KLEBSIELLA PNEUMONIAE INFECTION IN BROILER CHICKENS

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ABSTRACT

The epidemiology, Pathogenciety and the possible ways of treatment of Klebsiella pneumoniae (K. pneumoniae) infection as a respiratory pathogen in broiler chicken farms in different places in Gharbia Gornorate were investigated during 2014/2017. For this purpose 150 samples were collected from lung, trachea, liver and intestine samples of 150 diseased, 1-5-week-old commercial broiler chickens suffered from respiratory disorders, diarrhea, growth retardation and mortality with pneumonia, pericarditis, airsaculitis and enlarged liver. Bacteriological examination of swabs from trachea, lung, liver and intestine revealed Gram negative, non-motile rod-shaped organisms which were commonly isolated from lung, liver, intestine and trachea. The isolation trials revealed 10 K. pneumoniae positive cases with an incidence (6.6%). Identification of the isolated strains Biochemically by API 20 E and molecularly by polymerase chain reaction (PCR) revealed that the isolates belonged to K. pneumoniae. The K. Pneumoniae isolate was inoculated experimentally in 7 day-old chicks and clinical signs, lesion were reproduced. Clinical signs & P.M lesions characteristic in K. Pneumoniae infection was reproduced when the K. Pneumoniae isolates was inoculated in 7 day old chicks. K. Pneumoniae was also reisolated. Clinicopathological features and
re-isolation of the organism from experimentally infected chicks were recorded. The results of the in vitro antibiotic sensitivity test revealed that the isolated strains were highly sensitive to gentamycin, amikacin, sulphamethoxazole–trimethoprim, ciprofloxacin and chloramphenicol, considerably sensitive to kanamycin, norfloxacin, oxytetracyclin and neomycin but resistant to Ampicillin and Erythromycin.

The clinical signs, post-mortem findings and the histopathological lesions of tissue sections from different organs of experimentally infected chickens were less severe after treatment with gentamycin, amikacin combined with sulphamethoxazole–trimethoprim in drinking water for 3 consecutive days.

INTRODUCTION

Poultry diseases is one of the most important segment of the agriculture sector in Egypt, Where investment in this industry is about 20 billion Egyptian pounds. It contributes a large part of the country’s supply of animal protein (White meat and eggs). This sector has faced serious challenges through the respiratory infection.

Enterobacteriaceae infection is still causing severe losses especially in young birds. Klebsiella pneumoniae is one of more than 40 genera that comprise the Enterobacteriaceae family which plays as human and animal respiratory pathogen. Klebsiella infection in poultry has been recently reported to cause high mortality in balady chicks (Sarakby, 1979 and Karaman, 1980).
Klebsiella pneumoniae infection is conveyed to the developing and very young chicks after hatching but adult birds act as a carrier (Sekariah and Seth, 1957).

The upper respiratory tract of healthy birds can harbor Klebsiella microorganism which can behave as opportunistic pathogens (Sandra and Duarte Carreia 1998) K. pneumoniae can cause localized or systemic infection in poultry and other birds (Shivaprasad, 1998), celliulitis in turkey and could be associated with primary contact dermatitis and skin abrasions. (Gomis et al., 2001).

*K. pneumoniae* infections in growing broiler chickens did not receive much care in our country in spite of considerable importance which are found to be implicated in poultry diseases resulting in significant losses, therefore the work reported in this study was undertaken to investigate the role of *K. pneumoniae* organism in causing disease in growing broiler chickens at Gharbia Governorate, and confirming the pathogenicity by experimental infections using the isolated organism in 3 day old chicks further more studying the in vitro sensitivity test of the isolated organism against different antibiotics followed by treatment trial of experimentally infected chicks.

