PATHOGENICITY OF *E.COLI* O₅₅ K₅₉ TO 5 – 30 DAYS OLD BROILER CHICKENS

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ABSTRACT

Studying pathogenicity of E. coli O₅₅ K₅₉ strain to broiler chickens at three different ages of 5, 21 and 30 days old, the ability of this strain to induce clinical signs. lateral transmission to contact chickens and the effect of experimental and contact infection on the body weight gain. The study proved that pathogenicity of E. coli O₅₅ K₅₉ strain was differs according to age of chickens and route of inoculation and laterally transimeted by contact. When E. coli O₅₅ K₅₉ was inoculated orally to 5 days old chicks. resulted in clinical signs in infected and contact chicks as well as 26.67% mortality and the inoculated strain was reisolated from (dead, infected sacrificed, contact sacrificed) at percentages of 100%, 63.6% and 50%; respectively. When E.coli O₅₅ K₅₉ was inoculated subcutaneously to chicken at age of 21 days old, mortality was 13.33% while the organism was reisolated from dead, sacrificed infected and contact sacrificed at percentage of 100%, 84.6% and 70%; respectively. While inoculation into 30 days old, no mortality was observed and the organism was reisolated from both sacrificed infected and contact at percentage of 53.3% and 30% respectively. Clinical symptoms of respiratory disease in the inoculated birds at 21 or 30 days of age were recorded. We observe that the curve of reisolation from contact group at the three different ages was the same as that of infected group, at the three ages. Infected birds at 30 days old showed a reduction in body weight gain than control group 13.1%, 23.1%, 20.3% and 18.6% at 35, 40, 43 and 46 days of ages, respectively. Decrease in body weight gain of contact group than control. The conversion rate of infected chicken group was lowest (2.586) than that of contact group (2.356) than control negative group (2.306) as calculated at 46 days old.

INTRODUCTION

E.coli had been reported to be pathogenic or facultative pathogenic to many species of poultry including chicken (Gross, 1957 a &b; Gross and Siegle, 1959; Gross, 1961; 1964, Heller et al., 1972 and Shane, 1983). Broilers chickens that characterized with rapid growth rate which can be considered as on of the stressors can make such birds to be more susceptible to E.coli especially under deep litter system due to ammonia and high humidity.

E.coli was also investigation to be the causative agent of colibacillosis that resulted in severe losses in broiler chickens including high mortality, low performance and high medication costs as well as increase condemnation rate during processing (Awaad, 1972; Youssef et al., 1983; Khalid, 1990; Riad, 1994 and Radwan 2000).

Many E.coli serotypes had been demonstrated to be pathogenic including O_1 , O_2 , O_{124} , O_{78} , O_{114} , O_{119} , O_{128} , O_{127} (Awaad, 1972; Youssef *et al.*, 1983; Khalid, 1990; Riad, 1994 and Radwan, 2000).

While from our study we observed that the serotyped strain $O_{55}K_{59}$ was reported to be isolated in rate of 4.25% (**EL-Kady** *et al.*, 2004) from clinically affected broiler chickens. Therefore this study was carried out to study pathogenicity of *E.coli* O55 K59 to experimentally infected broiler chickens aged 5, 21 and 30 days.

MATERIAL AND METHODS

Experimental chicks:

One- day- old broiler chicks (150) of avian 43 as hatched were obtained from commercial hatchery. The chicks were reared on deep litter and fed on unmedicated starter commercial ration. These chicks were used for studying pathogenicity of isolated E.coli O_{55} K_{59} strain.

<u>Amoxicillin:</u>

The used drug was obtained from:

-Company: PHARCO pharmaceutica's as Amoxicillin Trihydrate chemically (6- $\{D\ (-)\ \alpha$ - amino- p- hydroxyl phenyl acetamidol $\}$ penicillinic acid trihydrate) Batch No 270400.

<u> Amoxiciilin medium:</u>

a-Amoxicillin nutrient broth media:

Amoxicillin broth medium was prepared by adding Amoxicillin in increasing concentrations (10 mg - 1000 mg/Lit. according to (Awaad, 1975). b- Amoxicillin MacConkey's agar:

One gram of Amoxicillin was added to milt sterilized MacConkey's agar at 45°C then distributed in Petri dishes. This medium used for reisolation of inoculated organisms from experimentally infected birds.

