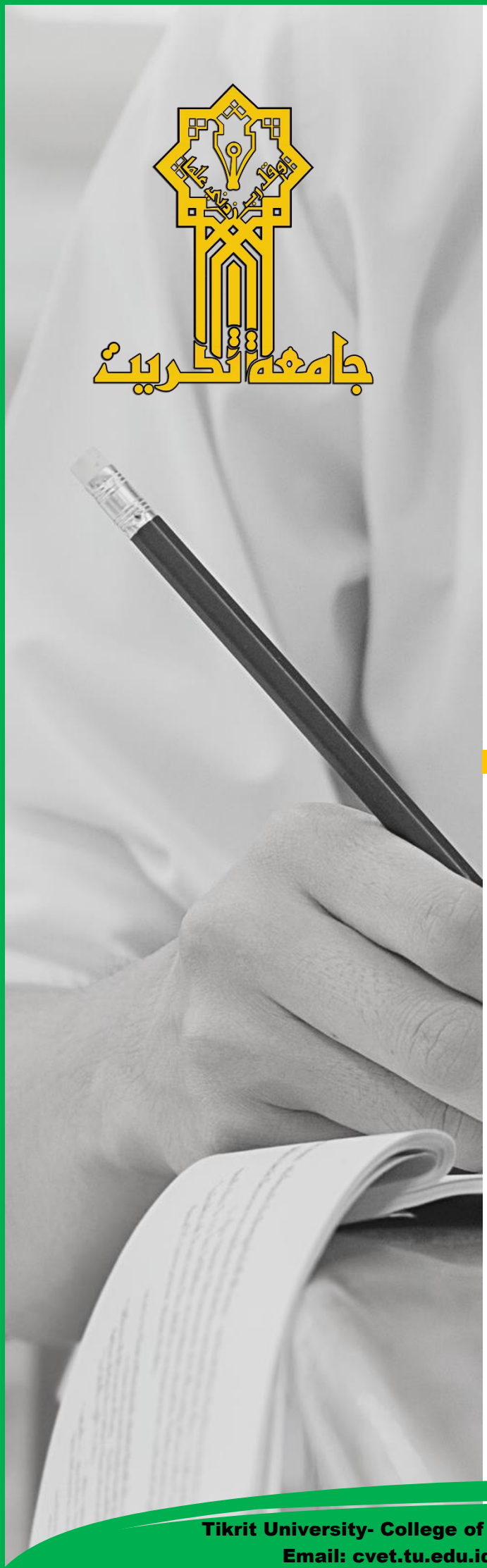

Tikrit University
College of Veterinary Medicine

Colibacillosis

Subject name: Poultry Diseases

Subject year:2024-2025

Lecturer name: Ismael I. Hasan

Academic

Email:ismailhasan@tu.edu.iq

Lecturers link

Colibacillosis

Summary

Colibacillosis is a localized or systemic disease of production poultry associated with the bacterial pathogen avian pathogenic Escherichia coli (APEC). The disease may present in a number of forms including colisepticemia, air sac disease, peritonitis, swollen head syndrome, and salpingitis among others. The previous school of thought was that E. coli was a secondary pathogen to other pre-disposing factors such as viral infection, stress, ammonia, etc. Current thought is that APEC can also be a primary pathogen. Syndromes of disease include colisepticemia, hemorrhagic septicemia, coligranuloma (Hjarre's disease), airsacculitis (chronic respiratory disease, CRD), swollen-head syndrome, venereal colibacillosis, coliform cellulitis (inflammatory or infectious process), peritonitis, salpingitis, orchitis, osteomyelitis/synovitis (including turkey osteomyelitis complex), panophthalmitis, omphalitis/yolk sac infection, and enteritis. Lesions alone should not be used to infer an E. coli infection without the descriptor "coli" or "coliform" being added, because other opportunistic bacteria can behave similarly to E. coli in secondary infections.

Classification

Escherichia is the type genus of the family Enterobacteriaceae, which is composed of organisms that can grow aerobically or anaerobically and utilize simple carbon and nitrogen sources. E. coli is the type species of the genus Escherichia. Additional species have been assigned to the genus but E. coli occurs most commonly and is most important as a pathogen. Although Shigella is still recognized as a genus with 4 species, they actually group genetically with E. coli

Morphology and Staining

Escherichia coli is a Gram-negative, non-acid-fast, uniform staining, non-spore-forming bacillus, usually 2–3 × 0.6μm. Organisms grown in culture are more variable in size and shape. Intracellular organisms are often smaller than extracellular bacteria. Most strains are motile and have peritrichous flagella.

Growth Requirements

Escherichia coli grow aerobically or anaerobically on ordinary nutrient media at temperatures of 18–44°C. It ferments carbohydrates, often producing gas. Generation time and growth rate are related to temperature

Colony Morphology

On agar plates incubated for 24 hours at 37°C, colonies are low, convex, smooth, and colorless. Colonies are bright pink and surrounded by a precipitate on MacConkey's agar, have a dark green-black metallic sheen on eosin-methylene blue (EMB) agar, and are yellow on tergitol-7 agar. Mucoid colonies are raised, larger, appear wet, and are sticky when probed. In contrast to the frequent occurrence of hemolysis by mammalian pathogenic E. coli on blood agar, hemolysis is not a common characteristic of APEC. E. coli rapidly produces diffuse turbidity in broth cultures.