**MATERIALS AND METHODS**

1. **Collected samples:**

Specimens were collected from 150 diseased, freshly dead and sacrificed broiler chicks suffering from diarrhea and sudden death obtained from 35 farms at different localities in El-Gharbia Governorate from 2014-2017 and individual cases subjected to clinical examination. Samples were subjected to clinical, post-mortem and bacteriological...
examinations. Under complete aseptic condition, loopfuls from liver, lungs, trachea and intestine were streaked on MacConkey’s agar and XLD agar. The inoculated plates were incubated micro-aerobically (10% CO₂) at 37°C for 48 hrs. The suspected colonies were picked up, purified and identified morphologically and biochemically using (API 20 E Micro Test Strip Bio-Merieux, France) according to Nawaz et al., (2006) and Kamran et al., (2014). Molecular identification of K. pneumonia suspected colonies were carried out using PCR.

2. Bacterial isolation:

Field strains of K. pneumonia isolated from diseased broilers were used for pathogenicity test by oral, intramuscular and subcutaneous injection (Wilki et al, 2000). The K. pneumonia isolates were grow on MacConkey’s agar plates for 48 hrs at 37°C micro-aerobically. A loopful of colonies was inoculated into brain heart infusion broth for serial ten-fold dilution of the organisms. Growth was adjusted by turbidity 10⁹ CFU/ml (Tatum et al., 2012).

3. Biochemical identification:

The biochemical identification of the isolated strains was performed using API 20 E strip (Bio-Merieux, Lyon, France).

4. Molecular detection (PCR):

The molecular detection of the isolated strains was performed by using PCR and the Amplification of DNA was performed on thermal cycler (Master cycler, Eppendorf, Hamburg, Germany) according to
Townsend et al. (1998). Primers were designed against (16S-23S rDNA) as previously described by Liu et al. (2008). Expected product size was 130 bp.

5. Experimental infection:

A total of 80, 1-day-old White Lohman chicks, free from Klebsiella pneumoniae infection were floor reared and fed on antibiotic-free ration. Feed and water were added ad libitum. Chicks were kept for one week before infection to insure that they were free from Klebsiella infection. Birds were randomly divided into 5 groups.

**Group (1):** of 10 chicks inoculated orally with 1 ml of sterile saline and kept as non-infected non-treated control group.

**Group (2):** of 10 chicks inoculated orally with 1 ml sterile saline (non-infected) and divided into 3 subgroups A, B, and C which treated with Gentamycine 0.2 mg/chick/day for consecutive days, Amikacin 15 mg/kg/day for 3 consecutive days and Sulpha-Trimethoprime 10/kg/day for 10 days, respectively.

**Group (3):** of 20 chicks were inoculated orally with 1 ml of broth culture containing $5 \times 10^9$ colony forming unit per ml (CFU/ml) and subdivided into 3 subgroups (A, B and C) treated with gentamycin, Amikacin and Sulphatrimethoprim, respectively with similar doses to group (2).

**Group (4):** of 20 chicks were intramuscularly infected with 1 ml broth culture contain $5 \times 10^9$ CFU/ml and subdivided into 3 subgroups (A, B and C) and treated similar to group (3).
**Group (5):** Of 20 chicks were subcutaneously infected with 1 ml broth culture contain $5 \times 10^9$ CFU and divided into 3 subgroup (A, B and C) and treated with gentamycin, Amikacin combined with Sulphmethoprime similar to group (4)

All chickens were kept under close observation for 3 weeks during which clinical signs and mortality were recorded. Dead and sacrificed chickens were subjected to postmortem and bacteriological examinations for re-isolation of the inoculated organism.