Bacterial isolate for Experimental infection:

a- E. coli O55 K59:

 O_{55} K_{59} was selected because it was isolated from flocks with high mortality rate and this isolate not fully discussed before.

b- Preparation of E.coli strain O = 5 K 59 resistant to Amoxicillin:

To prepare on *E.coli* strain O_{55} K_{59} resistant to amoxicillin using the method of (**Awaad, 1975**). This referred as Amoxicillin resistant *E. coli* (A R *E. Coli* O_{55} K_{59}).

c- Preparation of bacterial inoculum:

Three colonies of similar size were picked up for preparation of 24 broth culture, inoculated into nutrient broth and incubated for 24 hours at 37 °C. Then kept in refrigerator till bacterial count and use for experimental infection.

d-Plate counting:

The method of (Collins and Lyne, 1989) was adopted.

Dose for Experimental infection:

At 5 days old each chick was infected orally with 0.5 ml of standardized broth culture containing a dose of 2.5×10^7 CFU (**Awaad, 1972**) and (**Azzam, 1983**).

At 21 day old each chick was injected subcutaneously with 1.25 ml of standardized broth culture containing a dose of 1 x 10^8 CFU. (Singab, 1987).

At 30 days old each chickens was injected subcutaneously with 1.3 ml of standardized broth culture containing a dose of 3 x 10⁸ CFU (Aly, 1989).

The degree of pathogenicity was determined according to criteria reported by (Harry, 1964).

EXPERIMENT AND RESULTS

Experimental infection:

A total 150, one day-old chicks as hatched; were obtained from commercial source. The chicks were floor reared and feed on commercial balanced ration without feed additives.

At 1 st day of life 15 chicks were randomly collected, sacrificed, and samples from liver, heart blood, intestine and yolk sac were cultured to prove their freedom from E.coli O_{55} K_{59} . Rests of birds were divided into 3 groups (45 chicks) to be inoculated with E.coli O_{55} K_{59} ampicillin resistant at 5, 21 and 30 days.

At each time 45 birds were divided into 2 groups, Group I contain 25 chicks and group II contains 20 chicks, where 15 chicks out of group I (25 chicks) were infected while the other 10 were labeled and kept as contact. While birds of group II (20 chicks) were kept as non infected control group.

All experimental groups were kept under daily observation for 14 days for clinical signs and mortalities. Died birds after infection or sacrificed at the end of observation were subjected to post-mortem examination to record any gross lesions and try to reisolate inoculated *E.coli* from internal organs.

Body weight of infected birds aged 30 days were recorded at 35, 40, 43 and 46.

Results:

I- 5 days old chicks (Table 4):

Clinical Signs and mortalities:

- After 12 hours infected chicks showed signs of dullness, huddling together, depression and off food, while signs of sneezing, nasal discharge, inability to move, off food and rales were recorded at 24 hours. The recorded signs were developed to difficult respiration, coughing, nasal discharge, rhinitis, arthritis and decrease in body weight gain than control group at the 4th day, while at the 7th day infected chicks having diarrhea.
- Mortality started in infected group at 24 hours after inoculation where 3 birds were died followed by one bird at the 4th days with mortality rate of 26.67% while contact and control birds showed no mortalities.
- Contact birds showed signs of sneezing, nasal discharge and decrease in body weight gain than that of the control group, while no signs were observed on control negative birds.

Postmortem examination:

- Infected died birds 24 hours after experimental infection showed congestion of subcutaneous blood vessel, liver, spleen, petechial hemorrhages on liver and heart, while those died at the 4th day showed pericarditis, trachitis, perihepatitis, air sacculitis and unabsorbed yolk sac.
- Sacrificed birds, revealed lesions of trachitis, enteritis, enlarged gall bladder, unabsorbed yolk sac, fibrinous pericarditis, perihepatitis, airsaculitis and turbid air sac in infected, while only pericarditis, perihepatitis, airsacculitis and enlarged gall bladder were recorded 30% of contact.

Bacterial reisolation:

The inoculated *E.coli* could be reisolated from 100% in dead birds , 63.6% in sacrificed infected and from 50% in contact birds (Table 1).

- Regarding examined samples, the inoculated organism could be reisolated from all samples of only one bird while samples of liver and gall bladder were 100% positive, while only 75% of hearts, spleens and air sacs, were positive.
- Samples of sacrificed infected positive 7 birds organism was demonstrated of mainly in spleen (85.7%) followed by (57.1%) in air sac, (42.8%) in liver, (28.5%) in gall bladder and only (14.2%) of hearts, while Contact positive birds showed 100% of spleen, 40% of air sac and 20% for both liver and heart.

