villus Height, VW= Villus Width, CD=Crypt Depth, LPT= Lamina Propria thickness, MMT= Muscularis Mucosa Thickness, VSA= Villus Surface Area, MSA= Mucosal Absorptive Surface Area, VHCD= Villus Height to Crypt Depth ratio. * Within the same row, means with different superscripts are significantly different(P<0.05). The results are reported as means ± S.D



Detection of Colistin Resistance (MCR) genes among Multi Drug Resistant *Escherichia coli* isolated from beef samples

Zeeshan Nawaz*, Laiba Arshad, Abu Baker Siddique, Muhammad Asif Zahoor, Rasti Abbas and Muhammad Qasim
Department of Microbiology, Government College University, Faisalabad

Abstract

The emergence of drug resistance against colistin among *Escherichia coli* from different type of meat is a major public health threat. In present study, a total of 100 samples of fresh and frozen beef (n=50 each) were collected from different butcher's shops and super markets. The samples were processed for isolation and identification of *E. coli* by standard microbiological, biochemical and molecular procedures. Antibiotic susceptibility profiling was done by disc diffusion for the detection of multi drug resistant isolates. The phenotypic resistance of MDR isolates to colistin sulfate was detected by colistin broth disk elution test while presence of (MCR) genes was detected by polymerase chain reaction. The results of present study showed that out of total beef samples, 40 showed presence of *E. coli* including 22 from fresh beef and 18 from frozen beef samples. The isolates showed 100% resistance to ciprofloxacin, cefepime, tetracycline followed by 97.5% to cefotaxime and 95% to ampicillin, hence 38 (95%) were detected as MDR- *E. coli*. Among the MDR isolates, 30 (75%) showed resistance to colistin sulfate phenotypically. The presence of mcr-1 gene was detected in 18 (60%) while none of the isolate showed presence of mcr-2 gene among colistin resistant MDR strains of *E. coli*. The results indicated elevated presence of mcr-1 harboring *E. coli* among beef animals leading to its spread among humans posing a serious public health threat.

Background & Objectives

- Emergence of multidrug resistant bacteria among food animals is creating a serious public health threat
- Isolation and Molecular detection of *Escherichia coli* from fresh and frozen beef samples
- Detection of multi drug resistance (MDR) and colistin resistance genes from *E. coli* isolates

Methodology

Sample collection

Overall 100 fresh and frozen beef samples were collected from butcher's shops and super markets

Isolation and Identification of *Escherichia coli*

Molecular detection of *Escherichia coli* by using PCR

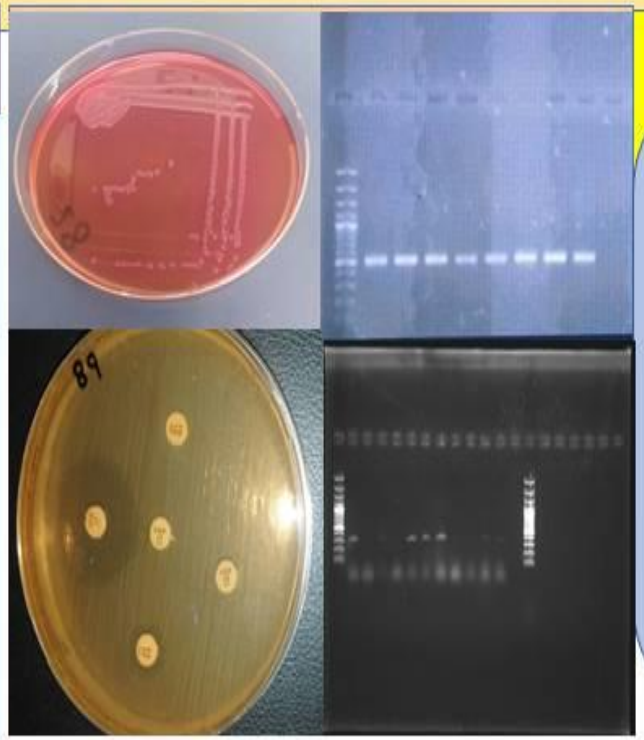
Congo Red Dye Test

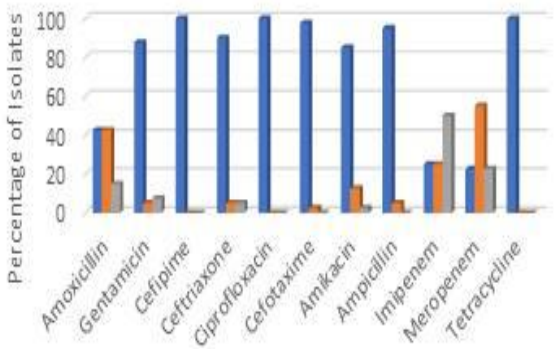
Antibiotic Susceptibility Test

Multi drug resistant *E. coli* (MDR-EC)

Colistin Broth Disc Elution Test

Detection of mobile colistin resistance (*mcr*) genes





Conclusion

- Beef samples are highly contaminated with pathogenic *E. coli* (40%).
- The percentage of MDR and colistin resistant isolates are at alarming levels.
- The mcr1 harbouring *E. coli* in food animals transmits colistin resistance to human beings

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ANTIMICROBIAL EVALUATION OF COPPER NANOPARTICLES SYNTHESIZED THROUGH GREEN SYNTHESIS METHOD USING AQUEOUS LEAF EXTRACT OF *AZADIRACHTA INDICA LINN.*

Ahmad Raza[1], Muhammad Adil Rasheed*[1], Muhammad Ovais Omer[1], Imran Iltaf[2], Usama Bin Naeem[1], Qurat ul Ain[1]

[1]Department of Pharmacology & Toxicology, University of Veterinary and Animal sciences, Lahore.
[2]Quality operational Laboratory, Institute of Microbiology, University of Veterinary and Animal sciences, Lahore.

