

Field Survey of Important Causes (Viruses, Bacterial, and Fungi) Agents Poultry in Tikrit City

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ABSTRACT

Keywords:

Field, Survey, Causative agents. Poultry, diseases.

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The Study have been arranged to identify the mainly popular agent's disease poultry which received via field consultants outside Tikrit urban during 1/1-31/12/2016.

A total 250 sick with death chickens collected 5 different fields. Diagnosis cases according to clinical times past, clinical symbols and postmortem examination while; confirmatory diagnosis via cultural media, Grams stain, biochemical examines, Lacto phenol cotton blue and ELISA.

In this study out of 120,40(33.3%) chickens were clinically diagnosing as *colibacillosis*, 60(50%) as *Salmonella spp* and 20 (16.7%) *Staphylococcus aureus*. In the same way, out of 90 chicken, 27(30%) were diagnosed an *Infectious bronchitis* (IB), 25(27.8%) *Infectious laryngotracheitis* (ILT), 23 (25.6 %) *Infectious Bursal Disease* (IBD), 8(8.9 %) as *Newcastle virus* and 7(7.8%) *Reo virus*. whilst fungi culture showed 25(62.5%) *Aspergillus Fumigatus* and 15 (37.5%) *Candidiasis*. antimicrobial susceptibility test isolated bacteria was done to establish susceptibility here.

مسح حقلي لاهم المسببات المرضية (الفيروسية، البكتيرية والفطرية) للدواجن في مدينة تكريت

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الخلاصة

اجريت هذه الدراسة لمعرفة معظم المسببات المرضية الشائعة والمستلمة بواسطة مستشاري الحقول في مدينة تكريت خلال المدة من 1/1 - 31/12/2016.

اشارت النتائج ل 250 عينة دجاج مريض او ميت جمعت من خمس حقول مختلفة للدواجن. تم تشخيص الحالات وفقا لتاريخ حاله السريرية العلامات السريرية، اجراء الصفة التشريحية لطيور المصابة وبعدها اكدت باستخدام الاوساط الزرع، صبغه كرام، الاختبارات الكيموحيوية المختلفة، Lacto phenol cotton blue ، كتأت الاليزا، واختبار فحص الحساسية للجراثيم.

في هذه الدراسة من مجموع 120 دجاجة 40(33.3%) شخصت سريريا *colibacillosis* و60(50%) *Salmonella spp* و20 (16.7%) *Staphylococcus aureus*. وبنفس الطريقة من مجموع 90 دجاجة 27(30%) شخصت سريريا كفيروس التهاب القصبات المعدي و25(27.8%) كفيروس التهاب الحنجرة والرغامى المعدي و23(25.6%) كفيروس التهاب غده فايبرشيا المعدي و 8 (8.9%) كفيروس النيوكاسل و7(7.8%) كفيروس التهاب المفاصل. بينما اشارت نتائج فحص الفطريات انه من مجموع 25 دجاجة (62.5%) شخصت سريريا ك *Aspergillus spp* و15(37.5%) *Candidiasis*. تم اجراء اختبار فحص الحساسية للجراثيم المعزولة لمعرفة حساسيتها للمضادات الحيوية.

الكلمات المفتاحية:

مسح، حقلي، المسببات

المرضية، الدواجن.

للمراسلة:

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Introduction:

In the last half century, significant increases in the productivity of modern poultry stocks have been achieved for both the meat and the egg production sectors of the global poultry industry. Synergies have resulted from advances made in all the major activities of poultry management and housing, nutrition and ration formulation, applying poultry genetics knowledge in commercial breeding programs and better diagnosis and control of avian diseases. Of all these core elements, poultry health and disease can be the least predictable (Pattison et al., 2008)

Although poultry diseases from nutritional and metabolic causes can be of concern, the emphasis in this information note is on controlling diseases that are caused by infectious agents, which can exert damaging – and sometimes immediate – negative effects on the profitability of commercial operations. The development of an intensive poultry industry in many of the countries discussed here depends on the growth in number and size of small and medium-sized commercial poultry operations. The emphasis in this review is therefore primarily on optimizing poultry health for this scale of operations. Because of the importance of small-scale village-based production units in many developing countries, however, the poultry health implications for and such flocks are also included (Jalloob *et al.*, 2009).