- Regarding positive organs to over all tested sacrificed infected it is clear that spleen showed the highest percentage (54.6) followed by air sac (36.4), liver (27.3), gall bladder (18.2) and heart (9.2), while sacrificed contact birds showed positive results in 50% of examined spleen, 20% in air sac, 10% in both liver and heart and 0%- of gall bladder.

II-21 days old chickens: (Table 2)

Clinical Signs and mortalities:

The infected birds started to show clinical signs at 12 hours after infection including general signs while signs of sneezing and nasal discharge were seen 24 hours after infection. Experimentally infected chickens showed nasal discharge, coughing, difficult respiration, inability to move, arthritis, decrease in feed consumption at the 48 hours from infection and stunted growth till the 7th day.

Contact birds showed no deaths while some chickens showed similar clinical signs to the infected.

Control negative group showed no detectable clinical signs allover the observation period (14 days).

One infected birds was died 24 hours post infection and other one at 48 hours with total mortalities of 13.33%, while both contact infected and control birds having no mortalities (0 %).

Post mortem examination:

- Birds dying 24 hours after experimental infection revealed septiceamic lesions while that died after 48 hours showed additional percarditis.
- Birds sacrificed at the end of experiment showed fibrinous pericarditis, periheaptitis, aisaculitis, enlarged gall bladder and enteritis in contact group some chickens showed pericarditis, distended gall bladder, turbid air sac and trachitis.

Bacteriological examination:

Inoculated *E.coli* could be reisolated from all examined organs of dead birds Table 2.

Sacrificed infected birds showed 11 positive birds from 13 in percentage of (84.6%) where the percentage of positive organs to over all examined was the highest (61.5%) from heart followed by (53.8%) from spleen, (46.1%) from liver and (30.7%) from both air sac and gall bladder.

Contact birds showed 70% bacteriologically positive birds with organ positivety of 50% in spleen, 20% in gall bladder and air sac and 20% in liver while heart samples showed 0% as shown in Table 2.

Regarding the percentages of positive organs to positive birds from sacrificed infected bird, it is clear that heart showed the highest isolation percentage (72.7%) followed by spleen (63.63%) then liver (46.1%), gall bladder and air sacs (36.36%). While organs of sacrificed contact birds

showed percentage of 20% for liver, gall bladder and air sac, 50% for spleen while heart was 0 %.

III- 30 days old chickens:

Clinical signs:

- 24 hours after experimental infection chickens showed depression, huddling together, dullness, off food, sneezing.
- 3 7 days some chickens showed coughing, sneezing, nasal discharge, rales difficult respiration, stunted growth and marked decrease in body weight gain than control group.
- Contact birds showed sneezing, nasal discharge and decrease in body weight gain while no signs were observed in control group.

No deaths were occurred in any group allover the observation period.

Post mortem examination:

P. M. examination 50% of sacrificed birds showed that infected group (50%) and 20% in contact group showed percarditis, air saculitis, enlarged gall bladder, enteritis as well as splenomegally (Table 3).

Bacteriological examination:

E.coli could be reisolated from sacrificed birds from 8 out of 15 infected birds (53.33%), only 2 birds showed positive isolation from all organs (13.33%), while over all liver showed the highest isolation rate (46.7%) followed by heart (33.3%), air sac (33.3%), gall bladder (26.6%) and spleen (26.6%).

Contact birds proved only 30% positive cases. Over all samples showed of liver, heart and spleen are positive reisolation as well as 10% of air sacs (Table 3).

Regarding the percentage of positive organs to positive birds from sacrificed infected birds, it is clear that liver showed the highest isolation percentage (87.5%) followed by both heart and air sacs (62.5%) for each then gall bladder and spleen 50% for each, while organs of sacrificed contact birds showed percentage of 20% for liver, heart and spleen followed by 10% for air sac then gall bladder 0%.

Regarding result of body weight (Table 4) infected birds showed lowest body weight (1058 gr) than contact birds (1082 gr) than control non- infected birds (1218 gr) at the 35 days, 5 days post infection in reduction rates of 13.1% and 11.2% in infected and contact as compared with control birds.

The reduction percentage in body weight was observed in all intervals where it reaches 18.6% and 10.4% at the 46th day of life (16 days post infection).

Infected and contact birds consumed 4.200 kg ration as compared with 4.6 kg in control group in 46 days of life.

The calculated conversion rate showed that the control negative birds showed conversion rate nearly higher (2.306) than the contact birds (2.356) then finally infected birds which showed the lowest rate (2.586).