Biochemical Properties

Acid and gas are produced from fermentation of glucose, maltose, mannitol, xylose, glycerol, rhamnose, sorbitol, and arabinose, but not dextrin, starch, or inositol. Substituting sorbitol for lactose in MacConkey agar is useful for distinguishing E. coli O157:H7 from other E. coli because O157:H7 typically does not ferment sorbitol and will appear colorless to straw yellow compared with typical E. coli isolates which will appear pink. Most E. coli isolates ferment lactose, but negative strains, which must be differentiated from Salmonella, are occasionally isolated. Fermentation of adonitol, sucrose, salicin, raffinose, and dulcitol is variable. Isolates that fermented raffinose and sorbose produced high mortality in an embryo lethality test . E. coli produces indole, a positive methyl red reaction, and reduces nitrate to nitrite. Voges-Proskauer and oxidase reactions are negative and hydrogen sulfide is not produced in Kligler's iron medium. E. coli does not grow in the presence of potassium cyanide, hydrolyze urea (urease negative), liquefy gelatin, or grow in citrate medium.

Susceptibility to Chemical and Physical Agents

Escherichia coli have susceptibility patterns to chemical and physical agents typical of vegetative, Gram-negative bacteria. Inactivation of most strains will occur at temperatures ranging from 60°C for 30 minutes to 70°C for 2 minutes. Thorough precleaning and/or presence

of a germicide enhance thermal inactivation. The organism survives freezing and persists for extended periods at cold temperatures. Thermal inactivation in litter to achieve a 90% reduction in the number of bacteria is dependent on time and temperature. Inactivation in litter is slower in the presence of high moisture, but more rapid when free ammonia is present. Reproduction of most strains is inhibited by a pH of less than 4.5 or greater than 9, but the organism is not killed. Some virulent strains, e.g., O157:H7, are acid tolerant, which permits them to pass through the stomach. Organic acids are more effective than inorganic acids at inhibiting growth. Treatment with citric, tartaric, or salicylic acids significantly reduces coliform counts in poultry litter. A salt concentration of 8.5% prevents growth but does not inactivate the organism. Stabilized chlorine dioxide is highly effective when used as a water disinfectant. Chlorate in feed selectively reduces the number of E. coli and related bacteria in the digestive tract by converting relatively nontoxic chlorate to highly toxic chlorite via the same pathway E. coli uses to convert nitrate to nitrite. Solar disinfection of water through the action of ultraviolet light and temperature is a low-cost method of treating drinking water for people that may have application in the poultry industry. Drying is detrimental to the organism. When samples of flooring from broiler transport coops were contaminated with E. coli and allowed to dry for 24 or 48 hours, only very few organisms were still viable. Washing before drying completely eliminated the organism.

Resistance to Heavy Metals, Disinfectants, and Antibiotics

Escherichia coli have the ability to acquire resistance to a broad range of heavy metals (arsenic, copper, mercury, silver, tellurium, zinc) and disinfectants (chlorhexidine, formaldehyde, hydrogen peroxide, quaternary ammonium compounds). Specific strains can vary substantially in their susceptibility to heavy metals and disinfectants. Strains develop resistance to disinfectants when subjected to environmental selection pressures, and such resistances are often encoded by large R plasmids. In addition to antibiotic resistance, the APEC IncHI2 plasmid, pAPEC-O1-R, conferred resistance to potassium tellurite, silver nitrate, copper sulfate, and benzalkonium chloride following transfer of the plasmid to a recipient strain by conjugation.

Antigenic Structure and Toxins

Serotypes of E. coli are classified according to the Kauffmann scheme. Currently there are approximately 180 O, 60 H, and 80 K antigens; the numbers change as new ones are identified and previous ones that are duplicated or attributable to another bacterial species are removed.

In most serologic typing schemes only the O and H antigens are determined, e.g., O157:H7. The O antigen determines serogroup; addition of the H antigen and sometimes K antigen determines serotype. Rough strains autoagglutinate and cannot be serotyped. Additional serotypes with O antigens that have not been recognized also are found in most surveys. Fimbrial (pilus) antigens are included in serotyping when considered important. Recent innovations have enabled assignment of E. coli to "serogroups" using polymerase chain reaction (PCR)-based schemes

Public Health Significance:

Though poultry has not been a significant source of shigatoxin producing E. coli (STEC) in human disease, continued vigilance is recommended because STEC, including E. coli O157:H7, an important enterohemorrhagic pathogen of humans, has been isolated from various types of birds and poultry products. More concerning is the possibility that APEC-contaminated poultry and eggs are a foodborne reservoir of ExPEC that cause human urinary tract infections, meningitis, and other extraintestinal diseases. This hypothesis is based on the remarkable similarities that some APEC and human ExPEC share, by contrast to other meat commodities, in their genomic sequences, serogroups, virulence genotypes, phylogenetic types, plasmid content, antimicrobial resistance patterns and abilities to cause disease in various in vitro and in vivo models of human disease.

Pathogenicity

The ability to cause mortality in embryos or chicks differentiates APEC from commensal E. coli strains. An embryo lethality test can be used to test avian E. coli isolates for virulence. Eleven 12-day-old chicken embryos are inoculated via the allantoic cavity with 100 cfu of the test organism. Two- day mortality is less than 10% for nonvirulent strains, 10–29% for intermediate strains, and more than 29% for virulent strains.

Pathogenesis

Escherichia coli enters host tissues following mucosal colonization or directly through breaks or openings in the skin. Mucosal colonization is dependent on adhesin factors that permit the bacterium to attach to receptors and subsequently reproduce. A variety of fimbrial and nonfimbrial adhesins are produced by E. coli, which facilitates their attachment to host cells.

Air sac epithelial cells round up and become vacuolated following exposure to virulent strains, which causes them to separate from each other providing bacteria access to systemic tissues. The ability of APEC to bind with fibronectin and laminin, 2 components of basement membranes, would aid in penetration through the damaged mucosa into host tissues. Toxins in cell-free culture filtrates, most likely endotoxin, produce the same acute inflammatory response as the living organism.

Alternatively, the initial portal of entry into the host's tissues, an essential first step in colisepticemia, may be transcellular through nonphagocytic cells. Certain strains of APEC have the ability to invade fibroblasts, much like virulent S. typhimurium. E. coli have been identified within air sac epithelial cells by electron microscopy. An APEC strain was able to readily adhere to and invade tracheal epithelial cells.