Poultry that have an infection show a variety of symptoms, such as respiratory problems, diarrhoea and paralysis. It should be emphasized at the outset that prevention of infection in a poultry flock through sound management is very important. This is because although some infectious diseases can be treated, for many it is a waste of time and money and infected birds should be disposed of immediately. (Janmatt *et al.*, 2011)

Poultry can be affected by many types of disease, and a broad variety of pests (and behavioral problems) including Viruses, Mycoplasma, Bacteria, Fungi, Protozoa, Internal Parasites, External Parasites, Metabolic disorder, environmental factors (Jaswinder *et al.*, 2004).

Although the relative importance of poultry diseases may differ between countries and geographical areas, there are few important diseases that are unique to particular parts of the world, at the global level, however, differences in distribution among regions are now apparent, because genetic variants have emerged within some of the major specific pathogens of chickens. This has become important for attempts to prevent the spread of virulent strains through international movements of poultry products (Shane , 2004).

Material and Methods:

The research was carried out 1/1/ - 13/12/2016 in microbiology laboratory in College of Veterinary Medicine. Using examination methods state history, appearance signs, postmortem examine while certain tests include cultural characteristic, Grams stain, biochemical test, Lacto phenol cotton blue, ELISA, and sensitivity tests. Specimens were collected infected chicken liver, lung, heart, spleen, intestinal contents, tracheal swab, heart blood. Blood samples (5ml) were obtain from each chicken from wing vein-puncture. sera sample be stored at -20c formed serological analysis. Cultural and morphological characteristic of the isolated colonies fungi and bacteria described by (Buxton and Fraser 1977),distinguish motile and non-motile bacterial isolated described by (Ryan and Ray 2004).

A- TSI gar:

Determine Sugar fomenters and also the bacteria which produce H₂S. The isolate bacteria were growth over the surface of the slants and bottom following incubate for 24 hr at 37°C.

B- Fermentation test:

This test used to distinguish between types of the bacterial which ability of ferment a wide groups from sugars and others ferment only a few described by (Ryan and Ray 2004)

C-Catalase and coagulase examinations:

Catalase and Coagulase tests use distinguish isolated bacterial consistent with method described by (Cheesbrough,2006). Coagulase examine specimens were confirmation consistent with method (Cowan and Steels 1985).

D-Indole, MR and VP tests:

Distinguish the isolation bacterial described by (Cowan and Steels, 1985).

E-Agar slant:

The bacterial was isolated inoculate into the brain heart infusion slop agar to preservation them (Cowan and Steels 1985).

F- Elisa Kits.

Identification of virus were used ELISA Kits for detection Abs in samples according to manufactured instruction by Biochek Smart Veterinary Diagnostics.

Sensitivity test:

The test carried out in order to identify the most effective drugs to treat. transport Small part bacterial colony to Mueller-Hinton broth tubes and incubated at 37°C for 24 h. Then antibiotic diffusion inside plate via forceps and incubated at 37°C for 24 h. zone inhibition measured millimeter as sensitive, intermediate and resistant to different antibiotics (Quinn et al., 2010).

Results:

Sum 250 chickens chosen study 120(48%) found positive bacteria growth, 90 (36%) positive virus analysis and 40(16%) positive fungi growth in chickens Table (1).

Table 1. popularity of pathogenic organism in chickens

	pathogenic organism	No of chickens	
		Number	%
1-	bacterial isolated	120	48
2-	Viral analysis	90	36
3-	Fungal growth	40	16
Total		250	

Clinical diagnosis diseases.

1- Colibacillosis

About 40 (33.3%) of the suspected chickens were diagnosed as colibacillosis. The most obvious clinical signs were diarrhoea, depression, soiling of cloaca with semisolid cheesy material, respiratory distress (coughing, sneezing), reduced egg production, loss of condition and death. Recorded postmortem lesions were omphalitis and fluid accumulation in the peritoneal cavity of chickens, dark-colored swollen liver and spleen, fibrinopurulent airsacculitis, pericarditis, perihepatitis hemorrhagic enteritis with fluid accumulation in ligated intestinal loops and diarrhea.

