Turkish Online Journal of Qualitative Inquiry (TOJQI)
Volume 12, Issue 8, July 2021: 813-819

Bacteriological Studies on Swollen Head Syndrome in Broiler Chickens

Ahmed H. Abed[1], Shaza Z. Mohammed[2]*,Khaled A, M. Shokier[3],Ismail A. Radwan[4]

[1]Bacteriology, Mycology and Immunology Department, Faculty of Veterinary Medicine, Beni-Suef University, Egypt.
[2*] Animal Health Research Institute Dokki, Beni-Suef branch, Egypt.

[3] Animal Health Research Institute Dokki, Beni-Suef branch, Egypt.

[4]Bacteriology, Mycology and Immunology Department, Faculty of Veterinary Medicine, Beni-Suef University, Egypt.
[*]**Corresponding author at:**Shaza Z. Mohammed**,**Animal Health Research Institute Dokki, Beni-Suef branch, Egypt.
Tel.: 010918116160

E-mail:Shazazeinelgaby@gmail.com.

Abstract

Swollen head syndrome (SHS) in poultry is a serious problem for the commercial broiler industry causing great economic losses.In the present study, the prevalence of SHS was studied in 500 broiler chickens and identification of the causative bacterial agents was conducted focusing. Samples were collected from the serous fluid from subcutaneous of the infra orbital sinuses. The prevalence rate of SHS in examined broiler chickens was 20%.The results of bacteriological examinationrevealed that 92% samples showed bacterial isolation, 72% of them had single bacterial isolates while 20% had mixed bacterial isolates. On the other hand,8% had negative bacterial isolation.A total of 112 bacterial isolates were recovered. *E. coli* was the most prevalent; recorded as 80 isolates (60 single and 20 mixed) followed by *P. aeruginosa*represented as 24 isolates (11 single and 13 mixed with *E. coli*), *Klebsiella* spp. represented as 4 isolates (one single 1% and 3 mixed with *E. coli*) and finally, *Salmonella* spp. represented as 4 isolates, all were mixed with *E. coli*.Results of serogrouping of *E. coli* isolates revealed that 6 O-serogroups were obtained of which O_{119} was the most prevalent (30%) followed by O_{18} (23.8%), O_1 (16.3%), O_{125} (15%), O_{158} (8.6%), and finally O_{169} (6.3%).On the other hand, serotyping of *Salmonella* isolates revealed the existence of the only serotype S. Kentucky; identifies in 50% of isolates, meanwhile 50% of isolates were untyped.

Keywords: *Broiler chickens,Swollen head syndrome, E. coli,P. aeruginosa,Klebsiella spp.,Salmonella.*

INTRODUCTION

Swollen-head syndrome (SHS) is acute respiratory disease affecting the upper respiratory tract of domestic poultry; includingbroilers and broiler breeders, characterized by swelling of headand facial edema;in periorbital and infraorbital sinuses, resulted from subcutaneous

accumulation of inflammatory exudatesin the head in response to secondary bacterial infection; usually *E. coli*, following the initial upper respiratory viral infection(**Shawki, *et al.*, 2017 and Abdelmoez*et al.*, 2019)**.SHS was first described in the South America **(Morley and Thomson, 1984)**and it has becomean important avian problem all over the world in the last few years **(Abdelmoez*et al.*, 2019)**.SHS can affect chickens of all ages mainly 4-6 weeks old causing considerable economiclosses in the avian industry due tomorbidity rate reaching to 10% and mortality about 3-4% as well as 2-3% reduction in the egg production **(Osman, *et al.*, 2015)**.

Although the etiology of SHS is uncertain anditsclinicalappearanceisquitevariable,it is a multi-factorial disease.SHS may be resultedfromamixedinfectionwith *E. coli* and paramyxovirus, coronavirus, or pneumovirus in which the initialviralinfection causes acute rhinitis that permits *E. coli* to invade the subcutaneousfacialtissues**(Osman, *et al.*, 2015)**.The severity of the disease depends on presence of some environmental factors such as accumulation of ammonia, dust, overcrowding and bad ventilation**(Abdelmoez*et al.*, 2019)**.In addition to *E. coli*, therearemanypathogenshadfarrelationwithSHSas *Pseudomonas* spp., *Klebsiella*spp. and *Enterococcus* spp.**(Osman, *et al.*, 2015)**.