DISCUSSION

Isolated *E. coli* serogroup O_{55} were chosen for studying pathogenicty, transmission of the *E.coli* O_{55} by contact and effect of infection on the body weight gain. {*E. coli* O_{55} was inoculated into broiler chickens at three different ages by different route of inoculation}.

Mortality rate was 26.7 % in 5 days old chicks were infected orally with 2.5×10^7 CFU of E. coli serotype (O_{55} K₅₉) (Table 1) and the percentage of positive reisolation was 73%. The inoculated serotype was reisolated from all dead birds (4 birds) with highest percentage (100%) from liver and gall bladder, followed by 75% for heart, spleen and air sac. While *E. coli* O_{55} K ₅₉ was reisolated from infected sacrificed birds at percentage of 63.6% with highest rate from spleen (54.6%) followed by air sac (36.4%), liver (27.3%), gall bladder (18.2%) the finally heart (9.1).

In relation to contact birds, inoculated isolate was reisolated in rate of 50% of sacrificed contact birds with over all highest reisolation rate from spleen 50% followed by 20% from air sac then 10% for each of liver and heart while the organism cannot be reisolated from gall bladder. It was observed that $E.\ coli\ (O_{55}\ K_{59})$ were localized in the liver and gall bladder in dead chicks at age 5 - 7 days while sacrificed birds at age of 18 - 20 days the best organ for reisolation were spleen followed by air sacs.

 $E.\ ccli\ (O_{55}\ K_{59})$ was inoculated subcutaneously into 21 days old chickens, the mortality was 13.3% in infected group while contact birds no mortality occur. $E.\ coli$ was reisolated from the two dead birds (100%) from heart and spleen followed by liver, gall bladder and air sacs 50% while O_{55} K₅₉ strains was reisolated from (84.6%) of infected sacrificed birds with highest reisolation rate from heart (61%) followed by spleen (53.8%), liver (54.1%). Then finally both gall bladder and air sacs (30.7%) for each.

In relation to contact birds, no deaths occurs and the inoculated isolate organism was reisolated from 70% of the birds with highest reisolation rate from spleen 50% followed by liver, gall bladder and air sac 20% for each and cannot be reisolated from heart. From (Table 2). It was observed that the percentage of reisolation of inoculated organism O_{55} from the spleen is highest in dead, sacrificed infected and sacrificed contact chickens.

No mortality was observed in both infected and contact chicken group aged 30 days old chickens and inoculated by subcutaneous route. The inoculated $E.\ coli$ strain, $O_{55}\ K_{59}$ was reisolated from 53.3% and 30% in infected and contact groups; respectively.

In relation to infected sacrificed birds the over all percentages of reisolation were highest in spleen 46.7% followed by both heart and air sac 33.3% finally both gall bladder and spleen 26.6%.

While contact sacrificed birds, the overall percentage of reisolation were highest in each liver, heart, spleen 66.6% for each followed by air sac 33.3% and the organism cannot be reisolated from gall bladder.

Body weight of chickens were recorded at 4 intervals of age (35, 40, 43 and 46) days and the conversion rates were recorded and there was a marked decrease in the mean body weight of infected chicken than that of both contact birds while conversion rates for negative control was the highest (2.306) than that of contact (2.356) and the infected birds show lowest conversion rate (2.586).

The results of clinical signs, post mortem, mortality, percentages of reisolation and effect on mean body weight of chickens indicated pathogenicity of inoculated *E. coli* O₅₅ K ₅₉ .Combating results of mortalities and severity of clinical signs was seen that age of birds at time of exposure and route of infection play a role in the above means. Where their was a some sort of age resistance could be seen in the area which come in agreement with these reported by Sojka and Caranphan, (1961); Awaad, (1972); Ibrahim *et al.*, (1998) and Azzam, (1983).

In conclusion E. coli O_{55} strain was of moderate pathogenicity to chickens of 5 days old when given orally and to chicken of 21 days old when given subcutaneously. While of low pathogenicity to chickens of 30 days old when inoculated subcutaneously. Also it was proved that the inoculated microorganism transmitted from inoculated group to contact one and the percentages of reisolation of the inoculated microorganism from contact groups at ages of 5, 21 and 30 days old were parallel to that of inoculated groups at the same ages.

Table (1): Results of *E. Coli* reisolation from dead infected, sacrificed infected and sacrificed contact 5-days old chickens.