2-Salmonellosis

Out of 60(50%) suspected chickens were clinically diagnosed as *Salmonellosis*. Most common clinical signs of *Salmonellosis* were drowsiness, huddled together, poor growth, chalky white diarrhoea with pasted vent, dehydration, reduced egg production and death .After necropsy the gross lesions were observed as peritonitis, unabsorbed yolk ,discrete, small, white, necrotic foci in the liver which became swollen and fragile with distinctive coppery bronze sheen on the surface ,turbid yellow color fluids in the peritoneal cavity and irregular, haemorrhagic ova with prominent thicken stalks.

3-Staphylococcus aureus

Sum 20(16.7 %) suspected chickens were clinically diagnosing *Staphylococcus aureus* the majority common clinical symbols of *Staphylococcus aureus* infections appeared in three forms—septicemia (acute), arthritic (chronic), and bumble foot.

4-Infectious Bronchitis:

Sum 30(33.3%) suspected chickens were clinical diagnose Infectious Bronchitis mainly common clinical signs of chick may cough, sneeze, and tracheal rales, Conjunctivitis and dyspnea. lesions sinusitis, nasal passageway full exudates, and air sacs some foamy exudates initially, progressing to cloudy thicken.

5-Infectious Laryngotracheitis (ILT):

Sum25(27.8%)suspected chickens were clinically diagnosed as *Infectious Laryngotracheitis* (ILT) Most common clinical signs of Chickens may be in acute infections, there may be nasal discharge, moist rales, coughing and gasping. The neck may be extended (“pump handle respiration”). The eyelids can be red with increased ocular drainage. In severe cases there is marked labored breathing and coughing of blood-stained mucus, which covers the wing and breast feathers as a result of the bird shaking the head with violent coughing. Blood and yellow exudate in the trachea cause death by suffocation. In mild cases, after gross lesions are in the trachea and larynx. In severe cases, the trachea contains clotted blood. White fibrinonecrotic plugs can fill the glottis and trachea.

6-Infectious Bursal Disease:

Sum 23 (25.6 %) suspected chickens were clinically diagnosed as *Infectious Bursal Disease*. Most common clinical signs of Chickens may depression and ruffling of feathers, poor or lack of appetite huddling, unsteady gate, reluctance to rise, and diarrhea (sometimes, bloody). Immuno suppressed survivors may be affected with other disease agents, resulting in various secondary infections that can end in death, or manifest as respiratory or gastrointestinal disease. after post-mortem lesions (identified upon autopsy) indicative of IBDV infection. The bursa of Fabricius is the main organ affected, showing swelling from edema and hemorrhage during the early stages of the disease and then shrinking (atrophy) 7-8 days following infection. Bleeding in the breast and thigh muscles may be noted due to impaired blood clotting.

7-Newcastle Disease:

Out of 8(8.9%) suspected chickens clinically diagnosed Newcastle Disease. Most frequent clinical symbols of Chicken may be Sudden dead, depression, In appetite, Coughing, Diarrhoea, Nervous symbols, paralysis, abnormal neck and Molt. lesion Airsacculitis, Necrotic plaques in proventriculus, intestine, caecal tonsil, bleeding in proventriculus.

8-Reo virus (viral arthritis):

Sum 7(7.8%) suspected chickens clinical diagnosed as Reo virus (viral arthritis) mainly common Clinically. The manifested lameness and swelling affecting primarily tarsometatarsal joints and the feet. after post-mortem assessment of affected birds, lesions discrete distension and oedema of the tendons, Swelling and collection of inflammatory fluid, Unilateral femoral head necrosis occurs in one cockerel, at the same time suffer tenosynovitis.

9-Aspergillosis:

Sum 26 (65%) suspected chickens were clinical diagnose Aspergillosis. Most frequent Clinically manifested by Gasping and quick breathing. gross lesions involve the lungs and air sacs primarily. Yellow-white pin head sized.

10-Candidiasis:

Out of 14 (35%) supposed chickens clinical diagnosed candidia . most common Clinically it is manifested by present in only severely affected birds as depression, retardation of growth, stunted appearance, reduced food intake ruffling feathers, listlessness, diarrhea, may drooling mucus exudates of offending odour from mouth. Drop of egg production in layers, pendulous crop in some of the affected birds may occur, when candidiasis occurs as a secondary infection, the lesion appears grossly as thickening of the mucosa with raised multifocal (circular) to confluent whitish cheesy material often there are pseudo or diphtheritic membranous necrotic patches that are peeled easily from the eroded mucosal surface sloughed ulcer formation may occur

laboratory diagnosis:

Result showed with 120 bacterial isolates belonging to 5 species, 90 virus belonging to 5 species and 40 fungi belonging 2 to species (Table 2).

