

Research Article

Evaluation of Antimicrobial Activity of Various Chemicals on Isolated Chicken and Mutton Spoilage Microorganisms

Ahmad Mir B¹, Amin Mir M^{2*}, Singh D¹ and Rao Z¹

¹Department of Microbiology and Zoology, Uttaranchal College of Biomedical Sciences and Hospital, India

²Research Scholar at Uttarakhand Technical University, India

*Corresponding author: Mohd Amin Mir, Research Scholar at Uttarakhand Technical University, Dehradun, India

Received: April 28, 2017; Accepted: May 22, 2017;

Published: May 29, 2017

Abstract

The study was carried out on the isolation of various micro-organisms, in which various samples of chicken and mutton were collected from different locations of the Dehradun. The process of slaughtering of the chicken and mutton as per the survey was of two types- viz, Jatka and Halal. The isolated micro-organisms from the chicken samples of Jatka type include- *Vibrio*, *Enterobacter Species*, *Clostridium*, *E.coli*, *Shigella*, where as the micro-organisms isolated from the chicken samples of Halal type include only *Enterobacter Species*, *Clostridium* and *E.coli*. The micro-organisms isolated from the mutton sample of Jatka type include, *Shigella*, *E.coli*, *Vibrio*, *staphylococcus*, and the micro-organisms isolated from the mutton samples of Halal type include only *E.coli*, *Vibrio*, and *staphylococcus*. The micro-organisms have been isolated and identified by various biochemical tests. The isolated micro-organisms have been then testified for antimicrobial susceptibility test against various chemical substances which include, Gentamicin, Naphthazarin, Azithromycin, Citric Acid, Amphotericin-B, Clindamycin, Cefixime, Norfloxacin, Streptomycin, Ampicillin. Among the various used chemical substances, Gentamicin have been found effective in inhibiting the growth of all the isolated micro-organisms, followed by Norfloxacin, Streptomycin, Ampicillin, Azithromycin, Citric Acid. Least antimicrobial activity was observed for Naphthazarin, Amphotericin-B, Clindamycin, and Cefixime. Among the isolated micro-organisms *shigella* species was found to be less prone against the applied anti-microbial drugs, and the most susceptible micro-organism among the isolated micro-organisms includes *E.coli*. Also from the collected chicken and mutton samples the samples of Halal type have been to be less effective against the growth of micro-organisms as compared to the Jatka type samples, which could be due to different procedure of slaughtering technique.

Keywords: Chicken and mutton samples; Anti-microbial activity; Halal and Jatka; *Vibrio*; *Enterobacter Species*; *Clostridium*; *E.coli*; *Shigella*; *Staphylococcus*

Introduction

Microorganisms can be used to transform raw foods into fermented delights, including yoghurt, cheese, sausages, Tempeh, pickles, wine, beers and other alcoholic products. On the other hand, foods also can act as a reservoir for disease transmission, and thus detection and control of pathogens and spoilage organisms are important areas of food microbiology. During the entire sequence of food handling from the producer to the final consumer, microorganisms can affect food quality and human health.

This study briefly summarizes current knowledge on the biological implications of biogenic amines on human health and collects data on the factors affecting their formation in meat and fermented meat products by Joanna Stadnik, Zbigniew J. Dolatowski [1]. George-John E. Nychas, Panos N. Skandamis, Chrysoula C. Tassou, Konstantinos P. Koutsoumanis [2] studied on meat spoilage during distribution can be considered as an ecological phenomenon that encompasses the changes of the available substrata (e.g., low molecular compounds),

during the prevailing of a particular microbial association, the so-called Specific Spoilage Organisms (SSO).

Microbial contamination of poultry carcasses can be influenced by many factors during transport and slaughtering. The study carried out by Irena Svobodová, Gabriela Bořilová [3] evaluated the impact of **Table 1:** Showing various bacterial strains isolated from the chicken and mutton samples.