The present study aimed to investigate the prevalence of SHS in broiler chickensas well as identification of the causative bacterial pathogens.

MATERIAL AND METHODS

Chickens.

A total of 500 diseased broiler chickens of different ages (2-5 weeks) from different farms in Beni-Suef and El-Fayoum Governorates were subjected to the present study during the period from January 2018 up to May 2019. These chickens were subjected to clinical and postmortem examinationsto detect SHS lesion.

Samples.

A total of 100 serous fluid samples collected were collected aseptically from subcutaneous of the infra orbital area in the affected head of slaughtered diseased and freshly dead chickens.

Bacteriological examination.

The collected samples wereaseptically inoculated into tryptone soya broth and incubated aerobically at 37˚C for 24 hrs. Then a loopful of the broth culture was streaked onto tryptone soya agar and MacConkey's agar and incubated aerobically at 37˚C for 24-48hr. The lactose fermenting (pink) colonies were further inoculated onto eosin methylene blue (EMB) agar medium and incubated at 37˚C for 18-24 hrs while pale colonies were streaked onto xylose lysine desoxycholate(XLD) agar and incubated aerobically at 37˚C for 24 hrs. Also, the green coloured colonies were further inoculated onto cetrimide agar medium and incubated at 37˚C for 18-24 hrs.

Identification of bacterial isolates.

Morphological and biochemical identification

All the recovered isolates were identified morphologically and biochemically according to schemes described by **Collee *et al.* (1996)** and **Quinn *et al.* (2011)**. The following tests were used; oxidase, catalase, indole, methyl red,VogesProskauer, citrate utilization, urease,H_2S production on TSI,nitrate reduction and sugar fermentationfor glucose, lactose, sucrose, mannose, arabinose, maltose and mannitol. Other non-biochemical tests including pigment production, motility test and haemolysis onto blood agar were applied.

Identification by using Microbact(GNB 12A) Gram-negative system.

The appropriateMicrobactkit (Microbact24E, Oxoid) was used for identification of the isolates of Family *Enterobacteriaceae*members and other Gram-negative bacteria. Microbactstrips should only be used to identify pure cultures. It was used according to the manufacturer's instruction.

Serological identification:

Serogrouping of *E. coli* isolates. *E. coli* isolates were serogrouped by slide agglutination test using standard polyvalent and monovalent *E. coli* antisera according to **Quinn *et al*. (2011)**.

Serotyping of *Salmonella*. *Salmonella* isolates were serotyped by slide agglutination test using diagnostic polyvalent and monovalent O and H *Salmonella* antisera**(DENKA SEIKEN Co., Japan)** according to Kauffman-white scheme **(Quinn *et al*., 2011)**.

RESULTS

Prevalence of SHS in the examined broiler chickens.

Out of 500 broiler chicken, 100ones (20%) showed SHS lesions in clinical and PM examination.

Bacteriological examination.

The results represented in **table (1)** revealed that out of 100 serous fluid samples collected from subcutaneous of the infra orbital area in the broiler chicken heads, 92 samples (92%) revealed bacterial isolation, of which 72 samples (72%) had single bacterial isolates while 20 samples (20%) had mixed bacterial isolates.On the other hand,8 samples (8%) had negative bacterial isolation.

Table (1): Results of bacteriological examination of samples collected from broiler chickens with SHS.

No. of samples	Positive bacterial isolation						Negative bacterial isolation	
	Single isolates		Mixed isolates		Total			
	No.	%	No.	%	No.	%	No.	%
100	72	72	20	20	92	92	8	8

%: was calculated according to the number (No.) of the samples.

Prevalences of different bacterial pathogens recovered from SHS lesions in broiler chickens.

Form all collected samples, a total of 112 bacterial isolates were recovered.*E. coli* wasthe most prevalentrepresented as80 *E. coli* isolates (80%); of which 60 single (60%) and 20 (20%) mixed with other bacteria. Then, *Pseudomonas aeruginosa*represented as 24 isolates(24%); of which 11 single (11%) and13 (13%) mixed with *E. coli*.Afterthat, *Klebsiella* spp.represented as 4 isolates; of which one single (1%) and3 (3%) mixed with *E. coli*. And finally, *Salmonella* spp.represented as 4 isolates(4%);all were mixed with *E. coli***(Table 2)**.