**Presenter e-mail address: dr_aadil@uvas.edu.pk*

ABSTRACT

Background: Antibacterial resistance is increasing day by day leading to therapeutic failure and increase in the cost of treatment. Change in drug delivery system to body may decrease resistance of bacteria against antibacterial agents.

Objective(s): This project is designed to evaluate antimicrobial activity of copper nanoparticle (CuNPs) against *Eschrchia coli* and *Staphylococcus aureus.*

Methodology: Copper Nanoparticles were prepared through green synthesis method using aqueous leaf extract of Neem (*Azadirachta indica*). Characterization of nanoparticles was done using UV-spectroscopy and Fourier transform infrared (FTIR) spectroscopy. Scanning electron microscope (SEM) was used to determine the size of copper nanoparticles. Antibacterial effect of nanoparticles was evaluated using well diffusion method against field strains of *Escherichia coli,* and *Staphylococcus aurous*.

Results: UV-spectroscopy exhibited resonance absorption peak at 570nm, which is a convenient region of the spectrum, FTIR spectroscopy showed peaks at 1630 and 3400cm-[1] which confirms the presence of copper nanoparticles. The size of copper nanoparticles was ranging from 39-62 nm with average size 49.44 nm. Inhibitory zones were measured and compared with the guidelines of CLSI. Data was expressed as means±SD. P value ≤ 0.05 considered as significant. The diameter of zones of inhibition on plates exposed with nanoparticles and crude extract of Neem was (20.96±1.06), (11.38±1.59) respectively. It is concluded that nanoparticles has produced significant antimicrobial effect against *E.coli* and *S.aureus*.

Significance: The Copper nanoparticle produced by aqueous leaf extract of Neem could be an alternative choice for treatment of diseases causing by *Eschrchia coli* and *Staphylococcus aureus*.

Keywords: Copper nanoparticles, Neem extract, antibacterial resistance, green synthesis

BACK GROUND

Antibacterial resistance is increasing day by day leading to therapeutic failure and increase in the cost of treatment.

Change in drug delivery system to body may decrease resistance of bacteria against antibacterial agents.

OBJECTIVES

This project is designed to evaluate antimicrobial activity of copper nanoparticle (CuNPs) against *Eschrchia coli* and *Staphylococcus aureus.*

METHODOLOGY

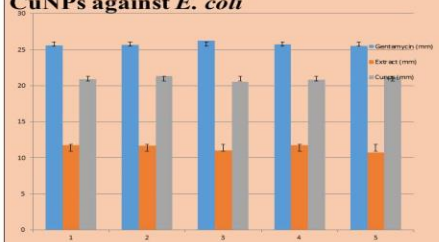
Copper Nanoparticles were prepared through green synthesis method using aqueous leaf extract of Neem (*Azadirachta indica*).

Characterization of nanoparticles was done using UV-spectroscopy and Fourier transform infrared (FTIR) spectroscopy. Scanning electron microscope (SEM) was used to determine the size of copper nanoparticles.

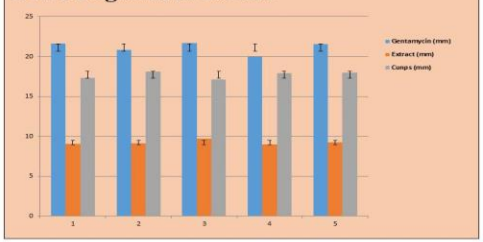
Antibacterial effect of nanoparticles was evaluated using well diffusion method against field strains of *Escherichia coli,* and *Staphylococcus aurous*.

RESULTS

Graphical representations of zones of inhibition by Gentamicin, Extract and CuNPs against *E. coli*



Graphical representations of zones of inhibition by Gentamycin, Extract and CuNPs against *S. aureus*



CONCLUSION

It is concluded that nanoparticles produced by neem leaves have significant antimicrobial effect against *E.coli* and *S.aureus* with average zone of inhibition of 15.66mm and 20.03mm respectively. Likewise the neem leave extract has less significant antimicrobial ability as compared to copper nanoparticles against *E.coli* and *S.aureus* with average zone of inhibition of 11.49mm and 9.21mm respectively.

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Spread of antimicrobial resistance of zoonotic diseases from animals to humans

Muhammad Hunain Ahmed[1], Muhammad Tariq Javed[1], Muhammad Haider Ali[1], Muhammad Hassan Tahir[1], Sami Ullah Khan Bahadur[1], Narmeen Tariq[2], Aira Tariq[3], Shaza Zarnab[1]

[1]Department of Pathology, University of Agriculture, Faisalabad, Pakistan.
[2]Department of Microbiology, University of Veterinary and Animal Sciences, Lahore, Pakistan.
[3]Department of Clinical Medicine and Surgery, University of Agriculture, Faisalabad, Pakistan.