S. No	Name of Bacteria Isolated			
	Chicken Sample		Mutton Sample	
	Halal	Jatka	Halal	Jatka
1	<i>Shigella</i>	-----	-----	<i>Shigella</i>
2	<i>Enterobacter Species</i>	<i>Enterobacter Species</i>	-----	-----
3	<i>Clostridium</i>	<i>Clostridium</i>	-----	-----
4	<i>E.coli</i>	<i>E.coli</i>	<i>E.coli</i>	<i>E.coli</i>
5	<i>Vibrio</i>	-----	<i>Vibrio</i>	<i>Vibrio</i>
6	-----	-----	<i>staphylococcus</i>	<i>Staphylococcus</i>

Table 2: Showing zone of inhibition against *Shigella* bacteria.

S. No	Strain Used	Chemical Used	Zone of Inhibition (mm)
1	<i>Shigella</i>	Gentamicin	29
2	<i>Shigella</i>	Naphthazarin	19
3	<i>Shigella</i>	Azithromycin	6
4	<i>Shigella</i>	Citric Acid	27
5	<i>Shigella</i>	Amphotericin-B	Nil
6	<i>Shigella</i>	Clindamycin	10
7	<i>Shigella</i>	Cefixime	Nil
8	<i>Shigella</i>	Norfloxacin	18
9	<i>Shigella</i>	Streptomycin	17
10	<i>Shigella</i>	Ampicillin	13

Table 3: Showing zone of inhibition against *E. coli* bacteria.

S. No	Strain Used	Chemical Used	Zone of Inhibition (mm)
1	<i>E.coli</i>	Gentamicin	28
2	<i>E.coli</i>	Naphthazarin	15
3	<i>E.coli</i>	Azithromycin	06
4	<i>E.coli</i>	Citric Acid	26
5	<i>E.coli</i>	Amphotericin-B	Nil
6	<i>E.coli</i>	Clindamycin	Nil
7	<i>E.coli</i>	Cefixime	Nil
8	<i>E.coli</i>	Norfloxacin	18
9	<i>E.coli</i>	Streptomycin	17
10	<i>E.coli</i>	Ampicillin	13

Table 4: Showing zone of inhibition against *Vibrio* bacteria.

S. No	Strain Used	Chemical Used	Zone of Inhibition (mm)
1	<i>Vibrio</i>	Gentamicin	30
2	<i>Vibrio</i>	Naphthazarin	16
3	<i>Vibrio</i>	Azithromycin	Nil
4	<i>Vibrio</i>	Citric Acid	25
5	<i>Vibrio</i>	Amphotericin-B	Nil
6	<i>Vibrio</i>	Clindamycin	Nil
7	<i>Vibrio</i>	Cefixime	Nil
8	<i>Vibrio</i>	Norfloxacin	Nil
9	<i>Vibrio</i>	Streptomycin	15
10	<i>Vibrio</i>	Ampicillin	Nil

four processing steps (plucking, evisceration, washing and chilling) on the Total Viable Counts (TVC), counts of *Escherichia coli*, *Salmonella* spp. and *Listeria* spp. incidence on broiler carcasses. Food borne illness is a major public health concern. The largest number of food borne illness cases attributed to poultry and poultry products are caused by paratyphoid serotypes of *Salmonella* and by *Campylobacter jejuni* as studied by P.L. White A.R. Baker & W.O. James [4]. Consumption of meat is continuously increasing worldwide. The annual per capita consumption increased from 10kg in the 1960s to 26kg in 2000 and will reach 37kg by the year 2030 as per the reports by Joanna Stadnik, Zbigniew J. Dolatowski, [5]. On the other hand, a significant portion of meat and meat products are spoiled every year. Kantor et al. (1997)

Table 5: Showing zone of inhibition against *Enterobacter* Species bacteria.