Table (2): Prevalence of different bacterial isolates from SHS in broiler chickens.

Bacterial isolate		No. of isolates	%
Single isolates	***E.coli***	60	60
	P. aeruginosa	11	11
	***Klebsiella* spp.**	1	1
	Total single isolates	72	72

Mixed isolates	***E.coli* + *P. aeruginosa***	**13**	**13**
	***E.coli* + *Salmonella* spp.**	**4**	**4**
	***E.coli* + *Klebsiella*spp.**	**3**	**4**
	Total mixed infection	**20**	**20**
Negative bacterial isolation		**8**	**8**
Over all total		**100**	**100**

%: was calculated according to the overall total number (No.) of the samples(*n=100*).

Serogrouping of *E. coli* isolates.

Out of 80 *E. coli* isolates, 6 O-serogroups were obtained. The serogroup O_{119} was the most prevalent represented in 24 isolates (30%) followed by serogroups O_{18}(*n=19*; 23.8%), O_1 (*n=13*; 16.3%), O_{125}(*n=12*; 15%), O_{158}(*n=7*; 8.6%), and finally O_{169}(*n=5*; 6.3%)**(Table 3)**.

Table (3): Serogroups of *E. coli* recovered from swollen head broiler chickens.

***E. coli*Serogroup**	**No.**	**%**
- **O_{119}**	**24**	**30**
- **O_{18}**	**19**	**23.8**
- **O_1**	**13**	**16.3**
- **O_{125}**	**12**	**15**
- **O_{158}**	**7**	**8.6**
- **O_{169}**	**5**	**6.3**
Total No. of isolates	**80**	**100**

%: was calculated according to the total number (No.) of isolates (*n=80*).

Serotyping of *Salmonella* isolates.

Out of 4 *Salmonella*enterica isolates, only oneserotypes was identified as *Salmonella* Kentucky which were represented in 2 isolates (50%) meanwhile the other 2 isolates (50%) were untyped with the available antisera.

DISCUSSION

Poultry industry has reached great levels of development and became one of the most dynamic areas of agriculture. Poultry is the main protein feed of animal origin for human consumption worldwide **(Milsavljevic, 2014)** due to efficiency of production cost as well as its short life cycle. In Egypt, great efforts were directed toward the poultry industry to meet the high requirements of animal protein.

SHS is a disease of upper respiratory tract and considered as one of these problems in last few years. The disease affects broilers and broiler breeders which resulted in inflammatory exudate beneath the skin **(Seifi *et al.*, 2015)**.SHS has been described as a multifactorial disease where the initial lesion mainly caused by viruses while the clinical signs are a consequence of secondary bacterial infections and the severity of the disease depends upon environmental factors **(Abdelmoez*et al.*, 2019)**.In this study, the prevalence of SHS in diseased 500 broiler chickensand identification of the different causative bacterial agents wereinvestigated.

In the present study, the resultsrevealed that 20% of diseases chickens showed SHS

lesions in clinical and PM examination.Such prevalence wasnearly similar to that recoded by **Georgiades*et al.*(2001)** who found that from 50 commercial broiler flocks concerning respiratory disease, signs of SHS were shown in 8 birds (16%).

In the current study, the results of bacteriological examinationrevealed that92% of serous fluid samples collected from subcutaneous of the infra orbital sinusesof the broiler chickens showed bacterial isolation,72% of them had single bacterial isolates while 20% had mixed bacterial isolates.On the other hand,8% had negative bacterial isolation. These results are coincided with those obtained in Egypt by**Abdelmoez*et al.* (2019)** who recorded bacterial isolation from all samples of chickens with SHS (100%) of them65% had single bacterial isolates while 35% had mixed bacterial isolates. Also, **Amer*et al.* (2019)** found that all heads samples were positive for bacteriological examination.