Background

Antibiotic development was considered as the most important advancement in modern science. However, the prolonged and overuse of antibiotic raised antimicrobial resistance, a significant threat to animal and human health. The resistant bacteria can be passed through affected animals to human through food chain, or by direct and indirect contact. The resistant genes are mostly found on gene cassettes, plasmids and transposons of various bacterial strains.

The comparative analysis of whole genome (human and animal strain) revealed different types of antimicrobial resistance genomes. These findings revealed the importance of One Health concept to find the association of resistance genes and zoonotic infectious pathogens.

According to Marshal and Levy, it can be seen that different resistance types have been transferred from animals to humans through commensals bacteria (as shown in the table)

Commensal	Resistance transferred	Evidence
E. coli	Tetracycline (From US Chickens to Caretakers, farm family)	Following introduction of tetracycline on a farm, resistant E. coli strains with transferable plasmids were found in caretakers' gut floras, with subsequent spread to the farm family
E. coli	Gentamicin (From Chinese chicken to poultry workers)	Increase in phenotypic, gentamicin resistance in workers through direct contact with chickens receiving gentamicicn prophylactically
E. coli	Apramycin (Not in humans) (From Chinese chicken to farm workers)	Detection of aac(3)-IV apramycin resistance gene in humans, with 99.3% homology to that animal strains
Salmonella Newport	Ampicillin, carbenicillin, tetracycline (From Beef cattle to patient with diarrhea)	Direct genetic tracking of rsistance plasmid from humberger meat to infected patient
E. Coli, Salmonella	Apramycin, Gentamicin (From Belgian cattle to hospital patients)	Plasmid based transfer of aac(3)-IV gene bearing ristance to a drug used only in animals (apramycin)
Enterococcus faecium	Vancomycin (From chickens to patient with diarrhea)	Clonal spread of E. faecium and horizontal transmission of vanA gene cluster (Tn1546) found between animals and humans
E. coli	Ciprofloxacin (From Spanish chickens to bacteremic hospital patients)	Multiple molecular and epidemiological typing modalities demonstrated avian source of resistant E. coli

Effects of antibiotic use in food animals on human

- ✓ Exposure to farm animals treated with antibiotics causes increased risk of resistant colonization or infection in humans.
- ✓ Consumption of food contaminated with antibiotic-resistant bacteria causes an outbreak of resistant diarrheal disease.
- ✓ Consumption of antibiotic-containing meat products induces resistance in normal flora of the human gastrointestinal tract.
- ✓ Transport of animals causes dispersion of resistant bacteria along route.
- ✓ Mobile genetic elements from antibiotic-resistant bacteria in animals are incorporated into pathogens that cause cases of human infection.
- ✓ Resistant bacteria from animal waste used as fertilizer cause contamination of water supply and alterations in human flora.

Conclusion

For the containment of AMR, we need to develop best alternatives to antibiotics, these could be improved managemental practices, use of vaccines, probiotics, use of IgY and nanoparticles.

References and Acknowledgement

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Marshal and Levy (doi:10.1128/CMR.00002-11)

Association of Type-III Secretions with Multi Drug Resistant *Pseudomonas aeruginosa* from Urinary Tract Infection

Muhammad Iqbal1, Abu Baker Siddique*1, Bilal Aslam1, Zeeshan Nawaz1, Muhammad Hidayat Rasool1 and Muhammad Asif Zahoor1

1 Department of Microbiology, Government College University, Faisalabad

Abstract

Pseudomonas aeruginosa is the prominent causative agent of urinary tract infections (UTIs) resulting in a high mortality rate worldwide. It has the ability to produce toxins and type III secretions that inhibit polypeptide elongation than can lead to a septic shock and systemic infection. The present study is conducted and main focus is to determine the resistance pattern and coexistence of type III secretion with MDR *P. aeruginosa*. A Total of 200 Urine samples were collected from UTI patients from different hospitals and processed on Pseudomonas Cetrimide agar. Different biochemical test was carried out in order to identify *P. aeruginosa* and molecular characterization is done by polymerase chain reaction. AST profiling done by performing the Kirby-Bauer disc diffusion technique.Molecular detection of Type III Secretions was done by using specific primers through PCR. Total 72 (36%) isolates of *P. aeruginosa* was detected on basis of PCR. Isolates of *P. aeruginosa* showed highest resistance towards ceftazidime (100%), ceftriaxone (100%), nalidixic acid (95%), piperacillin (85%), gentamicin (85%), aztreonam (85%), tobramycin (80%) and amikacin (80%), norfloxacin (70%), ciprofloxacin (60%) and Imipenem (70%). Gene specific PCR indicated 40 (56%) Isolates of *P. aeruginosa* with prevalence of ExoS, 36(50%) ExoU, 32(44%) ExoY and 24(33%) ExoT. It is concluded that T3SS (exoS and exoU) associates more than 50% in infections occurred due to MDR *P. aeruginosa*.

Background & Objectives

Uropathogenically isolated *P. aeruginosa* is most common cause of nosocomial infections & accounts 35% among all nosocomial infections.