S. No	Strain Used	Chemical Used	Zone of Inhibition (mm)
1	<i>Enterobacter</i> Species	Gentamicin	27
2	<i>Enterobacter</i> Species	Naphthazarin	20
3	<i>Enterobacter</i> Species	Azithromycin	Nil
4	<i>Enterobacter</i> Species	Citric Acid	24
5	<i>Enterobacter</i> Species	Amphotericin-B	Nil
6	<i>Enterobacter</i> Species	Clindamycin	13
7	<i>Enterobacter</i> Species	Cefixime	Nil
8	<i>Enterobacter</i> Species	Norfloxacin	Nil
9	<i>Enterobacter</i> Species	Streptomycin	15
10	<i>Enterobacter</i> Species	Ampicillin	Nil

Table 6: Showing zone of inhibition against *clostridium* bacteria.

S. No	Strain Used	Chemical Used	Zone of Inhibition (mm)
1	<i>Clostridium</i>	Gentamicin	27
2	<i>Clostridium</i>	Naphthazarin	17
3	<i>Clostridium</i>	Azithromycin	14
4	<i>Clostridium</i>	Citric Acid	25
5	<i>Clostridium</i>	Amphotericin-B	Nil
6	<i>Clostridium</i>	Clindomycin	19
7	<i>Clostridium</i>	Cefixime	Nil
8	<i>Clostridium</i>	Norfloxacin	20
9	<i>Clostridium</i>	Streptomycin	12
10	<i>Clostridium</i>	Ampicilin	10

Table 7: Showing zone of inhibition against *Staphylococcus* bacteria.

S. No	Strain Used	Chemical Used	Zone of Inhibition (mm)
1	<i>Staphylococcus</i>	Gentamicin	24
2	<i>Staphylococcus</i>	Naphthazarin	17
3	<i>Staphylococcus</i>	Azithromycin	Nil
4	<i>Staphylococcus</i>	Citric Acid	22
5	<i>Staphylococcus</i>	Amphotericin-B	Nil
6	<i>Staphylococcus</i>	Clindamycin	11
7	<i>Staphylococcus</i>	Cefixime	Nil
8	<i>Staphylococcus</i>	Norfloxacin	Nil
9	<i>Staphylococcus</i>	Streptomycin	13
10	<i>Staphylococcus</i>	Ampicillin	Nil

reported that approximately 3.5 billion kg of poultry and meat were wasted at the consumer, retailer and foodservice levels which have a substantial economic and environmental impact.

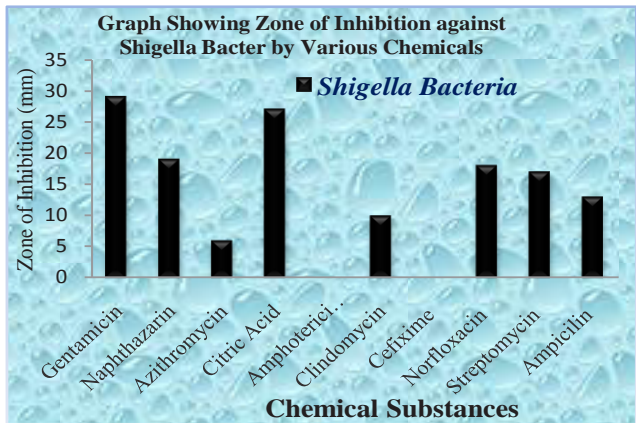
Material and Methods for Antimicrobial Analysis

The antimicrobial analyses of various “Chicken and Mutton” samples were carried out at Uttaranchal College of Biomedical Sciences and Hospital, Dehradun.

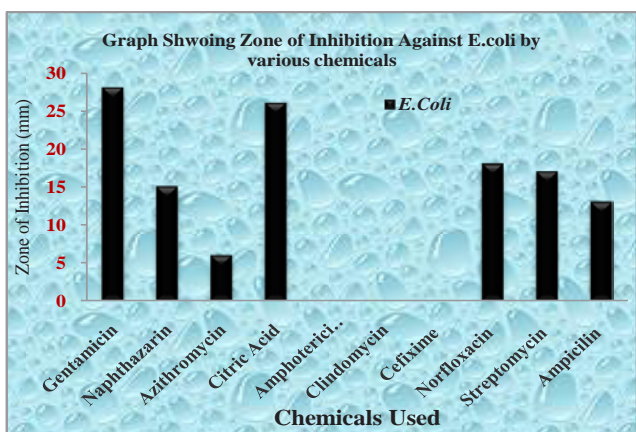
Microbial world is of great importance in the present world. They are beneficial in various ways to humans, not only they help

Table 8: Showing zone of inhibition against *Salmonella* bacteria.