Once E. coli becomes extramucosal, the environment it has entered is extremely hostile. Unless the organism is equipped with survival capabilities (e.g., "virulence" factors), it is rapidly destroyed by phagocytic cells such as heterophils, thrombocytes, and macrophages.

Macrophages located primarily in the spleen and liver phagocytize bacteria that gain access to the circulation. Complement and antibodies to O antigens (endotoxin), outer-membrane proteins (siderophores), and fimbriae serve as opsonins to promote phagocytosis and destruction of the organism. Endotoxin also decreases the bacteriocidal ability of pulmonary macrophages, which may aid in survival and dissemination.

Immediately after E. coli contacts host tissues, there is an acute inflammatory response. Acute phase proteins produced in the liver and cytokines IL-1, IL-6, and tumor necrosis factor increase rapidly following exposure to endotoxin or E. coli, which can serve as nonspecific indicators of early disease.

Acute phase effects of endotoxemia include hypothermia followed by hyperthermia, hypotension, decreased circulating heterophils associated with increased apoptosis and sequestration in the lung, and increased inflammatory mediators, TL1A, IL-1β, and IL-6.

Increasing amounts of endotoxin in the circulation causes decreased feed consumption and efficiency, decreased body weight and breast meat yield, decreased tibial bone size, weight, calcium content, and breaking strength, and increased mortality, liver weight, plasma ionized calcium, and antibody responses.

Vascular permeability increases leading to the accumulation of fluid and protein in the

tissues. Serous membranes become wet and edematous and liquid begins to accumulate in body cavities. Chemotactic factors attract heterophils, which marginate in postcapillary venules and emigrate into surrounding tissues. Between 6 and 12 hours, soft, gelatinous exudate becomes grossly visible. Heterophils can kill E. coli extracellularly by substances such as ß-defensins released as they degranulate and die.

After 12 hours there is a progressive shift in inflammatory cells from heterophils to macrophages and lymphocytes Exudate continues to accumulate and eventually undergoes caseation to form a firm, dry, yellow, irregular, cheese-like mass. Microscopically, caseous exudate consists of heterophilic granulomatous exudate containing variable numbers of embedded bacterial colonies.

A palisade of multinucleated giant cells and macrophages surrounds the exudate. Depending on the size of the mass of exudate, an extended period of time will be required for the exudate to be slowly eroded away by the action of the surrounding phagocytic cells.

Viable bacteria persist as microcolonies within the exudate. Epithelial tissue may be restored if damage has not been too severe, but usually there is some degree of fibrosis, which may be complete (scarring) if tissue destruction has been extensive.

Exudate containing fibrin undergoes organization and is eventually converted to scar tissue. Gross lesions are inversely related to virulence. Highly virulent strains cause mortality so quickly that gross lesions have little time to develop, whereas birds infected with less virulent strains survive longer and develop more extensive lesions.

Transmission

Horizontally

Incubation Period:

1-7 days

Clinical Signs:

1- Coliform cellulitis is typically not detected until the birds are processed.

2- Lameness and retarded growth are seen in birds with skeletal lesions that develop as a sequel to sepsis.

3- Affected birds are typically undersized for the flock and found at the ends of the house, along the side walls, or under feeders or waterers.

4- They may be victims of persecution (“cannibalism”) by other birds.

5- When joints or bones of 1 leg are affected, birds walk with a characteristic hopping motion to keep weight off the affected leg. Birds with lesions in both legs are either nonambulatory or have great difficulty in standing and walking.

6- When the thoracolumbar spine is affected, the birds have an arched back, sit on their hocks, and bear little or no weight on their feet.

7- Birds with chronic lameness have caking of droppings around the vent and on abdominal feathers.

8- Young birds with omphalitis and infected yolk sacs also may have difficulty in walking because of abdominal distention, which alters weight distribution and impairs balance.

9- Birds with colisepticemia are often terminally moribund or very lethargic. 10-

Decreased water consumption is associated with a poor prognosis.

11- Severely affected individual birds are unresponsive when approached, do not react to stimuli, and are easily caught and handled.

12- The beak may be inserted into the litter to support the head.

13- Dehydration is indicated by dark dry skin, which is especially noticeable in the shanks and feet.

14- Although, technically, death is not a clinical sign, this may be the main indication of an outbreak of colibacillosis in a flock.

Localized Forms of Colibacillosis

1- Coliform Omphalitis/Yolk Sac Infection.

Omphalitis is an inflammation of the navel (umbilicus). In birds the yolk sac is also usually involved (yolksacculitis) because of its close anatomic relationship to the umbilicus. Fecal contamination of the egg shell and unsanitary conditions in the hatchery are considered the most important sources of infection. Bacteria may be acquired in ovo if the hen has oophoritis or salpingitis or via contamination following artificial insemination. **Yolksacculitis** also can result from translocation of bacteria from the chick's intestine or from the bloodstream. In these cases the navel is not affected. Similarly, **peritonitis** can occur without involvement of the umbilicus. It is common to recover low numbers of E. coli from normal yolk sacs. Between 0.5% and 6% of eggs from normal hens contain E. coli.

E. coli and Enterococcus faecalis infections accounted for approximately half of the mortality that occurred in layer chicks during the first week after hatching. First week mortality was significantly correlated with total mortality in the flock but not flock uniformity. For good flock performance, first week mortality needs to be less than 1%.

A high percentage of E. coli isolates from eggs, dead embryos, and chicks that died between placement and 7 days of age possessed the virulence genes.