In developing countries, UTIs are listed as one of the most prevalent rank 3rd among other infections.

acute pyelonephritis or cystitis , chills, fever and flank pain

Antimicrobial Resiatance in *P. aeruginosa*

Super bug

Mechanisms of AMR

- ❑ Intrinsic mechanism
- ❑ Acquired mechanism
- ❑ Adaptive mechanism

Type III Secretion System

- complex of needle like machines
- Direct injection of toxins into the cytosol of host cells
- Translocation apparatus
- Chaperone proteins
- Enzymes of T3SS

Methodology

Sample collection

→ Overall 200 Urine samples were collected from three tertiary health care hospitals of FSD

Isolation and Identification of *Pseudomonas aeruginosa*

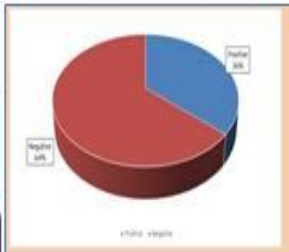
Molecular detection of *P. aeruginosa* by using PCR

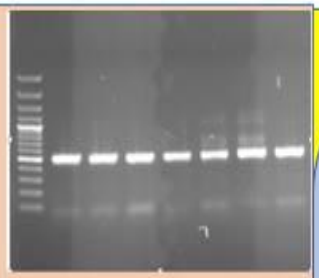
→ Genus-specific Primer oprL
Specie -pecific Primer oprl

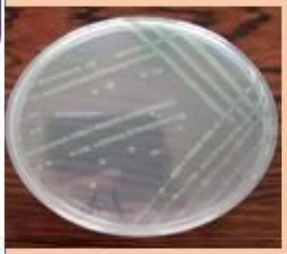
Molecular detection of type-III Secretion genes by using PCR

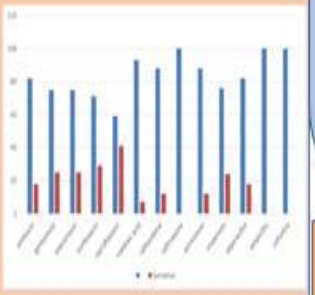
→ Genes specific primers for T3SS genes including ExoS, ExoT, ExoU, ExoY

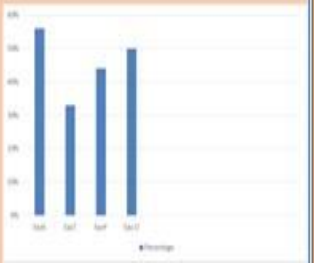
Antibiotic Susceptibility Test











Conclusion

- Major pathogen found in the UTIs are P. aeruginosa - 36%
- Occurrence of T3SS gene harboring isolates was ExoS, 36(50%) ExoU, 32(44%) ExoY and 24(33%) ExoT.
- T3SS (exoS and exoU) associates more than 50% in infections occurred due to MDR *Pseudomonas aeruginosa*.

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Longitudinal study of experimental induction of AA amyloidosis in mice seeded with homologous and heterologous AA fibrils

Dr. Muhammad Naeem

Abstract

To investigate pathogenesis and kinetics of experimentally induced murine AA amyloidosis seeded with homologous(murine) and heterologous(bovine) AA fibrils .Severity of AA amyloidosis induced with homologous AA fibrils is higher compared to heterologous AA fibrils.

Background & Objectives

- Amyloidosis is a group of protein miss-folding diseases afflicting animals and humans. Amyloid A(AA) amyloidosis involves the pathological deposition of insoluble AA fibrils in various body tissues and organs.
- Precursor protein of AA amyloidosis is serum amyloid A (SAA), which is an acute phase reactant primarily produced by hepatocytes.
- The objective is to investigate pathogenesis and kinetics of experimentally induced murine AA amyloidosis seeded with homologous (murine) and heterologous (bovine) AA fibrils

Methodology

- Experimental AA amyloidosis was induced by administration of inflammatory stimulus and preformed AA fibrils to a total of 111 female C57/Black mice.
- In this longitudinal study, heterologous (bovine) as well as homologous (murine) AA fibrils were injected intraperitoneal to mice in various combinations. Re-stimulation
- was done at 120 or 300 days post first inoculation. To analyze the intensity of amyloid depositions in mice organs, immunohistochemical techniques and image J software were used.
- Assessment of cytokines level in sera was done using a Mouse Th1/Th2/Th17 Cytokine CBA Kit.

Results

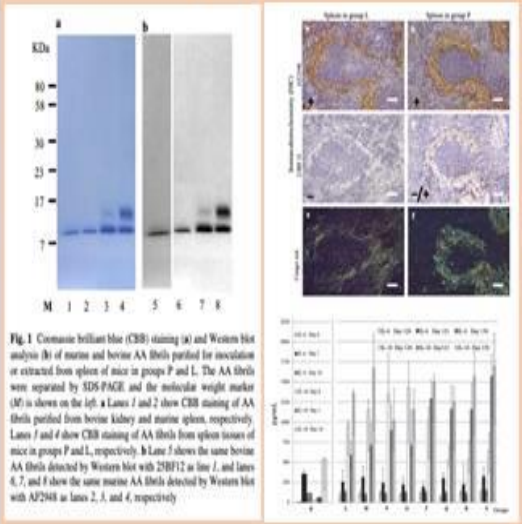
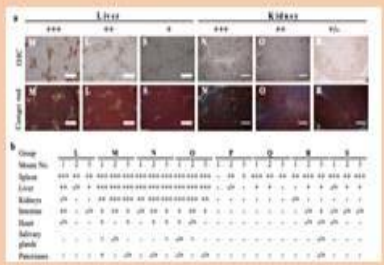