Regarding the prevalences of different bacterial pathogens recovered from SHS lesions in broiler chickens, the results illustrated in **table(2)** showed that a total of 112 bacterial isolates were recovered.*E. coli* was the most prevalent; recorded as80 isolates (60 single and 20 mixed) followed by*P. aeruginosa*represented as 24 isolates (11 single and 13 mixed with *E. coli*),*Klebsiella* spp. represented as 4 isolates (one single 1% and 3 mixed with *E. coli*) and finally, *Salmonella* spp. represented as 4 isolates, all were mixed with *E. coli*.Such results were nearly similar to those recorded by **Abdelmoez*et al.* (2019)** whofound that28/40 samples were positive for *E. coli*isolation (16 single and12 mixed) and10/40 samples were infected had*P. aeruginosa*isolates (4 single, 6 mixed with*E. coli*or*P. mirabilis*). Also, **Osman, *et al.*(2015)** examined bacteriologically 250 samples of SHS from broiler chicken and isolated 70*E.coli*(28%), 7*K. pneumoniae*(2.8%) and 5 *P. aeruginosa*(2%).**Georgiades*et al.*(2001)**found that *E. coli* was the most prevalent isolate from infraorbital sinuses of the affected birds as 87.5% of samples. Moreover,**Hafez and Löehren (1990)** and **Al-Ankari*et al.* (2004)** isolated pure *E. coli* culture from cases of SHS.

Additionally, in the current study there was an unprecedented result where 4*Salmonella* spp. (4%) were recovered from SHS lesions and that was considered; as we believe, the first record for *Salmonella*spp. as a cause of SHS.

Results of serogrouping of *E. coli* isolates were illustrated in **table (3)**.Six O-serogroups were obtained of which O_{119} was the most prevalent (30%) followed by O_{18} (23.8%), O_1 (16.3%), O_{125} (15%), O_{158} (8.6%), and finally O_{169} (6.3%).The distribution of O antigens was nearly similar to that reported in previous studies **(Gomis*et al.*, 2001;Schouler*et al.*, 2012 and Hasan *et al.*, 2020)** who recovered nearly the same serogroups; beside other serogroups. On the contrary they differed from those obtained by **Wang *et al.* (2010)** who recovered 8 different serogroups; O_{65}, O_{78}, O_8, O_{120}, O_2, O_{92}, O_{108}, and O_{26}, and **Tana*et al.* (2013)** who recovered 8 serogroups*E. coli* including O_2, O_8, O_{15}, O_{73}, O_{86}, O_{102}, O_{115} and O_{139}.

On the other hand, serotyping of *Salmonella* isolates revealed the existence of the only serotype S. Kentucky; identifiesin 50% of isolates, meanwhile 50%of isolates were untyped.These results run parallel to those recorded by **(Radwan*et al.*, 2016 andandHasan *et al.*, 2018)**as *S.* Kentucky was one of the most frequent broiler chicken isolated serovars. Also, **Helal*et al.*, (2019)** isolated different *salmonella* serotypes and found that 12.5% of isolates were*S. Kentucky*.

CONCLUSION

Swollen-head syndrome is a serious problem for the commercial broiler industry causing great economic losses.The prevalence rate of SHS in examined broiler chickens was 20%. *E. coli* is the most prevalent bacterial agent causing SHS followed by*P. aeruginosa*, *Klebsiella* spp. and*Salmonella* spp.