Some embryos may die before hatching, particularly late in incubation; whereas others die at or shortly after hatching. Surviving infected chicks can be a source of E. coli for other chicks in the same hatch. The incidence of birds with omphalitis increases after hatching and declines after about 6 days with occasional losses continuing up to 3 weeks. As few as 10 organisms of serotype O1a:K1:H7 caused 100% mortality in day-old chicks following yolk sac injection.

Swelling, edema, redness, and possibly small abscesses characterize acute inflammation of the navel. The abdomen is often distended and blood vessels are hyperemic.

In severe cases, the body wall and overlying skin undergo lysis and are wet and dirty leading to the term “mushy” chicks or poults. Other nonspecific changes such as dehydration, visceral gout, emaciation, vent pasting, and enlarged gall bladder may be seen.

The yolk sac is typically distended because yolk has not been absorbed and inflammatory products have been added. Yolk is abnormal in color, consistency, and smell, and may contain visible exudate. Blood vessels of the yolk sac are often prominent.

Chicks or poults with infected yolk sacs that live more than 4 days also may have **peritonitis**, **pericarditis**, or **perihepatitis**, indicating **local and systemic** spread of the organism from the yolk sac.

Microscopically the wall of the infected yolk sac is **edematous** with mild inflammation. There is an outer connective tissue zone adjacent to a layer of inflammatory cells containing **heterophils** and **macrophages**, a layer of giant cells, a zone of **necrotic** heterophils and masses of **bacteria**, and then the inner, abnormal yolk contents. A **few plasma** cells may be found in some yolk sacs.

Consequences of yolk sac infection include deprivation of nutrients and maternal antibodies, absorption of toxins, and spread of E. coli by extension into the body cavity (peritonitis) or systemically to produce colisepticemia and its sequelae (polyserositis, arthritis). Survivors are usually stunted and do poorly. Birds that survive the acute infection have small, firm, persistent yolk sacs (often referred to as “retained” yolk sacs) that contain inspissated exudate and yolk material. E. coli persists in these chronically inflamed yolk sacs and can be isolated from them for weeks to months after hatching. Adhesions to intestines, especially the tip of the duodenal loop, or other visceral organs are common. Rarely the elongated stalk of the yolk sac will wind around the

intestine and cause strangulation.

2-Coliform Cellulitis (Avian Cellulitis, Inflammatory Process, Infectious Process). Coliform **cellulitis** is characterized by sheets of serosanguineous to caseated, fibrinoheterophilic exudate in subcutaneous tissues. Lesions, often referred to as "plaques," are located in the skin over the abdomen or between the thigh and midline. Other colibacillosis lesions, or reduced productivity, occasionally accompany coliform cellulitis, but usually lesions are discovered at processing **when inspectors open** the thickened yellow abdominal body wall of an otherwise normal carcass.

Cellulitis isolates of E. coli belong to the same serogroups as those that cause other forms of colibacillosis. They usually produce **colicin** and **aerobactin**. Virulence properties and molecular characteristics are similar among isolates from **cellulitis and colisepticemia lesions and normal birds**. However, isolates from cellulitis lesions have a **greater ability** to produce cellulitis in experimentally exposed birds than E. coli isolates from **airsacculitis** lesions or feces of healthy chickens.

A vacuolating cytotoxin produced by cellulitis E. coli isolates is also produced by isolates from chickens with colisepticemia and swollen head syndrome but not by isolates from healthy chickens. The cytotoxin is similar to one produced by **Helicobacter pylori**, except that H. pylori cytotoxin is **specific for mammalian** cells whereas the avian E. coli cytotoxin is specific for avian cells.

These organisms persist for at least 6 months, irrespective of partial or complete cleaning and disinfection as performed in the field, and cause coliform cellulitis in successive flocks.

Regional differences in the prevalence of coliform cellulitis emphasize the important roles of environmental and management factors in occurrence of the disease. Increased condemnation rates caused by coliform cellulitis during the past 25 years indicate that changes have occurred in either the occurrence or characteristics of risk factors associated with coliform cellulitis.

Fast-growing, heavy broiler strains are more likely to have an increased prevalence and severity of skin scratches, which predispose to coliform cellulitis. Coliform cellulitis occurs more frequently in males than females. The gene responsible for sexing regulates feather growth. Slower feathering males may be more vulnerable to skin injuries because of greater exposure of the skin to potential physical damage. Sex may also contribute to coliform cellulitis because of its association with weight, aggressiveness, or management practices.

Significant variables, indicated a 40%– 60 % increase in cellulitis as temperatures increased over a range of approximately 60°F (15.5°C) from 29°F (–1.7°C) to 94°F (34.4°C). An increase in relative humidity from 36% to 93 % was predicted to increase cellulitis from 0.3% to 0.9 % in low-prevalence flocks and from 1.0% to 1.9 % in high-prevalence flocks.

The occurrence of coliform cellulitis was higher in vegetarian broilers compared with broilers fed feeds containing animal products. Condemnation rates for birds fed a standard diet, which contained growth promotants, antibiotics, and anticoccidials, was substantially lower (0.26%) than for birds fed a vegetarian or organic feed without additives (1.18%).

Providing vitamin E at 300mg/kg or vitamin A at 60,000IU/kg improved the resistance of 6-week-old broilers against E. coli infection. Birds fed both vitamin E at 48 IU/kg and a zinc- protein complex at 40ppm of zinc decreased the occurrence of coliform cellulitis.

3- Swollen Head Syndrome.

Swollen head syndrome (SHS) is an acute to subacute cellulitis involving the periorbital and adjacent subcutaneous tissues of the head. SHS was first described in broilers in South Africa associated with E. coli and an unidentified coronavirus infection. Swelling of the head is caused by inflammatory exudate beneath the skin that accumulates in response to bacteria, usually E. coli, following upper respiratory viral infections (e.g., avian metapneumovirus, infectious bronchitis virus). Ammonia aggravates the disease. The portal of entry is considered to be the **conjunctiva** or inflamed mucous membranes of the **sinuses** or **nasal** cavity. Possible infection via the **Eustachian tube** also has been suggested.