Fig. 1 Coomassie brilliant blue (CBB) staining (a) and Western blot analysis (b) of murine and bovine AA fibrils purified for inoculation or extracted from spleen of mice in groups P and L. The AA fibrils were separated by SDS-PAGE and the molecular weight marker (M) is shown on the left. a Lanes 1 and 2 show CBB staining of AA fibrils purified from bovine kidney and murine spleen, respectively. Lanes 3 and 4 show CBB staining of AA fibrils from spleen tissues of mice in groups P and L, respectively. b Lane 5 shows the same bovine AA fibrils detected by Western blot with 25BF12 as line 1, and lanes 6, 7, and 8 show the same murine AA fibrils detected by Western blot with AF2948 as lanes 2, 3, and 4, respectively



Increase in the level of proinflammatory cytokine IL-6 was observed after first inoculation, while second inoculation caused a further increase in the level of anti inflammatory cytokine IL-10.

Conclusion

- AA amyloidosis can be induced by heterologous as well as homologous AA fibrils.
- Severity of AA amyloidosis induced with homologous AA fibrils is higher compared to heterologous AA fibrils.

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GENE-EDITING BASED MODERN TECHNIQUES FOR MOSQUITO CONTROL

Maria Kausar*[a], Muhammad Sohail Sajid[a], Muhammad Abdullah Malik[a]

a: Department of Parasitology, University of Agriculture, Faisalabad, Pakistan.

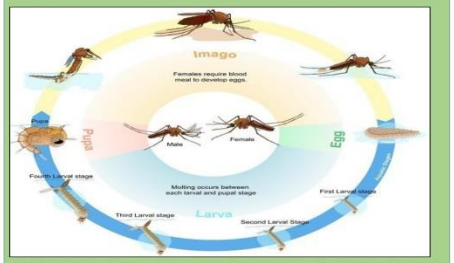
*Corresponding Author: Maria Kausar rkousar371@gmail.com

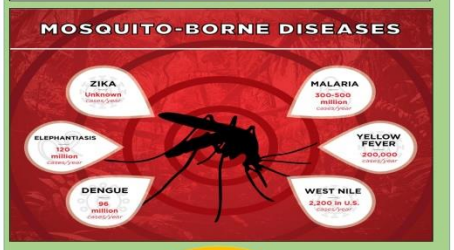
ABSTRACT

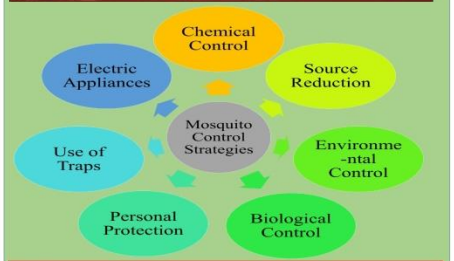
Mosquito control strategies include chemical control, source reduction, environmental control, biological control, personal protection, and use of traps. But the present techniques are ineffective, offering a continuously global threat demanding advanced genetic tools to control mosquito-transmitted diseases. Genome editing techniques like RNA interference (RNAi), zinc finger nucleases (ZFN), transcription activator-like effector nucleases (TALEN), clustered regulatory interspaced palindromic repeats (CRISPR), and associated protein (Cas 9), and immune-related gene modification with the development of antiviral immunity. RNAi uses double-stranded RNA (dsRNA), and small interfering RNA/short interfering technique (siRNA), single stranded hairpin micro-RNA (miRNA), and short hairpin RNAs (shRNA) to halt the protein synthesis through post-transcriptional mRNA silencing. RNAi has knocked down many lethal genes, and identified essential genes like *IAP1*, *Ubi-p63E* and *CG11700* genes along with-it DNA binding proteins *bss* and *CG1545*. But this tool is laborious, limited to RNA based gene silencing with high-off target effects and there might be an immune response stimulation by the introduced RNA. The most recently used tool CRISPR: a gold standard to knocks out the genes on DNA level with no off-target effects. CRISPR has knocked down many genes like *AGAP005958*, *AGAP011377,* and *AGAP007280 (female fertility genes)*. It causes the permanent disruption of gene resulting in robust signal, lowers the risk of immune modulation with a flexible time frame for assay. By employing CRISPR researchers can impede protein synthesis, mating, and consequently control of mosquito and mosquito borne pathogens.

Keywords: Mosquitoes, control techniques, gene-editing, CRISPR

BACKGROUND & OBJECTIVES

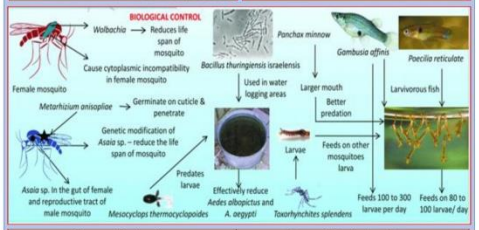


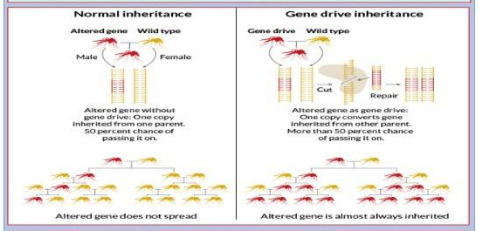


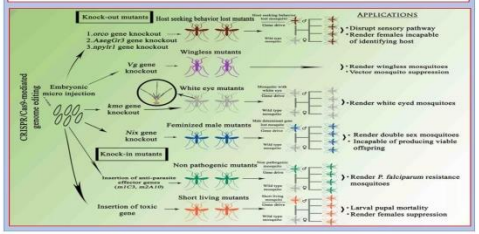


- To elaborates the pros and cons of earlier mosquito control strategies
- Review based evaluation of modern mosquito control techniques