**Table (1): experimental infection**

| Group codes | Infection and treatment | Breed          | NO of birds | Inoculation and dose                                                                 |
|-------------|-------------------------|----------------|-------------|-------------------------------------------------------------------------------------|
| 1           | Non infected –non treated | White Lohman   | 10          | Inoculated orally with 1ml of sterial saline (Blank control)                        |
| 2A          | Non infected treated with Uccmagent | White Lohman | 3           | Inoculated injectable with 1 ml of sterial saline then give Uccmagent 0.2 mg /chicks once for 3 days (control –ve)3 |
| 2B          | Non –infected treated with Amikin | White Lohman   | 3           | Inoculated injectable with 1 ml of sterial saline then give Amikin 15mg/kg/day for 3 days (control –ve) |
| 2C          | Non –infected treated with Septazole | White Lohman | 4           | Inoculated orally with 1 ml of sterile saline then give Septazole40 mg/kg/day for 10 days (control –ve) |
| 3A          | Infected orally and treated with Uccmagent | White Lohman | 6           | Inoculated orally with 1 ml of 5×10^9after two weeks divided into 3 groups and NO 1 treated with Uccmagent |
| 3B          | Infected orally and treated with Amikin | White Lohman | 6           | NO 2 treated with Amikin and                                                        |
| 3C          | Infected orally and treated with septazole | White Lohman | 8           | NO.3 treated with Septazole.                                                        |
| 4A          | Infected I/Mand treated with Uccmagent | White Lohman | 6           | Inoculated I/M with 0.25 ml of 5×10^9 after two weeks divided into 3 groups and NO1 treated with Uccmagent |
| 4B          | Infected I/M and treated with Amikin | White Lohman | 6           | NO 2 treated with Amikin and                                                        |
| 4C          | Infected I/M and treated with Septazole | White Lohman | 8           | NO 3 treated with Septazole.                                                        |
| 5A          | Infected S/C and treated with Uccmagent | White Lohman | 6           | Inoculated S/C with 0.25 ml 5×10^9 after two weeks divided into 3 groups and NO1 treated with Uccmagent |
| 5B          | Infected S/Cand treated with Amikin | White Lohman | 6           | NO 2 treated with Amikin and                                                        |
| 5C          | Infected S/C and treated with Septazole | White Lohman | 8           | NO 3 treated with Septazole.                                                        |

6. Antibiogrmme:
The antibiotic sensitivity test of the isolates was investigated against 20 antimicrobial agents using the disc diffusion technique according to *Cruick-shank et al., (1975)*. The test procedure was that recommended by the National Committee for Clinical Laboratory Standards (1990).

**7. Treatment trials:**

Uccmagent (Gentamycin) (Uccma-pharm) 2-4mg/kg body weight. every 8 hours for 3 days, Amikin (each vial contain 250 mg amikacin sulfate) (Smithklin Beecham - Pharm), 15-20 mg/kg body weight for 3 days. and Septazole suspension (Sulfamethoxazole+ Trimethoprim) (Alexandria Pharm), 4--20 mg/kg body weight were used in drinking water for 5 consecutive days for treatment of experimentally challenged chickens with *K.Pneumoniae*.

**8. Histopathology:**

Specimens of lung, liver and intestine were taken from experimentally infected birds fixed in 10% neutral buffered formalin, washed, dehydrated in different concentrations of alcohols, cleared in xylol and embedded in paraffin wax by routine methods. They were then Sectioned at 4 μm, stained with haematoxylin and eosin (HE) stain, and examined by light microscopy (*Bancroft and Gamble, 2007*).

**9. Statistical analysis:**

Statistical analysis of the obtained data was carried out according to *Petri and Watson (1999).*

**RESULTS AND DISCUSSION**
Ten isolates suspected to be *K. Pneumoniae* were isolated from 150 diseased broiler chickens with (6.6) isolation rate. Table (2).

Examined chickens suffered from respiratory disorder, septicamiemia, peritonitis, salpingitis, air sac diseases, omphalitis, artheritis, panophthalmitis and intestinal disturbances. Such diseases cause great economic losses in poultry industry not only due to high mortality rate in young bird, slow growth and poor feed conversion rates in growing birds but also due to decrease in egg production and hatchability of the infected eggs. *Plessor et al., (1975); Mahalingam et al., (1988) and Rennie et al., (1990).*

The prevalence of *K. Pneumoniae* isolated from various internal organs of examined chichs was described in Table (3). The isolation rate was higher in lungs (60%) than the liver (40%) and the lowest rate of isolation was from the intestine (10%). Similar results were reported by *Gylstorff and Gerlach (1974) and Buxton and Fraser (1977).*