Microscopic lesions include fibrinoheterophilic inflammation and heterophilic granulomas in the air spaces of the cranial bones, middle ear, and facial skin. Lymphoplasmacytic conjunctivitis and tracheitis with formation of germinal centers have also been observed.

4- Diarrheal Disease.

Primary enteritis is a common manifestation of E. coli infections in mammals including humans, but is considered rare in poultry. Diarrhea results from infections with enterotoxigenic (ETEC), enterohemorrhagic (EHEC), enteropathogenic (EPEC), or enteroinvasive E. coli (EIEC); each type possessing certain virulence factors that determine the characteristics for each type of enteric disease. EHEC and EPEC strains produce attaching and effacing lesions on intestinal mucosal surfaces. Collectively, these strains are called attaching and effacing E. coli (AEEC).

ETEC that elaborated toxins capable of causing fluid accumulation in ligated intestinal loops of chickens were recovered from chickens with diarrhea, and an O15 APEC strain that

produced heat-labile toxin II was isolated from ostrich chicks experiencing severe diarrhea and high mortality.

Infections with infectious bursal disease (IBD) virus in chickens and adenovirus infection in the pigeon were considered possible predisposing factors to AEEC infection.

Birds infected with AEEC may be clinically normal or have diarrhea and be dehydrated. In clinically affected birds, the intestines are pale and distended with fluid, which may contain visible flecks of mucus and exudate. Ceca are often the most obviously affected part of the digestive tract. They are typically distended with pale brown fluid and gas.

Bacteria intimately attach to the surface of enterocytes causing effacement of microvilli, pitting, and pedestal formation, which are best seen by electron microscopy. Lesions are most common in the ceca. Organisms are readily identified in tissue sections using Giemsa stain or by immunohistochemical methods.

5- Venereal Colibacillosis (Acute Vaginitis).

Venereal colibacillosis is an acute and frequently fatal vaginitis that affects turkey breeder hens shortly after they are first inseminated usually in the first weeks of egg production. Puncturing the hymen of young turkey hens can lead to a severe localized E. coli infection characterized by vaginitis, cloacal and intestinal prolapse, peritonitis, egg binding, and internal laying.

The affected mucosa is markedly thickened, ulcerated, and covered with a diphtheritic, caseonecrotic membrane, which causes obstruction of the lower reproductive tract. The thickness of these membranes posed an obstruction to egg passage leading to internal laying and egg peritonitis. The upper oviduct is grossly and histologically normal.

Flocks can have losses of up to 8% because of increased mortality and culling. Egg production is decreased and there is an increased number of cull eggs because of small size. Swabs from cloaca and vagina produced numerous colonies of only E. coli. No other infectious agents have been identified as contributing to the disease. Anecdotal reports indicate a similar disease can affect young broiler breeders as they come into production.

6- Coliform Salpingitis/Peritonitis/Salpingoperitonitis (Adult).

Inflammation of the oviduct caused by E. coli results in decreased egg production and sporadic mortality. It is one of the most common causes of mortality in commercial layer and breeder chickens and also affects other female birds, especially ducks, geese, and quail.

Accumulations of caseating exudate in the body cavity resemble coagulated yolk, which is

the reason for the common name “egg peritonitis.” Yolk peritonitis is a mild to moderate diffuse peritonitis without exudate resembling coagulated yolk that results from free yolk in the body cavity. Yolk peritonitis is usually associated with bursting atresia that occurs during acute ovarian regression.

Marked exudation, extensive inflammation, and positive cultures characterize coliform peritonitis and serve to distinguish it from yolk peritonitis. Salpingitis and egg binding may occur concurrently, which can cause confusion because both result in an obstructive mass within the oviduct. If an egg is not grossly visible, cutting through the mass in the oviduct and finding a shelled egg in the center indicates egg binding.

Mucosal infections with viruses (e.g., infectious bronchitis virus) or mycoplasmas also may predispose a bird to salpingitis. M. synoviae infection increased the occurrence of coliform peritonitis. Coinfection with E. coli and Tetratrichomonas occurred in Pekin duck breeders with salpingitis. Heavy egg production and associated estrogenic activity predispose hens to salpingitis by relaxing the sphincter between the vagina and cloaca. Spread of E. coli to the oviduct from an airsacculitis is also possible, but this form of salpingitis occurs more frequently in young birds as part of a systemic infection. Isolates of APEC from birds with salpingitis have similar virulence characteristics to those that cause airsacculitis.

In chronic cases, the oviduct is markedly distended and thin-walled with single or multiple masses of caseous exudate in the general form of the oviduct. The mass of exudate may expand to the point that it fills most of the body cavity. Rupture of the oviduct is possible. Exudate is laminated, often contains a central egg, shells, and/or membranes, and is malodorous. Spread of the organism into the body cavity through the compromised oviduct wall or the open end of the infundibulum leads to concurrent peritonitis, which is termed salpingoperitonitis when there is involvement of both the oviduct and peritoneum. Acute cases have less exudate in the oviduct or peritoneum that tends to be soft and not as caseated. Affected birds are incapable of laying eggs although they typically continue to ovulate. Repeated ovulations and albumen secretion are responsible for the laminated appearance of the oviductal masses. Abdominal laying and misovulated ova may accompany salpingitis and contribute to peritonitis.

Microscopically, the tissue reaction in the oviduct is surprisingly mild in view of the marked gross lesions. It primarily consists of multifocal to diffuse heterophil accumulations subjacent to the epithelium and caseous exudate in the lumen, which often contains bacterial colonies. Lymphoid foci develop in the mucosa with time and indicate chronicity.

7- Coliform Orchitis/Epididymitis/Epididymo-Orchitis.