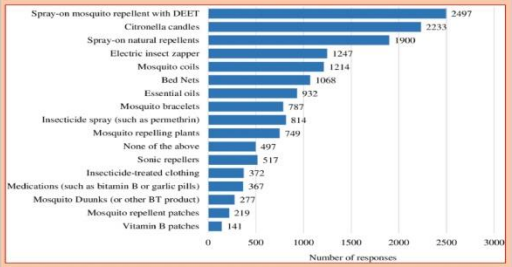
METHODOLOGY

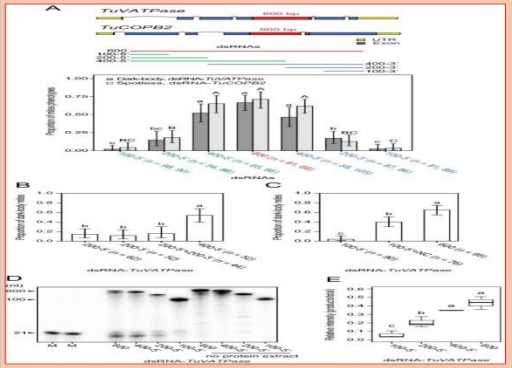






RESULTS





CONCLUSION

- The present techniques are ineffective, offering a continuously global threat demanding advanced genetic tools to control mosquito-transmitted diseases
- RNAi has knocked down many lethal genes, and identified essential genes like *IAP1*, *Ubi-p63E* and *CG11700* genes along with-it DNA binding proteins *bss* and *CG1545*. But this tool is laborious, limited to RNA based gene silencing with high-off target effects and there might be an immune response stimulation by the introduced RNA
- CRISPR-Cas9 causes the permanent disruption of gene resulting in robust signal, lowers the risk of immune modulation with a flexible time frame for assay. By employing CRISPR researchers can impede protein synthesis, mating, and consequently control of mosquito and mosquito borne pathogens

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Effect of prebiotic and Selenium-nanoparticles alone or in combination on ameliorating higher stocking density stress on absolute and relative organ weights, and meat quality in broiler chickens

HF Rehman[1]*, H Zaneb[1], S Masood[1], MS Yousaf[2], S Ashraf[1], MM Usman[1], K Hayat[1], S Ijaz[1], M Zeeshan[1], KA Majeed[2], MAA Saeed[2], I Khan[3], MS Yasin[4], H Rehman[2]
[1]Department of Anatomy and Histology, University of Veterinary and Animal Sciences, Lahore, Pakistan [2]Department of Physiology, University of Veterinary and Animal Sciences, Lahore, Pakistan [3]College of Veterinary Sciences and 3Animal Husbandry, Abdul Wali Khan University, Mardan, Pakistan [4]Institute of Biochemistry & biotechnology University of Veterinary and Animal Sciences, Lahore, Pakistan
*For correspondence: faseeh.rehman@uvas.edu.pk