*K. Pneumoniae* isolated from internal organs of diseased chickens on MacConkey agar, were Gram negative bacilli, non-spore forming, capsulated, non-motile and arranged singly . Colonies appear lactose fermenting, dome shaped, 3-4 mm diameter after overnight incubation at 37° C. Fig. (1). Similar results were obtained by *Mona Mohammed Aly (2014).*

The biochemical activity of the recovered isolates were typed to that of *K. Pneumoniae* and described in Table (4). The isolates were negative in motility, Indole test was variable, Methyle red and H$_2$S production were negative, Vogas proskuaer, Citrate utilization, Urease
and Sugar fermentation were positive, Similar results were reported by *Kawakib Ibraheem Al Zubaid* (2009).

*Molecular identification by using PCR revealed that 10 isolates were positive and amplified at 130 bp Fig. (2).* Similar findings were obtained by *Fang et al. 2004; Ku et al. 2008 and Cheng et al. 2010*. Also *Yu et al. (2007)*.

The experimentally infected, 7-day-old broiler chickens suffered from decreased body weight gain, ruffling of the feather, progressive weakness and prostration. Respiratory symptoms appeared on the second day after infection and were manifested by gasping, abnormal breathing while intestinal troubles were represent by yellow-greenish diarrhea. The mortality rate ranged from 15-20%. Table (5). Similar results were reported to *Abd-Alla (1981)*.

The post-mortem lesions of dead and sacrificed birds revealed hyperemia of the lunge, congestion of the liver, spleen, enlargement of the gall bladder and sometimes enteritis. Fig. (2). the organisms were re-isolated from lung and liver. These results were similar to that reported by *Ann Moursy et al (1982) and Dessouky et al. (1982)*.

The results of the in vitro antibiotic sensitivity test revealed that all tested *K.pneumoniae* isolates were highly sensitive to gentamycin, Amikacin and Sulphamethoxazal, moderate sensitive Ciprofloxacine and Chloramphenicol and low sensitive to Noreflaxacine, Neomycin and Oxytetracycline but resistant to Cefotaxim, Amoxacillin and Cephalothin. Table (6), (7). These results were similar to that obtained by *Sundaresan et al. (2007)*.
Treatment trials:

The results of treatment trial of experimentally infected chicks were described in table (8). The clinical signs and post-mortum lesion were improved and mortality were disappeared in experimentally infected chicks and treated with Gentamycin 2-4 mg/kg. Body weight for three days, Amikacin 15/20 mg/kg. Body weight for three days and Sulphamethoxazol-trimethoprim 40mg/kg+ 8 mg/kg for three days.

In this study, Klebsiella isolates showing 20% resistance to amikacin. A low prevalence of amikacin resistance (7%) to Klebsiella spp. isolated from meat samples were also reported by Gundogan et al. On the other hand, Ullah et al. (2009) reported that 63.04% of Klebsiella isolates were susceptible to amikacin.

Klebsiella strains recorded high antibiotic resistance with multiple antibiotic resistance (MAR). Rate of multiple antibiotic resistances was extremely high which may be due to the hazard routinely use antibiotics for treatment and control of bacterial diseases in poultry farms. When these antibiotics are administered to the birds at low levels for a long period, certain bacterial species become resistant (Kilonzo-Nthenge et al. 2007). These antibiotic-resistant bacteria can reach to human through consumption of food products from animal origin and by direct contact (Van den and Stobberingh, 2000). Our finding is in accordance with a previous literature reported by Davies et al. (2016) who reported a 25% multiple drug resistance of K. pneumoniae isolates from psittacines. Also Bonnedahl et al. (2014) detected 13% MDR strains from the samples they collected from ageese and free-living gulls in Alaska, USA.
Histopathological examination of lungs of experimentally infected chickens revealed diffuse interstitial pneumonia characterized by infiltration of monocular cells and hyperplastic bronchial epithelium Fig. (a), the liver of experimentally infected chickens showed vacuolar degeneration and sporadic necrotized hepatocytes Fig. (b) and the intestine showed necrosis, destruction and infiltration of mononuclear cells in the intestinal lumen Fig.(g).