	Bird	Infected dead 4 birds						Infected sacrificed 11 birds							Contact infected sacrificed 10 birds						
	No.	L	H	GB	SP	AS	Result*	Bird No.	L	H	GB	SP	AS	Result*	Bird No.	L	Н	GB	SP	AS	Result*
	1	+	_	+	+ :	+	+	1	-	-	+	+	+	+	1	-	-		+	+	+
	2	+	+	+	+	+	+	2	+	+	-	+	+	+	2	+	+.	 -	+	-	- i-
	3	+	+	+	-	+	+	3	+	-	-	-	+	+	3	-		-	+	-	+
	4	+	+	+	+		+	4	-		-	+	-	+	4	T-	_	-	+	-	+
								5	-	_	-	+	-	+	5	-	-	-	+	+	+
								6		-	+	+	+	+	6 - 10	-	-	-	-	-	5 - ve
	[7	+	-	·-	• +	-	+							
								8 - 11	_		-		-	5 - ve							·
Total +ve	4	4	3	4	3	3	4	11	3	1	2	6	4	7	10	1	1	-	5	2	5
	ositive	 					<u> </u>	 			 		 	<u> </u>						 	
organ to	positive ses	100	75	100	75	75	100		42.85	14.30	28.60	85.70	57.10	63.60		20 %	20 %	% 0	% 001	40 %	50 %
Over	all%								27.3	9.1	18.2	54.6	36.4		1	10	01	0	50	20	_

L = liver

H = Heart

GB = Gallbladder

SP = Spleen

AS = Air sac

* Results of reisolation

Table (2): Results of *E. Coli* O₅₅ K₅₉ from infected dead, infected sacrificed and sacrificed contact 21-days old birds.

	Bird No.	Infected dead 2 birds						Infected sacrificed 13 birds							Contact infected sacrificed 10 birds						
!		L	Н	GB	SP	AS	Result*	Bird No	L	Ĥ	GB	SP	AS	Result*	Bird No	L	Н	GB	SP	AS	Result*
	1	+	+	-	+	+	+	1	+	+	_	_	+	+	1	-	-	-	+	+	+
	2	-	+	+	+	-	+	2	-	_	<u> </u>	+	-	+	2	+		-	+	-	+
				 	 	1		3	-	+	+	-	-	+	3	-	-	+	-	-	+
		ļ	 					4	+	+	_	- -		+	4	-	_	-	+	 -	+
			,					5	+	+	+	+	+	+	5	-	_	+	-	-	+-
		 			,			6	-	+	-	-	+	+	6	-	_	-	+	+	+
								7	+	+	+	+	-	+	7	+	-	-	+	-	+
								8	+	+	-	+		+	8-10		-	-	-	-	-
		 				 		9	+		_			+						 	
								10		+	+	+	+	+							
	-					<u> </u>		11	+	-	-	_	2 -ve	-	1	1	<u> </u>		ļ	<u> </u>	
								12-13	-	-	-	-	-	4-							
Total +\e	2	1	2	1	2	1	2	13	6	8	4	7	4	11	10	2	0	2	5	2	7
ortan to	positive o positive ases	50	100 .	50	100	50	100		54.54	72.7	36.36	63.63	36.36	84.6		28.57	0	28.57	71.42	28.57	70
Over	r all%								46.1	61.5	30.7	53.8	30.7			20.0	0	20.0	50.0	20.5	i

L = liver

H = Heart

GB = Gallbladder

SP = Spleen

AS = Air sac

* Results of reisolation

Table (3): Results of reisolation of E. Coli O₅₅ K₅₉ from sacrificed infected and contact chickens 30-

days old.

	uays		Infe	cted sacri	ficed 15 b	oirds		Contact infected sacrificed 10 birds								
	Bird - No.															
		L	Н	GB	SP	AS	Result	Bird No.	L	Н	GB	SP	AS	Result*		
	1	+	-	-	+	-	+	1	-	+	-	+	-	+		
	2	-	+	+	-	-	+	2	+	+	-	_	+	+		
	3	+	+	-	_	+	+	3	+	-	_	+	_	+		
	4	+	+	+ +	+	+	+	4 -10	-	-	-	-	-	-		
	5	+	-	-	-	+	+	<u></u>	<u> </u>							
	6	+	+	-	•	+	+				-					
	7	+	-	÷	+ •	-	+	<u> </u>		1.	,		-			
	8	+	+	+	+ .	+	+									
	9-15	-	-	-	-	-	-						-			
Total +ve	15	7	5	4	4	5	8	10	2	2	0 .	2	1	3		
% of positive	e organ to cases	87.5	62.5	50	50	62.5	53.33		9:99	9.99	0.0	9:99	33.3	30%		
% of +v	% of +ve / all		33.36	26.6	26.6	33.3	. 1		20	20	0	20	10	1		

L = liver

H = Heart

GB = Gallbladder

SP = Spleen AS = Air sac * Results of reisolation

Table (4): Mean body weight (grams) and reduction percentages of subcutaneously infected chickens with $E.\ coli\ O_{55}\ K_{59}$ at 30 days age.