S. No	Strain Used	Chemical Used	Zone of Inhibition (mm)
1	<i>Salmonella</i>	Gentamicin	27
2	<i>Salmonella</i>	Naphthazarin	16
3	<i>Salmonella</i>	Azithromycin	Nil
4	<i>Salmonella</i>	Citric Acid	27
5	<i>Salmonella</i>	Amphotericin-B	Nil
6	<i>Salmonella</i>	Clindamycin	Nil
7	<i>Salmonella</i>	Cefixime	Nil
8	<i>Salmonella</i>	Norfloxacin	Nil
9	<i>Salmonella</i>	Streptomycin	15
10	<i>Salmonella</i>	Ampicillin	Nil



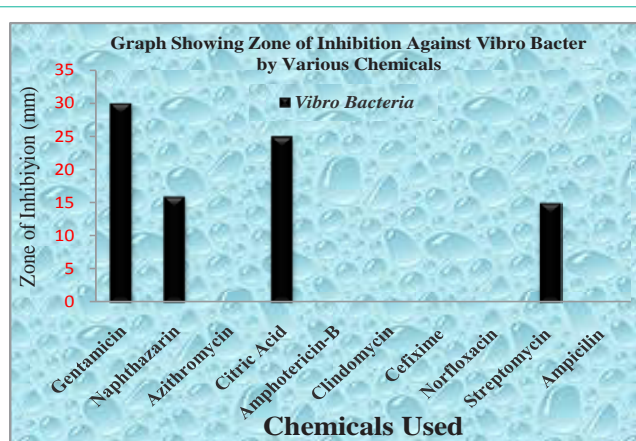
Graph 1: Showing effect of chemicals on *Shigella* Bacteria.



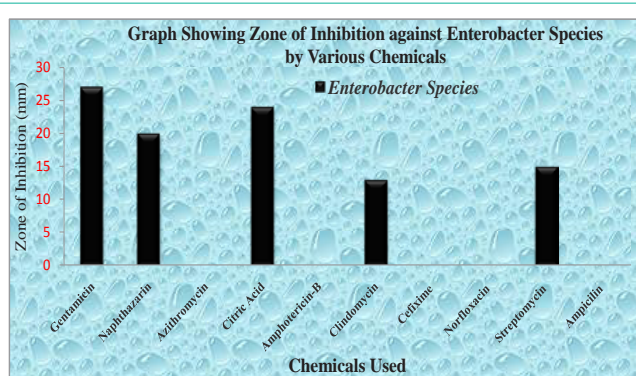
Graph 2: Showing Effect of chemicals on *E.coli* bacteria.

to maintain the environment clean, but also they furnish various types of antibiotics which are useful against various diseases. So this microbial analysis was carried out. All the chemicals used were of analytical grade.

Different samples of chicken and mutton were collected from the local shops at D. L. Road and Sahastradhara crossing, Dehradun then leaved all of these samples for 4 days for complete spoilage.



Graph 3: Showing Effect of chemicals on *Vibrio* bacteria.



Graph 4: Showing Effect of chemicals on *Enterobacter* Species bacteria.

Identification methods

All the bacterial colonies were identified by Gram’s staining method as they are Gram -ve or Gram +ve & several biochemical characterization methods.

Litmus milk test, Gelatin Agar test and Urea agar media were used to determine the load of viable cells followed by several biochemical tests for this purpose.