The lung of experimentally infected chicken with *K. Pneumoniae* and treated with Gentamycin showed few areas with mild interstitial pneumonia and congestion of the large blood vessels. Fig. (d). The liver of experimentally Infected chicken with *K. Pneumoniae* and treated with Amikacin showed necrotic foci and mild mononuclear cell infiltration in the parenchyma Fig. (h). Similar results were obtained by *Eman (1998)*.

The treatment improved the infected cases by different degrees, the best results obtained by gentamicin in which all organs seemed to be normal followed by Amikacin and sulphamethoxazol which leave mild effect on tissues on infected birds neither clinically nor histopathologically.

**Table (2): Results of Klebsiella Pneumoniae isolation**

| Total No. of chicks samples | Klebsiella infection | Non klebsiella infection |
|-----------------------------|----------------------|--------------------------|
|                             | No. | %    | No. | %       |
| 150                         | 10  | 6.6  | 140 | 93      |

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Table (3): Prevalence of *Klebsiella Pneumoniae* isolation in various organs of the examined chicks

| Organs      | Total NO. of examined | NO. of the isolates | Precentage of the isolation (%) |
|-------------|-----------------------|----------------------|---------------------------------|
| Lungs       | 150                   | 6                    | 4%                              |
| Livers      | 150                   | 3                    | 2%                              |
| Intestine   | 150                   | 2                    | 1.3%                            |

Fig. (1): *Klebsiella Pneumoniae* on MacConkey agar (lactose fermenter colonies).

Table (4): Biochemical tests for identification of *K.pneumoniae*:

| Biochemical test             | *K.pneumoniae* |
|------------------------------|----------------|
| Motility                     | -              |
| Indole                       | V              |
| Methyle red                  | -              |
| VogesProskuaer               | +              |
| Citrate utilization          | +              |
| Urease                       | +              |
| H2S                          | -              |
| Nitrate reduction            | V              |
| Gelatin liquefaction         | -              |
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| Biochemical test | K. pneumoniae |
|------------------|---------------|
| ODC              | -             |
| LDC              | +             |
| Arginine dihydrolase | -        |
| ONPG             | V             |
| Sugar fermentation |             |
| Lactose          | +             |
| Sucrose          | +             |
| Dulcitol         | V             |
| Salicin          | +             |
| Arabinose        | +             |
| Inositol         | +             |
| Xylose           | +             |

(+): Most strain positive.
(-): Most strains negative.
(v): Some strains positive, other negative, Variable.

Fig. (2): Agarose gel electrophoresis of PCR of 16S-23S rDNA "ITS" (130 bp) specific for detection and identification of Klebsiella pneumoniae

Lane M: 100 bp ladder as molecular size DNA marker
Lane C+: Control positive K. pneumoniae for 16S-23S ITS.
Lane C-: Control negative.
Lanes from 1 to 9 & 11: Positive K. pneumoniae strains.
Lane 10: Negative K. pneumoniae strain.
**Table (5):** Result of experimental infection of 7 - day - old broilers with *K.pneumoniae*.

| Group No. | No. of infected birds | Death time | Mortality |
|-----------|-----------------------|------------|-----------|
|           |                       | No. of dead chicks at different intervals post-infection/ day |           |
|           |                       | 2 | 3 | 4 | 7 | 9 | 13 | 14 | 16 | 18 | 20 | 21 | No. | % |
| 1         | 10                    | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 2         | 10                    | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 3         | 20                    | 2 | 0 | 0 | 0 | 1 | 0 | 1 | 0 | 0 | 1 | 1 | 6 | 30 |
| 4         | 20                    | 2 | 2 | 1 | 1 | 0 | 1 | 1 | 0 | 1 | 1 | 2 | 12 | 60 |
| 5         | 20                    | 3 | 0 | 1 | 1 | 1 | 0 | 1 | 0 | 0 | 1 | 1 | 9 | 45 |

**Fig. (3):** Experimentally infected chickens with *K.Pneumoniae* showing the clinicopathological features of the disease, diarrhea and pasty vent were observed in chicks and congestion of the lung.