An ascending E. coli infection of the male reproductive tract, analogous to that resulting in salpingitis in the hen, occurs infrequently in roosters. Testicles are swollen, firm, inflamed, irregularly shaped, and have a mosaic of necrotic and viable tissue when opened. Heavy growth of E. coli can be obtained from the testicle and epididymis.

B-Systemic Forms of Colibacillosis 1-

Colisepticemia.

Presence of virulent E. coli in the blood stream defines colisepticemia. Virulence and number of organisms balanced against efficacy of host defenses determine duration, degree, and outcome of the disease, as well as the pattern and severity of lesions. Colisepticemia progresses through the following **stages**: acute septicemia, subacute polyserositis, and chronic granulomatous inflammation. Whereas lesions are typical of colisepticemia, other bacteria capable of producing septicemia also can cause similar changes.

Characteristic features of colisepticemia at necropsy are tissues that develop a green discoloration following exposure to air and a characteristic odor, possibly related to indole produced by the organism.

The bursa of Fabricius is often atrophic or inflamed as a result of colisepticemia. It should not be interpreted that a small bursa is evidence of a prior immune-suppressing disease such as IBD.

Pericarditis is common and is a characteristic of colisepticemia. It is usually associated with myocarditis, which results in marked changes in the electrocardiogram, often before gross lesions appear. Vessels in the pericardium become increasingly prominent because of hyperemia and the pericardium becomes cloudy and edematous. Initially, fluid and soft masses of pale exudate accumulate within the pericardial sac followed by fibrinous exudate. Exudate can be seen loosely adhering to the epicardium when the pericardial sac is opened. As the disease progresses, exudate increases, becomes more cellular (fibrinoheterophilic), and undergoes caseation. The pericardial sac adheres to the epicardium with chronicity.

Microscopically, the same progression of lesion development is seen. Serous and serofibrinous exudate is seen initially followed by increasing numbers of heterophils and subsequently macrophages. Within the myocardium, particularly close to the epicardium, there are accumulations of lymphoid cells and by 7–10 days there also are many plasma cells. Subsequently,

exudate in the pericardial sac undergoes organization, which, in survivors, eventually results in constrictive pericarditis and liver fibrosis caused by chronic passive congestion. Cardiac lesions reduce arterial blood pressure from a norm of about 150mmHg to about 40mmHg just before death. Several distinct clinical forms of colisepticemia can be distinguished depending on how the organism gains access to the circulation and the age and type of bird.

2- Respiratory-Origin Colisepticemia.

Respiratory-origin colisepticemia affects both chickens and turkeys and is the most common type of colisepticemia. E. coli gains access to the circulation following damage to the respiratory mucosa by infectious or noninfectious agents.

Severity of the resulting disease, which is commonly referred to as airsacculitis, CRD, or multicausal respiratory disease, is directly related to the **number of agents** that are involved.

A diversity of E. coli serotypes can be identified in the tissues are usually different from those in the intestinal tract of the same bird but can be found in the intestinal tracts of other birds and the environment.

Susceptibility is increased when IBV, IBD or NDV infection occurs. **Five days** after administration of a vaccine strain of NDV, clearance of aerosol-administered E. coli is reduced. **Microscopically**, 3–8 layers of immature, nonciliated cells replace the pseudostratified, columnar epithelium of the trachea. Mixed IBV- E. coli infections are more severe than those caused by either agent alone.

Coinfection with IBV or NDV in addition to mycoplasma further decreases resistance to E. coli and the period of increased susceptibility begins earlier and persists longer.

Exposure to chicken-house dust and ammonia results in deciliation of the upper respiratory tract of birds permitting inhaled E. coli to colonize and cause respiratory infection.

Lesions are prominent in respiratory tissues (trachea, lungs, air sacs), pericardial sac, and peritoneal cavities and are typical of the subacute polyserositis stage of colibacillosis. Infected air sacs are thickened and often have caseous exudate on the respiratory surface.

Microscopically, the earliest changes consist of edema and heterophil infiltration. Mononuclear phagocytes are frequently seen 12 hours after inoculation. Later, macrophages become common, with giant cells along margins of necrotic areas. There is fibroblast proliferation and accumulation of vast numbers of necrotic heterophils in caseous exudate. Bacterial colonies are often present in caseous exudate and contain numerous organisms. E. coli

colonies have a typical appearance in histologic sections. They are usually circular with concentrated bacilli forming a distinct smooth perimeter with fewer bacilli and spaces centrally. They stain negative with tissue Gram stain.

Lesions of predisposing respiratory disease are usually present in the trachea and lungs and consist of lymphoid follicles, epithelial hyperplasia, and epithelium-lined air passages that may contain heterophils. Pneumonia and pleuropneumonia are more common in turkeys whereas chickens usually have pleuritis or pleuropneumonia with less lung involvement.

Extension of the disease process into the oviduct from the left abdominal air sac may occur and cause salpingitis in juvenile birds.

Airsacculitis occurs within 1.5 hours. **Bacteremia** and **pericarditis** develop within 6 hours. In birds that survive, lesions are well-developed by 48 hours postinoculation. Most mortality occurs during the first 5 days.

Recovery is usually rapid if birds survive the initial infection, although a few with persistent anorexia become emaciated and die.

3- Enteric-Origin Colisepticemia.

Enteric-origin colisepticemia is most common in turkeys. E. coli gain access to the circulation and tissues following damage to the intestinal mucosa by infectious agents.

The most common predisposing agent is **hemorrhagic enteritis virus**. Usually only 1 or 2 types of E. coli are involved in the disease outbreak, and those in the tissues and intestinal tract of each bird tend to be the same.

Lesions are typical of the acute septicemic stage of colibacillosis. Affected birds are in good physical condition and often have full crops containing feed and water. The most characteristic lesions are congestion or green discoloration of the liver, marked enlargement and congestion of the spleen, and congested muscles.