REFERENCES

1. Abdelmoez, N. S.; Shawky, M. M.; Abdelhady, H. A.; Lebdah, M. A. and Salama, S. S. (2019): Isolation and identification of some possible causative agents of swollen head syndrome (SHS) in broiler chickens in Egypt. Slov. Vet. Res.; 56 (22): 781–788. doi:10.26873/SVR-819-2019
2. Al-Ankari, A.S.; Al-Ramadan, A.M. and El Demerdash (2004): Risk factors associated with prevalence of swollen head syndrome (SHS) in broiler chickens in eastern province-Saudi Arabia. Int. J. Poult. Sci.; 3 (10): 646–650.
3. Amer, M.M., Mekky, H.M., Fedawy, H.S., Elbayoumi, Kh. M., &Sedeek, D.M. (2019): Antibiotic Profile of Bacterial Species Isolated from Broiler Chickens with Cellulitis. World's Veterinary Journal, 9(4): 268-279.
4. Collee, J. G.; Miles, R. S. and Watt, B. (1996): Tests for identification of bacteria, In: Collee, JG, Mackie, TJ andMcCartney, JE (Eds.) Mackie &Mccartney Practical Medical Microbiology. Elsevier (A Divisionof Reed Elsevier India Pvt. Limited), New York, USA.
5. Georgiades,G.; Iordanidis, P. and Koumbati,M. (2001):Cases of Swollen Head Syndrome in Broiler Chickens in Greece.Avian Diseases, 45(3):745-750.
6. Georqiades G., Iordanidis P., Koumati M. (2001): Cases of S-H-S in broiler chickens in Greece. Avian dis.; 45 (3): 745–50.
7. Gomis, S. M.; Riddell, C.; Potter, A. A. and Allan, B. J. (2001): Phenotypic and genotypic characterization of virulence factors of *Escherichia coli* isolated from broiler chickens with simultaneous occurrence of cellulitis and other colibacillosis lesions. Can. J. Vet. Res., 65:1-6.
8. Hafez, H.M. and Löehren, U. (1990): Swollen head syndrome: clinical observations and serological examinations in West Germany. Dtsch. Tierarztl. Wochenschr.; 97: 322–324.
9. Hasan, W. H.;Abd El-Latif, M.A. A. and Abed, A. H. (2020): Bacteriological and molecular studies on *E. coli* isolated from broiler chickens. Assiut Vet. Med. J.; 66 (16): 34-47.
10. Hassan, W.H.; Abed, A.H.; Thabet, A.; El Nady, E.A.M. (2018): Genetic analysis of multidrug resistant Salmonella isolated from broiler chickens. *J Vet Med Res, 25 (1)*: 121-131. doi: 10.21608/JVMR.2018.43309
11. Helal, G.; Tarabees, R.; Younis, G. (2019): Molecular characterization of virulence genes associated with *Salmonella* spp. isolated from poultry. Journal of Current Veterinary Research, 1(2):36-46.
12. Milsavljevic, T. (2014): Ascites poultry. J. Dairy Vet. Anim. Res., 1(2):18-20. doi: 10.15406/jdvar:2014.01.00006.
13. Morley, A.J. and Thomson D.K. (1984): Swollen head syndrome in broiler chickens. Avian Dis. 28:238-243.
14. Osman, K., A. Orabi, I. radwan, and monaarfat (2015): Antibiogram for Major Pathogens Recovered From Broilers swollen Head Syndrome", Journal of Global Bioscience, 4: 2650-2655.
15. Quinn PJ, Markey BK, Leonard FC, FitzPatrick ES, Fanning S and Hartigan PJ, 2011. Veterinary Mcrobiology and Microbial Disease, 912-912 pp.
16. Radwan, I.A.; Abed, A.H.; Abd Al-Wanis, S.A.; Abd El-Aziz, G.G (2016): Antibacterial effect of cinnamon and oreganium oils on multidrug resistant *Escherichia coli* and *Salmonellae* isolated from broiler chickens. *J Egy Vet Med Ass, 76 (2)*: 169-186.

17. Schouler, C.; Schaeffer, B.; Bree, A.; Mora, A.; Dahbi, G.; Biet, F.; Oswald, E.; Mainil, J.; Blanco, J. and Moulin-Schouleur, M. (2012): Diagnostic strategy for identifying avian pathogenic *Escherichiacoli* based on four patterns of virulence genes. J. Clin. Microbiol.; 50: 167-1678.
18. Seifi, S. and Boroomand, Z. (2015): The role of avian metapneumo-virus in respiratory complex disease circulating in broilers in Northern Iran. Trakia J. Sci.; No 2, pp 175-179.
19. Shawki, M. M.; Lebdah, M. A.; Shahin, A. M. and Nassif, S. A. (2017): Some Studies on Swollen Head Syndrome in Broiler Chickens in Egypt. Zagazig Veterinary Journal, 45 (1): 132-141.
20. Tana, S. K.; Elena, C.; Pranvera, A. S. and Antonio, C. (2013): Serotyping and biotyping prevalence of avain pathogenic *E. coli* in Albanian poultry industry. Agriculture University of Tirana, Faculty of Veterinary Medicine, Albania and UniversitadegliStudi "Aldo Moro", Facolta di MedicinaVeterinaria, Italy 1098-1104.
21. Wang, X. M.; Liao, X. P.; Zhang, W. J.; Jiang, H. X.; Sun J.; Zhang M. J.; He X. F.; Lao, D. X. and Liu, Y. H. (2010): Prevalence of serogroups, virulence genotypes, antimicrobial resistance, and phylogenetic background of avian pathogenic *Escherichia coli* in south of China. Food borne Pathog Dis.; 7(9):1099-1106.