Observations

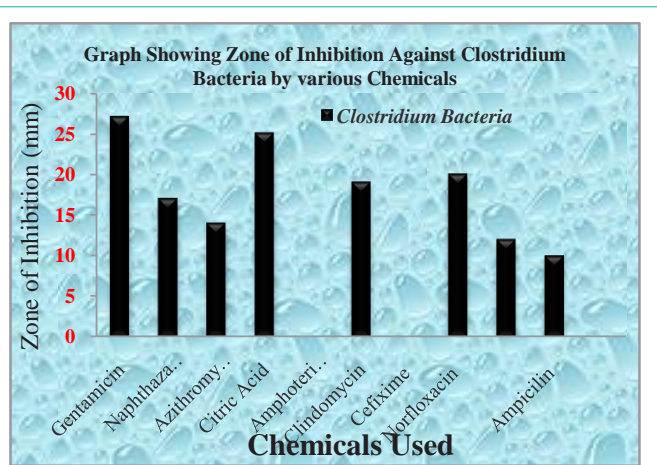
Identification of Bacterial strains: So far our observation is taken into consideration the following types of microorganisms have been isolated from the meat and chicken samples and are mention in the following tables (Table 1-8).

Antimicrobial Susceptibility Testing: Antimicrobial activity of various chemical substances against various isolated bacterial strains from chicken and mutt on samples (Graph 1-7).

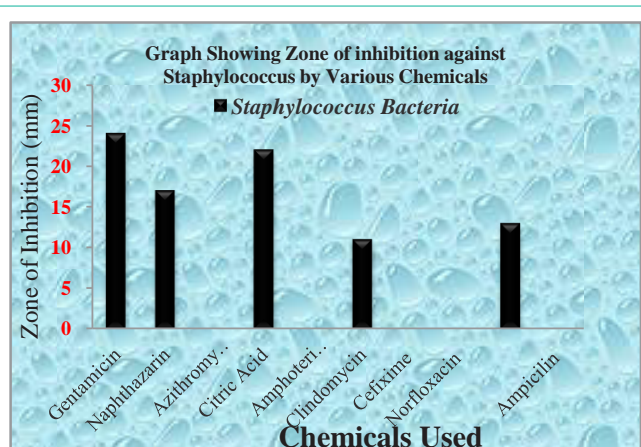
Results and Discussion

Altogether 06 bacterial isolates were obtained from chicken sample and 04 isolates from mutton sample. The bacterial isolates were screened for their thermo-tolerance property in different temperatures starting from 50°C to 90°C.

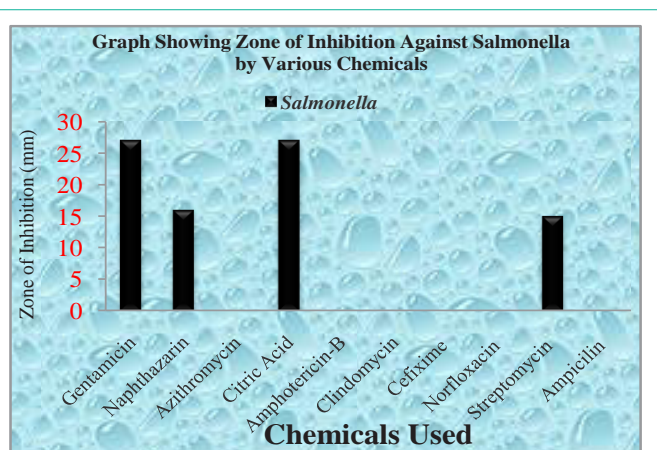
The bacterium *Staphylococcus* species is a spherical bacterium



Graph 5: Showing Effect of chemicals on Clostridium bacteria.



Graph 6: Showing Effect of chemicals on Staphylococcus bacteria.



Graph 7: Showing Effect of chemicals on Salmonella bacteria.