Age of birds		Infected birds			Control birds		
	Mean ± SD	Reduction	Reduction%	Mean ± SD	Reduction	Reduction%	Mean ± SD
35	1058 ± 84	160	13.1	1082 ± 32	136	11.2	1218 ± 157
40	1216 ± 39	366	23.1	1433 ± 72	149	9.4	1582 ± 64
43	1409 ± 175	357	20.3	1588 ÷ 130	180	10.2	1768 ± 148
46	1624 ± 127	370	18.6	1782 ± 126	208	10.4	1994 ± 177
Feed/gm	4200			4200			4600
Conversion rate	2.586			2.356			2.306

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الملخص العربي

المورة عنزة الميكروب القولوني K_{59} المبداري المسمون عمر K_{59} فيداري المسمون عمر K_{59}

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أستاذ ورئيس قسم أمراض الدواجن بكلية طب بيطرى بنى سويف جامعة القاهرة
** أستاذ مساعد أمراض الدواجن بكلية طب بيطرى بنى سويف جامعة القاهرة
*** أستاذ مساعد أمراض الدواجن المركز القومي للبحوث القي الجيزة

درست ضراوة معزولة من الميكروب القولونى نتتمى للمجموعة المصلية $O_{55}K_{59}$ لبدارى دجاج التسمين أعمار \circ ، ۲۱، \circ يوما بالتعرف على قدرة العترة على أحداث الأعراض الأكلينيكية ، الأنتقال الجانبى للدجاج المخالط وكذلك تأثير العدوى الصناعية على معدلات أكتساب الوزن.

أوضحت الدراسة أن العترة المستخدمة $O_{55}K_{59}$ كانت ضارية للدجاج المعدى صناعيا وأن هذه الضراوة تتأثر شدتها بعاملي العمر وطريقة العدوى كما أنها تتثقل جانبيا للدجاج المخالط.

أدت العدوى الصناعية لكتاكيت عمر ٥ أيام الى ظهور أعراض مرضية فى الكتاكيت المعدية والمخالطة وكانت نسبة النفوق ٢٦,٦٧% فى الدجاج المعدى. تم أعادة عزل الميكروب من الكتاكيت النافقة والمذبوح المعدى والمذبوح المخالط بنسب ١٠٠ %، ٢٣,٣٠%، ٥٠% على التوالى. بلغت نسب النفوق ١٣,٣٣% فى الدجاج المعدى بالحقن تحت الجلد عند عمر ٢١ يوما وكانت نسب أعادة العزل ١٠٠ %، ٢,٤٨%، ٥٠٠% من النافق والمذبوح المحقون والمخالط على التوالى. بينما لم تؤدى عدوى كتاكيت عمر ٣٠ يوما الى حدوث وفيات وكانت نسب أعادة عزل الميكروب من الدجاج المذبوح المعدى والمخالط عند نهاية فترة الملاحظة ٣٠٣٠%، ٣٠% على التوالى. كما تم ملاحظة الأعراض التنفسية عند عدوى أعمار ٢١، ٣٠ يوما وأن منحنى أعادة العزل من الدجاج المخالط للاعمار الثلاثة كان متماثلا مع ذلك فى الدجاج المعدى.

أوضحت الدراسة أن العدوى الصناعية لكتاكيت عمر ٣٠ يوما أدى الى أنخفاض معدل أكتساب الوزن عن المجموعة الحاكمة عند عمر ٣٥ ، ٤٠ ، ٤٣ ، ٤١ يوما لتصل الى ١٣,١%، أكتساب الوزن عن المجموعة الحاكمة عند عمر ٣٥ ، ٤٠ ، ٤٣ ، ٤١ يوما لتصل الى ١٣,١%، ١٣,١ المخالطة عن المجموعة الحاكمة غير المعدية. وكانت معدلات التحويل الغذائي في الدجاج المعدى الأقل (٢,٥٨٦) عن المجموعة المحلوعة الحاكمة (٢,٣٠٦) عند حساب هذا المعدل على عمر ٤٦ يوما من العمر.