Table 2. Causative agent Isolated from different samples

	Caustive diseases	Number	%
1-	<i>Escherichia coli</i>	40	33.3
2-	<i>Salmonella pullorum</i>	30	25
3-	<i>Salmonella paratyphoid</i>	20	16.7
4-	<i>Salmonella gallinarum</i>	10	8.3
5-	<i>Staphylococcus Spp</i>	20	16.7
Sum		120	
6-	<i>Infectious Bronchitis(IB)</i>	27	30
7-	<i>InfectiousLaryngotracheitis (ILT).</i>	25	27.8
8-	<i>Infectious Bursal Disease(IBD).</i>	23	25.6
9-	<i>Newcastle Disease(ND).</i>	8	8.9
10-	<i>Reo virus</i>	7	7.8
Sum		90	
11-	<i>Aspergillosis Spp</i>	25	62.5
12-	<i>CandidiasisSpp</i>	15	37.5
Sum		40	

1-Colibacillosis:

Smooth, large, rounded colonies blood agar, pink color MacConkey agar, greenish-yellow color on BGA, green sheen on EMB agar, pink color S.S agar and yellow color slope with gas in bottom TSI agar

Gram's stain and motile examine:

Bacterial Isolated G- rod shape, set in single, pairs or short chains. the test organisms were motile.

Biochemical examination:

Bacterial isolates produced acid and gas via sugar fermentation. microorganisms positive methyl red and negative voges-proskauer reactions H₂S wasn't formed but Catalase and Indole test positive.

Table 3. Biochemical examination

microorganism	Sugar fermentation					MR	VP	Indole	Catalase	H ₂ S
	Dex	Mal	Lac	Suc	Man					
<i>E.Coli</i>	Acid/gas	Acid/gas	Acid/gas	Acid/gas	Acid/gas	+	-	+	+	-
<i>Salmonella Spp</i>	AG/A	-/A	-	-	AG/A	+	-	-	+	+
<i>Staphylococcus spp</i>	-	+	+	+	+	-	-	-	+	-

2-Staphylococcus aureus:

The organism grow on different culture media formed turbidity in nutrient broth; around, flat, golden yellow color Mannitol salt agar, whitish, flat colonies gamma hemolysis blood agar.

Gram's staining ,Catalase and coagulase activity test.

The bacterial isolate G+ cocci, arranged grape like clusters ,positive coagulase and catalase test .

Biochemical examinations:

Bacterial isolate sugar fermentation and produced acid and gas ,negative methyl red , voges-proskauer without produce H₂S .

3-Salmonellosis.

Bacterial growth in different cultures broth and produced turbidity, round, smooth, and transparent NA, colorless with dark center S.S agar, colorless EMB and TSI agar slope *Salmonella gallinarum* formed dark color with yellow slope while *Salmonella pullorum* produced white colony red slant.

Gram's staining and motile examination:

G- short, straight rods. *Salmonella gallinarum* and *Salmonella pullorum* non-motile.

Biochemical examinations:

lactose and sucrose non fermentation while fermented dextrose and mannitol produce acid and gas. *Salmonella gallinarum* created only acid while *Salmonella pullorum* acid and gas. total isolates bacteria indole, VP negative and methyl red positive.

4-Aspergillosis:

The organisms isolated on sabaroud dextrose agar black-green dusty. Lacto phenol cotton blue observe conidiophores' smooth, pale to light green close to vesicle, enlarge form a flask- shaped vesicle.

5-Candidiasis:

Organisms isolated growth on sabaroud dextrose white to cream color, flat, glabrous. Under microscopic sphere-shaped budding yeast-like cells.

Antibiotics sensitivity test:

Sum (120)bacterial isolated result shown on *Escherichia coli* and *salmonella* spp are sensitive Ciprofloxacin, Sulphanomide, Amoxcyline and Tetracyline, intermediate Neomycin and Resistant penicillin and Erythromycin. whereas *Staphylococcus aureus* sensitive Ciprofloxacin penicillin and Erythromycin, moderately Tetracyline and Resistant to Sulphanomide, Amoxcyline.

Table4. antibiotics sensitivity tests.