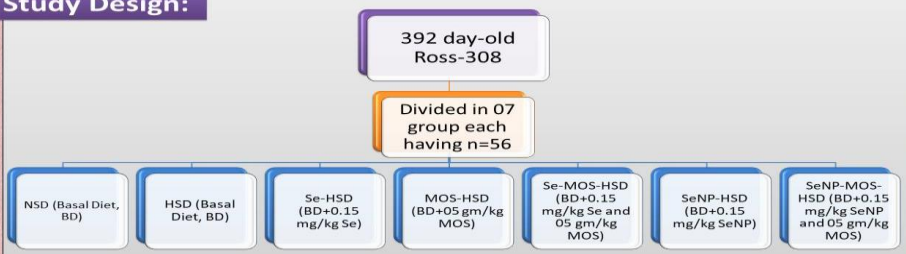
Abstract

Objective: Current study was carried out to investigate the effect of mannan-oligosaccharide (MOS) and selenium-nanoparticle (SeNP) alone or in combination in broiler reared under higher stocking density (HSD) on absolute and relative visceral organ weights and certain meat quality parameters.
Methodology: The 392 day-old chicks Ross-308 were randomly divided in seven groups with eight replicates (n=7) as NSD (basal diet (B-D)+ normal stocking density: 10 bird/m^2), HSD (basal diet+ higher stocking density: 16bird/m^2), Se-HSD (B-D + Se-0.15mg/kg), MOS-HSD (B-D + MOS-05gm/kg), Se-MOS-HSD (B-D + Se-0.15 mg/kg + 05gm/kg of MOS), SeNP-HSD (B-D + SeNP 0.15mg/kg) and SeNP-MOS-HSD (B-D + 0.15 SeNP + 05gm/kg of MOS). Each group consisted of eight replicates (n=7). On 21st and 42nd day, two birds from each replicate were slaughtered for measuring absolute and relative organ weights and sampled breast muscle for pH at 0-hour (0-H) and 24-hours (24-H) post slaughter and drip loss percentage.
Result: At 21st day, absolute weights of liver, small intestine (filled and empty), large intestine (filled and empty) and absolute length of small intestine were lowest ($p < 0.05$) in HSD group and highest($p < 0.05$) in NSD group, whereas for Se-MOS-HSD and SeNP-MOS-HSD it was comparable with NSD group. At 42nd day, absolute weights of liver, Bursa of Fabricius, spleen and small intestine (empty) were lowest ($P < 0.05$) in HSD group. Liver weight was highest ($p < 0.05$) in SeNP-HSD group; Bursa of Fabricius weight was highest ($p < 0.05$) in SeNP-MOS+HSD group. Spleen weight was highest ($p < 0.05$) in Se-MOS-HSD, SeNP-HSD, SeNP-MOS-HSD and NSD-control group. While relative organ weight, at both 21st day and 42nd day, did not vary significantly ($P > 0.05$) for all investigated visceral organs amongst experimental groups. At 21st day, there was no significantly difference ($P < 0.05$) in pH-0H, pH-24H and water holding capacity of breast meat. At 42nd day pH-i was lower (P < 0.05) in all HSD groups than NSD group, while pH-u was lower ($P < 0.05$) in HSD group than NSD group and it was comparable in all HSD-supplemented groups with NSD-control group. Water holding capacity was highest with low drip loss % ($P < 0.05$) in NSD and SeNP-MOS+HSD groups, lowest ($P < 0.05$) in HSD and MOS-HSD.

Background:

Broiler chicken welfare is under increasing scrutiny due to welfare concerns regarding growth rate and stocking density. Stocking density in broiler production is perceived as a topic of major importance; however, no consensus has been reached on what density would allow for good welfare as well as economic benefits to producers.

Study Design:



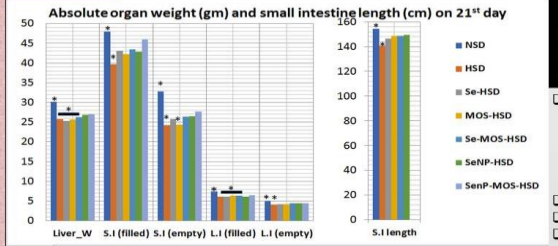
- Trail duration was 42 days.
- **NSD**: Normal stoking density (10 bird/m^2) & **HSD:** High stoking density (16 bird/m^2)
- Sampling done on 21st and 42nd day

The parameters measured were:

- Organ Weight (Absolute & Relative)
- Breast Muscle:
  - pH at 0-hour (0-H) and 24-hours (24-H) post slaughter
  - Drip loss percentage

Statistical analysis : One way ANOVA was used on Statistical Package for Social Science (SPSS for Windows version 13, SPSS Inc., Chicago, IL, USA) for analysing data. Data was presented as mean for each group.

Result:

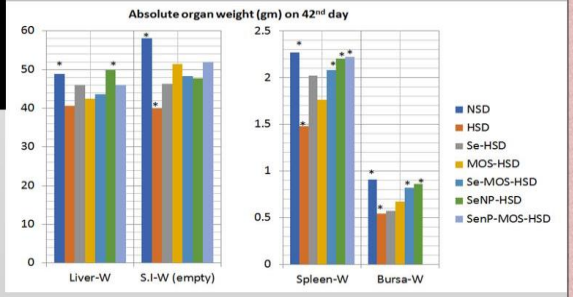


Effects of SeNP and MOS supplementations on Absolute organ weight and length on 21st day in broilers reared under normal stocking density and high stocking density conditions

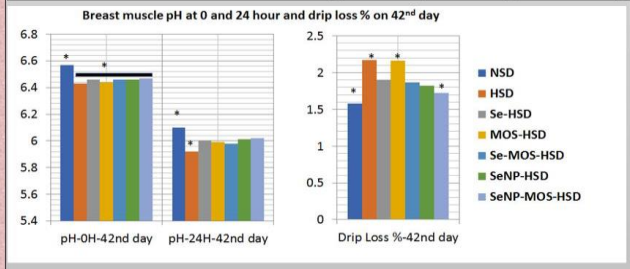
- Graph of Absolute organ weight and length on 21st day shows that broilers supplemented with SeNP, Se-MOS and SeNP-MOS improved absolute organ weight (P<0.05) and comparable with the Normal stocking density group.
- Data is presented as mean of each group
- * on bars differed significantly (P<0.05).
- Bars without * on bars did not differ significantly

Effects of SeNP and MOS supplementations on Absolute organ weight on 42nd day in broilers reared under normal stocking density and high stocking density conditions

- Graph of Absolute organ weight on 42nd day shows that broilers supplemented with SeNP, Se-MOS and SeNP-MOS improved absolute organ weight (P<0.05) in high stocking density groups and comparable with the Normal stocking density group.
- Data is presented as mean of each group
- * on bars differed significantly (P<0.05).
- Bars without * on bars did not differ significantly



Effects of SeNP and MOS supplementations on breast muscle pH and drip loss % in broiler reared under normal stocking density and high stocking density conditions



- Graph of Absolute pH and drip loss % shows that broilers supplemented with SeNP, Se-MOS and SeNP-MOS improved breast muscle pH and Water holding capacity on 42nd day (P<0.05) in high stocking density groups and comparable with the Normal stocking density group.
- Data is presented as mean of each group
- * on bars differed significantly (P<0.05).
- Bars without * on bars did not differ significantly

- **NSD:** Normal stoking density (10 bird/m^2)
- **HSD:** High stoking density (16 bird/m^2)
- **Se:** 0.15 mg/kg Selenium selenite
- **SeNP:** 0.15 mg/kg Selenium nano-particles
- **MOS:** 05 gm/kg Mannan oligosaccharide

Conclusion

Based on results, we concluded that higher stocking density is a stressful situation for bird which can negatively influence absolute organ weight and meat quality. Supplementations of Se-MOS and SeNP-MOS in higher stocking density partially mitigated HSD effects in aforementioned parameters. In current experimental condition SeNP-MOS performed better than all other supplementations.