Microscopically the spleen is congested with proteinaceous fluid in sinuses and has multifocal necrosis, often containing intralesional bacteria. Fibrin thrombi are present in liver sinusoids and occasionally renal glomeruli. In some cases, multiple, pale foci in the liver are seen. Microscopically these are areas of acute necrosis initially, but with time they evolve into granulomatous hepatitis in survivors.

After a few days birds eventually develop lesions similar to those of respiratory-origin colisepticemia.

4-Hemorrhagic Septicemia.

This form of colibacillosis occurs in turkeys and is characterized by generalized circulatory disturbances, discoloration of subserosal fat, bloody fluid on serosal surfaces, pulmonary edema and hemorrhage, and enlargement of the liver, spleen, and kidneys.

Generalized necrosis of the liver and multifocal necrosis of the spleen are seen on cut surfaces. Pure cultures of E. coli are obtained from the liver, spleen, and pericardium.

Characterization of isolates from 7 affected flocks showed they belonged to multiple serogroups (O1, O2, O18, O78, and O111). For each flock, isolates were clonal based on serotype, plasmid profiling, ribotyping, and MLST. Virulence factors were similar among all isolates in spite of differences in serotype and phenotype, and included F11 pili, iucD, iss, vat, tsh, and colicin V.

5- Neonatal Colisepticemia.

Chicks are affected within the first 24–48 hours after hatching. Mortality remains elevated for 2–3 weeks and usually totals 10%–20%. Up to 5% of the flock may be stunted and require culling. Unaffected birds grow normally and the disease does not appear to spread.

Initial lesions consist of congested lungs, edematous serous membranes, and splenomegaly. The proventriculus and lungs develop a dark color that can approach black as the interval between death and necropsy increases.

Microscopically, bacteria are numerous in affected tissues and easily identified. After a few days the typical pattern of acute, fibrinoheterophilic polyserositis involving the pericardial sac, pleura, air sacs, and peritoneum becomes evident. Lesions are often extensive and severe in birds that survive into the second week.

Occasionally birds with arthritis or osteomyelitis may be found late in the disease. Most affected birds have yolk sac abscesses suggesting the navel is the portal of entry. Alternatively, in ovo infection may be responsible.

6- Layer Colisepticemia.

Colisepticemia is usually a disease of young birds, but occasional outbreaks of acute E. coli infection resembling fowl typhoid or fowl cholera occurs in mature chickens and turkeys. Acute colibacillosis in layers is being seen with increasing frequency.

The majority of outbreaks are associated with onset of egg production, but less frequently

they occur at an older age, or may continue as the flock ages and potentially spread to older flocks on the same farm.

Death usually occurred suddenly without premonitory signs, although depression and/or dirty vents were observed in some affected hens in approximately half of the flocks.

Weekly mortality was significantly higher in affected flocks than age-matched control flocks (0.26%–1.71% vs. 0.07%–0.30%). Cumulative mortality ranged up to 10% and mortality remained elevated for 3–10 weeks.

Polyserositis (perihepatitis, pericarditis) and peritonitis associated with free yolk in the peritoneal cavity were present in most birds at necropsy. Oophoritis and salpingitis occurred less frequently.

The pathogenesis of the disease is unknown, but stress associated with onset of egg production is believed to be an important contributing factor. Ascending infections via the oviduct have been suggested as a means by which E. coli gain access to systemic tissues, but in a recent study, higher colonization rates of the trachea, but not the oviduct, in affected flocks suggests layer colisepticemia may be aerogenous.

Risk factors for developing layer colisepticemia include close proximity to other poultry farms and higher stocking density.

Control has been through **chlorination of water** or treatment of the flock with appropriate **antibiotics**.

Colisepticemia Sequelae.

Death is the usual outcome of colisepticemia, but some birds may **completely recover** or **recover with residual sequelae**. If the bird does not control E. coli, it can **localize** in poorly protected ("immunologically privileged") sites including the **brain**, **eyes**, **synovial** tissues (joints, tendon sheaths, sternal bursa), and **bones**. In immature females, **salpingitis** can occur when there is involvement of nearby **air sacs**.

After E. coli is no longer present, **constrictive pericarditis** develops as exudate in the pericardial sac undergoes organization, and **liver becomes fibrotic**. **Ascites** may develop because of residual pulmonary damage from combined E. coli–IBV infection. Endotoxin causes **vasoconstriction** leading to **pulmonary hypertension** and the potential to develop ascites (pulmonary hypertension syndrome).

7- Meningitis and Meningoencephalitis.

E. coli localization in the brain, although uncommon, has been reported. Meninges are affected (meningitis) but in some birds there also is involvement of the brain (encephalitis) and ventricles (ventriculitis). Meningeal lesions are evident at necropsy as areas of discoloration, often adjacent to major blood vessels. Fibrinoheterophilic to heterophilic exudate is seen microscopically early in the infection; the lesion becomes more granulomatous with time. Bacteria are usually numerous within lesions but may not form distinct colonies.

8- Panophthalmitis.

As with the brain, involvement of the eye is uncommon. However, if it is infected the resulting panophthalmitis is severe. Typically there is hypopyon and hyphema, and infection is unilateral. The eye is swollen, cloudy to opaque, and may be hyperemic initially. Later the eye shrinks as it undergoes atrophy. Fibrinoheterophilic exudate and numerous bacterial colonies are present throughout the eye. Inflammation, especially adjacent to necrotic tissue, becomes granulomatous with time. Varying degrees of retinal detachment, retinal atrophy, and lysis of the lens also may be seen. The organism persists in the diseased eye for long periods of time.

9- Osteoarthritis and Synovitis.