**Table (6):** Results of in vitro antibiotic sensitivity test of *K. pneumoniae* isolates against different 20 antimicrobial agents.

| Antimicrobial agent     | S |   | I |   | R |   |
|-------------------------|---|---|---|---|---|---|
|                         | NO| % | NO| % | NO| % |
| Cephalothin (CN)        | - | - | - | - | 10| 100|
| Amoxicillin(AMX)        | - | - | 1 | 10| 9 | 90 |
| Erythromycin (E)        | 1 | 10| 2 | 20| 7 | 70 |
Table (7): Antimicrobial resistance profile of *K.pneumoniae* strains (n=10).

| NO | Strain          | Antimicrobial resistance profile                     | MAR index |
|----|----------------|------------------------------------------------------|-----------|
| 1  | *K.pneumoniae*  | CN, AMX, E, CF, AM, T, NOR, C, N, CP, K, SXT, AK, G | 1         |
| 2  | *K.pneumoniae*  | CN, AMX, E, CF, AM, T, NOR, C, N, CP, K, SXT, AK    | 0.928     |
| 3  | *K.pneumoniae*  | CN, AMX, E, CF, AM, T, NOR, C, N, CP, K, SXT        | 0.857     |
| 4  | *K.pneumoniae*  | CN, AMX, E, CF, AM, T, NOR, C, N, CP                | 0.714     |
| 5  | *K.pneumoniae*  | CN, AMX, E, CF, AM, T, NOR, C, N                    | 0.643     |
| 6  | *K.pneumoniae*  | CN, AMX, E, CF, AM, T, NOR                           | 0.500     |
| 7  | *K.pneumoniae*  | CN, AMX, E, CF                                     | 0.286     |
| 8  | *K.pneumoniae*  | CN, AMX                                            | 0.143     |
| 9  | *K.pneumoniae*  | CN, AMX                                            | 0.143     |
| 10 | *K.pneumoniae*  | CN                                                  | 0.071     |

Average 0.529
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CN: Cephalothin  AMX: Amoxicillin  E: Erythromycin  CF: Cefotaxim
AM: Ampicillin  T: Oxytetracycline  NOR: Norfloxacin  C: Chloramphenicol
N: Neomycin  CP: Ciprofloxacin  K: Kanamycin  SXT: Sulphamethoxazol
AK: Amikacin  G: Gentamicin

Table(8): Treatment trials of experimentally infected broilers with *K. Pneumoniae*.

| Group No. | No. of birds | Treatment                  | Drug used                          | Mortality |
|-----------|--------------|----------------------------|------------------------------------|-----------|
|           |              |                            |                                    | No.  %    |
| 1         | 10           | Non Infected +Non treated  | -                                  | 0          | 0         |
| 2         | 10           | Non Infected + treated     | Uccmagent+ Amikin+ Septazole       | 0          | 0         |
| 3         | 20           | Non Infected + treated     | Uccmagent+ Amikin+ Septazole       | 6/20 30%  |
| 4         | 20           | Non Infected+ treated      | Uccmagent+ Amikin+ Septazole       | 13/20 65% |
| 5         | 20           | Infected + treated         | Uccmagent+ Amikin+ Septazole       | 9/20 45%  |

Fig. (a): Lung of a chick Infected orally with 1ml broth culture contain $5 \times 10^9$ CFU/ml. of *K.*

*Pneumoniae* (group3) showing hyperplastic bronchial epithelium (H&E, X100).
Fig. (b): Liver of a chick Infected orally with 1ml broth culture of *k. Pneumoniae* contained $5 \times 10^9$ CFU/ml. (group3) showing vacuolar degeneration and sporadic necrotized hepatocytes (H&E, X400).