(coccus) which on microscopic examination appears singly, in pairs, or bunched, grape-like clusters. They are Gram-positive, facultative anaerobes, but grow rapidly under aerobic conditions and were characterized by morphological observation and biochemical tests. Salmonella is a rod-shaped, Gram negative, non-sporulating,

facultative anaerobic motile bacterium. They are mesophiles with a growth temperature range of 5 to 46°C and optimum growth temperature of 35 to 37°C. Litmus milk test, Gelatin Agar test and Urea agar media viable cells followed by several biochemical tests lead to the identification of Salmonella. Shigella was identified as Gram-negative, non-motile, non-spore forming rod-shaped bacteria as per Litmus milk test, Gelatin Agar test and Urea agar media followed by several biochemical tests. The strains grow between 7 and 46°C, with an optimum at 37°C. Litmus milk test, Gelatin Agar test and Urea agar media followed by several biochemical tests identified E. coli as harmless, Gram negative, motile, non-sporulating; rod shaped facultative anaerobic bacterium. Vibrio bacterium as identified as Gram-negative, non-sporulating motile curved rods with an optimum temperature of 30°C to 37°C, but can grow over a temperature range of 5 and 42°C as per Gelatin Agar test and Urea agar media followed by several biochemical tests. The identification of Enterobacter bacteria was carried out by Gelatin Agar test and Urea agar media followed by several biochemical tests and had been identified as motile, rod-shaped cells, some of which are encapsulated. Clostridium was identified as an anaerobic, Gram-positive, spore-forming rod that produces a potent neurotoxin by various tests like Gelatin Agar test and Urea agar media followed by several biochemical tests.

Preliminary Screening for the anti bacterial activity was performed using Agar Well diffusion method. The mentioned Chemical substances showed an excellent antibacterial activity against the bacteria Shigella, Enterobacter Species, Clostridium, E. coli, Vibrio, and Staphylococcus. As per the microbial susceptibility tests Gentamicin showed highest antibacterial activity against Shigella bacterium followed by citric acid, Naphthazarin, Norfloxacin, Streptomycin and less activity was observed for Azithromycin. Gentamicin showed highest antimicrobial activity against E.coli, followed by citric acid, Norfloxacin, Streptomycin and Naphthazarin. Least activity was observed by azithromycin antibiotic. For Vibrio bacterium, the highest zone of inhibition was showed by Gentamicin followed by citric acid, Naphthazarin and less activity was observed for streptomycin. Clindamycin, Azithromycin, Amphotericin-B, Cefixime, Norfloxacin, and Ampicillin showed no any activity against Vibrio Bacterium. Enterobacter species have no any effect upon the utilization of Azithromycin, Amphotericin-B, Cefixime, Norfloxacin, and Ampicillin. The concerned bacterium is adducted against these antibiotics. Gentamicin have a highest response against Enterobacter as per antimicrobial susceptibility is taken into consideration followed by citric acid, Naphthazarin and Clindamycin. Clostridium bacteria have no any effect against antibiotics like Cefixime, and Amphotericin-B. Ampicillin have less activity against Clostridium bacteria and highest antimicrobial susceptibility was observed for Gentamicin followed by citric acid Norfloxacin, Clindamycin, Naphthazarin, Azithromycin, Streptomycin. The Highest Zone of Inhibition was observed by Gentamicin against Staphylococcus, followed by Citric Acid, Naphthazarin, Streptomycin and Clindamycin. No any activity was observed by the use of Azithromycin, Amphotericin-B, Cefixime, Norfloxacin and Ampicillin antibiotics. Salmonella bacterium a strong bacteria and some antibiotics like Gentamicin, Naphthazarin and citric acid have some antimicrobial activity against salmonella. Rest of the used antibiotics like, Ampicillin, Norfloxacin, Cefixime, Amphotericin-B and azithromycin have no any activity against the salmonella bacterium.

Conclusion

From the above mention research it could be concluded that the spoiled chicken and mutton samples should not be consumed. Being rich in protein content the chicken and the mutton samples are frequently affected by various bacterial strains. Also from the collected chicken and mutton samples the samples of Halal type have been found to be less effective against the growth of micro-organisms as compared to the Jatka type samples, which could be due to different procedure of slaughtering technique. In case of Jatka the animal is being slaughtered at once and the blood gets coagulated within no time, but during the slaughtering process in which the animal is being slaughtered in a Halal way, makes whole blood to come out of the body of an animal. So as per this research it could be concluded that Halal type of flesh should be consumed than the Jatka type.

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