Localization of E. coli in bones and synovial tissues is a common sequel to colisepticemia. The term **osteoarthritis** is used when a **joint** is inflamed and **1 or more bones** making up that joint have osteomyelitis. **Polyarthritis** refers to involvement of more than 1 joint. **Bacterial chondronecrosis with osteomyelitis (BCO)** is another name that has been used. Affected birds likely have insufficient resistance to clear bacteria completely.

Hematogenous spread of E. coli following hemorrhagic enteritis virus infection of turkeys resulted in synovitis, osteomyelitis, and green liver discoloration.

Mild to severe lameness and poor growth are seen clinically and affected birds are more likely to be victims of persecution ("cannibalism"). Often multiple sites are involved. Bacteria colonize the vascular sprouts that invade the physis of a growing bone provoking an inflammatory response that results in osteomyelitis.

Transphyseal blood vessels in birds serve as conduits for the process to spread into the joint and surrounding soft tissues.

Lame turkeys had the following: higher splenic and liver weights, lower body and bursal weights, decreased cellular immunity, normal to increased humoral immunity, decreased circulating lymphocytes, increased circulating total leukocytes, monocytes, and heterophils, normal to

marginally depressed phagocyte function, increased serum protein, uric acid, and blood urea nitrogen (BUN), and decreased hemoglobin, iron, alkaline phosphatase, and gamma- glutamyl-transferase.

Bones most often affected are the tibiotarsus, femur, thoracolumbar vertebra, and humerus. Proximal physes of long bones are more frequently affected than distal physes.

Lesions typically form where endochondral ossification is occurring and extend proximally to involve the adjacent physeal cartilage. It is common to find both osteomyelitis and tibial dyschondroplasia together, but this is most likely because of their occurrence at the same location rather than a cause and effect relationship.

Osteomyelitis is easily recognized on gross examination of bones opened to expose the physes, but small lesions that can only be seen microscopically also occur. Hock, stifle, hip, and wing joints and articulations of the free thoracic vertebra are sites where arthritis is most likely to occur.

Trauma to joints and growing bones may predispose to the development of lesions. Tenosynovitis frequently accompanies arthritis.

Less commonly, spread of the inflammatory process from a joint into the periarticular tissues occurs. An infectious sternal bursitis is also common but must be distinguished from **traumatic sternal bursitis** in which **fluid is seen but not exudate**. When inflammatory lesions involve the shoulder joint or proximal humerus, extensive exudate can accumulate between the superficial and deep pectoral muscles. Lesions that develop in joint spaces of the articulating free thoracic vertebra are characterized by spondylitis, which results in progressive paresis and paralysis.

10- Turkey Osteomyelitis Complex.

Turkeys with turkey osteomyelitis complex (TOC) have infectious, inflammatory lesions in bones, joints, or periarticular soft tissues and enlarged, green discolored livers that are used at processing to indicate the possible presence of intraosseous lesions.

Green liver discoloration is rarely identified in the field even in lame turkeys. Most often green discolored livers are detected at processing and result in downgrading and, depending on severity, partial or whole bird condemnation.

Feed and water withdrawal do not cause green liver discoloration, but infection with **M. synoviae can** be associated with a high percentage of carcasses with green discolored livers in

the absence of TOC.

The **Food Safety Inspection Service** in the United States uses green discoloration of the liver at processing to identify carcasses that may have TOC. **When vitamin D** metabolites were administered, TOC was reduced as with vitamin D treatment, but there were toxic effects in dexamethasone-treated turkeys challenged with E. coli.

11- Coligranuloma (Hjarre's Disease).

Coligranuloma of chickens and turkeys is characterized by multiple granulomas in liver, ceca, duodenum, and mesentery, but not spleen. The disease also has been described in quail. Coligranuloma is an uncommon form of systemic colibacillosis that usually occurs sporadically in individual birds but can cause mortality as high as 75% when a flock is affected.

Serosal lesions resemble leukosis tumors. Early in the disease there is confluent coagulation necrosis involving as much as half the liver. Only scattered heterophils are seen, and at the edge of the necrotic areas there are a few giant cells. Subsequently, typical heterophilic granulomas are present in the affected tissues. Pyogranulomatous typhlitis and hepatitis, which may be related to coligranuloma, have been described in turkeys with cecal cores and ruptured ceca.

Diagnosis:

1- Clinical signs and gross and histologic lesions. 2-solation and Identification of Causative Agent

3- RT-PCR. 4-ELISA

Differential Diagnosis

1- Acute septicemic diseases may result from pasteurellae, salmonellae, teurellae, or streptococci, pericarditis or peritonitis, mycoplasmas, Chlamydophila,

2- synovial lesions

Treatment

1- Antibiotics:

A-Fluoroquinolones

B-(APEC frequently are resistant to tetracyclines, sulfonamides, ampicillin, and streptomycin) C-A high percentage of E. coli isolates from turkeys are resistant to gentamicin

D-ampicillin (1.7 and 5 g/ton) developed resistance

E-Paradoxically, certain antimicrobials and anticoccidials commonly used at subtherapeutic levels in poultry for growth promotion and coccidiosis control inhibited transfer of a plasmid that is

responsible for multiresistance in E. coli.

F-Resistance to florfenicol and chloramphenicol, which had never been used in poultry in the United States, was found in E. coli isolates from chicken

G-Water administration of apramycin proved effective in reducing the numbers of organisms in the digestive tract and preventing bacteremia in chickens

H-Neomycin reduced mortality in turkey poults exposed naturally to litter from flocks with colibacillosis

I-Monensin reduced colonization of chickens with E. coli O157:H7 to undetectable levels 14 days postexposure compared with nonmedicated controls and chickens receiving other coccidiostats.

J-Bacitracin alone provided no protection against colibacillosis

K-Antimicrobial resistance associated with colistin has recently emerged as a significant concern worldwide

2- Anti-inflammatory:3-Supplements: 4-Local antiseptics