Fig. (c): Liver of a chick Infected orally with 1ml broth culture of *K. Pneumoniae* contained $5 \times 10^9$ CFU/ml. (group3) showing infiltration of monocular cells and sometimes neutrophils in the portal areas (H&E, X400).
Fig. (d): Lung of a chick orally Infected with 1ml broth culture of *K. Pneumoniae* contained $5 \times 10^9$ CFU/ml. and treated with Gentamycin (group 3A) showing few areas with mild interstitial pneumonia and congestion of the large blood vessels (H&E, X40).

Fig. (e): Lung of a chick intramuscularly Infected with 1ml broth culture of *K. Pneumoniae* contained $5 \times 10^9$ CFU/ml. (group 4) showing diffuse interstitial pneumonia characterized by infiltration of monocular cells in the interstitial tissue (H&E, X100).
Fig. (f): Trachea of a chick infected intramuscularly with 1ml broth culture of <i>K. Pneumoniae</i> contained $5 \times 10^9$ CFU/ml. (group 4) showing trachitis, desquamation of tracheal epithelium with sub-epithelial edema and infiltration of mononuclear cells (H&E, X40).

Fig. (g): Intestine of a Chick intramuscularly infected with 1ml broth culture of <i>K. Pneumoniae</i> contained $5 \times 10^9$ CFU/ml. (group 4) Showing necrosis, destruction and infiltration of mononuclear cells in the intestinal lumen (H&E, X40).
Fig. (h): Liver of a chick intramuscularly infected with 1ml broth culture of *K. Pneumoniae* contained $5 \times 10^9$ CFU/ml and treated with Amikacin (group 4B) showing necrotic foci and mononuclear cell infiltration in the parenchyma (H&E, X400).

Fig. (i): Intestinal glands of a chick intramuscularly infected with 1ml broth culture of *K. Pneumoniae* contained $5 \times 10^9$ CFU/ml and treated with Amikacin (group 4B) showing destructed, degenerated and shrinked...
intestinal glands with mononuclear cell infiltration in the lamina propria (H&E, X40).

**Fig. (j):** Intestine of a Chick subcutaneously Infected with 1ml broth culture of *K. Pneumoniae* contained $5 \times 10^9$ CFU/ml (group 5) showing necrosed, destructed and shrinked intestinal villi (H&E, X40).

**CONCLUSION**

This study spotlight the prevalence and incidence of *Klebsiella Pneumoniae* in clinically diseased chicken suffered from respiratory manifestation. The study confirmed the pathogenic potential of the isolated strains and their association with clinical manifestations in respiratory tract infections of broiler chicken. Antimicrobial susceptibility pattern showed high multiple antibiotic resistances which require strict regulations of the use of antibiotics in veterinary therapy to minimize the emergence of resistant bacteria in chickens which may increase the public health problem.

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...نسبة 6.6%.

تم عمل العدوى الاصطناعية لمعزولات الكيبسيلا وتم تسجيل الأعراض الإكمينيكية والصفة التشريحية ونسبة النفوق وإعادة عزل الميكروب المحقون.

أوضحتي النتائج أن العجزات المعزولة كانت شديدة الحساسية لكل من (الجنتاميسين، الأميكانسين سلفاميساكرزول)، ونورفوكوناسين والكلورامفينكول وقيل الحساسية للنيوميسين، سيفاميساكرزول والأوكسي تتراسيكلين ولكنه مقاوم أموكسيسيمن، سيفاميساكرزول وسيفاميساكرزول.

بينما تم علاج الدجاج المعدي اصطناعيا بحقن جنتاميسين والأمكين لمدة ثلاثة أيام متتالية وإعطاء سلفاميساكرزول في ماء الشرب في نفس الوقت. لوحظ اختفاء الأعراض الإكلينيكية والصفة التشريحية المميزة للمرض.

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