Bacterial isolate	Total	Ciprofloxacin	Sulfamide	Amoxicillin	tetracycline	Neomycin	penicillin	Erthyo- mycin
<i>Escherichia coli</i>	90	S	S	S	S	I	R	R
Sal. Spp	80	S	S	S	S	I	R	R
<i>Staphylococcus aureus</i>	18	S	R	R	I	R	S	S

T, Tetracycline(30µg); Sul, Sulfamide, (25µg); AMO, amoxicillin(10µg); P, penicillin(10IU); Cip, Ciprofloxacin(5µg); E, erythromycin(30µg); Neo, Neomycin, (30µg).

Discussion:

This type of study is regarded as a introduction survey in Tikrit province, although a number of previous studies have been made in other governorates (Jallob,2009and Mostafa etal., 1968).

The present study showed that bacterial diseases are the most prevalent one 120(48%) and this agreed with other observations (Mostafa etal.,1968). Diseases occurs due to lack of proper care and organization, insufficient nutritious feeding and some added factors.

The percentage of Salmonellosis and Colibacillosis represent the highest level 50%,33.3% respectively. Like types of findings were described by (Chauhan,2003;Rakibul etal., 2010;Lutfu, 2010 and Butcher,2015)

The study has been concluded that viral diseases constitute the second main line in problems that thread poultry industry. (Table2) showed that IB create a terrible sense for the poultry manufacturing, because of the mortality that are attributed to secondary bacterial infection that will go with the actual cause create further complication for the course of disease give high mortality that may exceed 80%. The disease cannot be overcome by vaccination processes owing to the presence of several serotypes which lack the cross protection or cross- neutralization. These findings were supported by Igajatovic(2010).

Infectious laryngotracheitis (ILT) is other chief problem among viral diseases (Table2). is a highly contagious respiratory disease in poultry that has been recognized in most countries about the world and remnants a threat to the intensive chicken industry. The first Australian outbreak of ILT was recorded in NSW in 1935, and the disease has since been reported in all other States of Australia. These findings supported by Kirkpatrick etal., (2014).

IBD is other severe problem in poultry disease. Its incidence was 25.6% (Table2) with a mortality rate of 10- 30%. The importance of this disease is its effect on the immune system, where immune suppression has been recognized as a significant sequel to the disease. These findings supported by (Daral etal., 2014).

Newcastle disease is other main problem between viral diseases (Table2). Some birds have get the infection, although they were vaccinated two or three times in different routes with high mortality reaching up to 90%. This require to re-evaluate the dose and to give a special attention for the local strain. These findings supported by (Dennis ,2014).

Reo virus have been observed with 7.8%. It seem that the greatest term used is infectious stunt syndrome because of the poor growth of the affected chicks .No available vaccine at a time, since the real causal agent is not confirmed, it may be caused by calicivirus, enterovirus, parvovirus, reovirus or togovirus ,the chief problem with the disease that stunted chicks are given sufficient feed and extra time, but feed change rate are poor ,The stunted chickens may be uneconomic to process and will create problems with the abattoir machinery as they are too small, in such cases it may be most excellent to slaughter them as they are caught. These findings supported by Jallob(2009).

The study has been concluded that fungi diseases make up the third main line in problems that thread poultry manufacturing. (Table2) showed that Aspergillosis and Candidiasis generate disease in two ways viz. produce pathogenic symbols and lesions of disease by invade, harm and destroying body tissues of the host; and by producing some toxins known as mycotoxins (aflatoxins, ochratoxins, ergot, fusarium toxins etc.) in food grain and feed during crop production, harvesting and storage steps, the intake, use and subsequent intoxication of which produce disease, immunosuppressive situation and hampers production potential Sporadic infections are common but sometimes they may get the form of outbreak These findings supported by (Merchant and Packer1967)and (Dhama etal., 2003).

The antibiogram of isolated bacteria show that most *Escherichia coli* and *Salmonella spp* are sensitive to Ciprofloxacin, Sulphanomide, Amoxcyline and Tetracyline, intermediate to Neomycin and Resistant for penicillin and Erythromycin correspond, whereas *Staphylococcus aureus* are sensitive Ciprofloxacin penicillin and Erythromycin, moderately for Tetracyline and Resistant to Sulphanomide, Amoxcyline.(Young etal., 2003;White etal., 2003) .

Conclusions:

Finally such type of study should be apply regionally at least every two year in order to obtain a good information about the poultry diseases.

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