Poultry zoonotic diseases and concept of one health

Zain Ul Abadeen[1]*, Muhammad Tariq Javed[1], Syed Muhammad Faizan[1]

[1]Department of Pathology, University of Agriculture, Faisalabad-38040, Pakistan.

(Corresponding Author email: drzain47@gmail.com)

Poultry zoonotic diseases

Zoonotic diseases are characterized as diseases transmitted by natural means between humans and vertebrate animals. A list of zoonotic diseases including Avian tuberculosis, Salmonellosis, Ornithosis, Avian influenza, Cryptosporidiosis, and Campylobacteriosis that are transmitted from poultry to humans beings. These diseases are mainly caused by pathogens like viruses, fungi, parasites, and bacteria and carried by free-range animals. Direct and indirect contact between humans and animals are responsible for the transmission of these pathogens. The presence of poultry zoonotic diseases is observed around the world including in developing, underdeveloped and developed countries that affect the economy of the countries and public health. In viral diseases, avian influenza outbreaks exert enormous economic losses in terms of loss of poultry and poultry products. The foodborne zoonotic diseases of bacterial origin are about 90% of all intestinal infection cases in human beings and species of *Salmonella* and *Campylobacter* bacteria are mainly involved. In poultry meat and eggs, the presence of *Salmonella* species is commonly observed @ 5.6% and 1%, respectively. These diseases produce their impact in terms of (a) damaging the safety and quality products (b) higher production cost (c) decreased production (d) lower quality products (e) compromised biosafety (f) poor hygiene (g) infections to handlers (h) low labor availability (i) shortage of safe food for human beings. These factors exert serious effects on public health along with decreased availability of animal-source protein.

The efforts including training of staff, government agencies involvement, public awareness, more research and funding along with the implementation of effective policies should be adopted to control and prevent such infections.

One health

The term 'One Health' could be defined as the interdisciplinary approaches and actions involving strong cooperation primarily between physician and veterinarian so that health of people and animals could be ensured including the safety of the environments through with improved cooperation between physicians and veterinarians. The concept of 'One Health' represents a unique and significant opportunity for veterinary medicine to be in a leadership role and to work collaboratively for ecosystem health (a community of humans, animals, and plants interacting with one another and with their physical environment) for the greater good of sense.

Conclusion: It is concluded that one health can play valuable and diversified roles in society by collaborating and developing communication with other medical professionals for providing facilities for humans, animals, and environmental protection. Despite several achievements of 'One Health' for example control of Avian Influenza HPAI H5N1, more collaborations are crucial to connect subject matter experts from different disciplines originating from local, regional, national and global level for better 'One Health' activities to ensure safe health and a better world.

History of One health

'One Health' is a new phrase, but the concept was originated as far back as to the time of Aristotle in 500 BC and Hippocrates of Cos in 340 BC. Aristotle wrote the 'Historia Animalium' to elaborate on the natural zoonotic history of animals. Effect of environmental factors on human health could be traced by the Greek physician Hippocrates (c. 460 BCE – c. 370 BCE) in his text 'On Airs, Waters and Places'. The Italian Physician Giovani Maria Lancisi (1654– 1720) also talked on the role the environment in the spread of disease in animals and humans. Lancisi is one of those few people who first suggested on the mosquito nets for prevention malaria in humans (Drake, 1832) but was also a pioneer in the control of rinderpest in cattle. Later on, Louis-René Villerme (1782–1863) and Alexandre Parent-Duchatelet (1790– 1835) talked about linkages of human and animal health with environment and developed the specialty of concept of public hygiene (LaBerge, 1992). Although human medicine developed many years back, the veterinary medicine appeared as a distinct discipline in China during the dynasty of Zhou between 11th to 13th century (Driesch and Peters, 2003). However, in 1762, the first veterinary school was established in Lyon, France. The term 'zoonosis' was first sued by the German physician and pathologist Rudolf Virchow (1821–1902) in 19th century. Canadian physician Sir William Osler (1849–1919) was one of the pioneers appointed as faculty combindly to work in the Montreal Veterinary College and the Medical School of McGill University (Kahn et al., 2007). The term 'One Medicine' was developed and promoted by veterinary epidemiologist and parasitologist Calvin W. Schwabe (1927–2006) in 1984. In 2003, the term 'One Health' draw the attention across the globe since it was mentioned in Washington Post by Dr. William Karesh to describe the outbreaks of Ebola hemorrhagic fever in Africa. He said, 'Human or livestock or wildlife health can't be discussed in isolation anymore. There is just One Health. And the solutions require everyone working together on all the different levels'. This was followed by a series of conferences across the globe with the theme of 'One World – One Health'.

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