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Pneumonia in domestic rabbits in Egypt  
strain types and methods of control.

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INTRODUCTION

Respiratory ailments are common among domestic rabbits in Egypt. In rational production , they essentially strike breeding adults. In a farm rabbitary young rabbits can also be affected (*Webster , 1924 , 1925*). Where such ailments are endemic , losses are especially to be feared among the females in which the disease become chronic , leading to production stoppages and mortalities among the nursing youngs (*Weisbroth et al. , 1974* ). Respiratory diseases usually remain endemic but abrupt epidemics which can decimate the flock in a few weeks sometimes break out in farm rabbitaries (*Griffen , 1952* ).

Pasteurellosis caused by *Pasteurella multocida* is one of the most important pathogens inducing respiratory problems in farm rabbitaries in Egypt (*Gergis et al. , 1992* ). A number of clinical forms of infection can occur including upper respiratory infection ( Snuffles ) ; otitis media ; enzootic pneumonia , conjunctivitis ; pyometra ; orchitis ; abscesses and septicaemia (*Digiacoimo et al. , 1983* ). Commercial rabbitaries suffer economic losses due to Pasteurellosis and concomitant infection of laboratory rabbits complicate research projects.

The purpose of the study reported here is to review the most important findings of intensive research conducted in our laboratory concerning the bacterial species causing pneumonia in rabbits ; the serotypes of *P. multocida* isolated from rabbits affected with different forms of Pasteurellosis and the antibiogram of the isolated organisms. Also the methods used for controlling and preventing Pasteurellosis are presented.

## MATERIALS AND METHODS

### Samples :

A total of 129 lung samples of dead rabbits of different sex and ages were examined by both gross observation and bacteriological examination.

Also 312 samples were collected from the internal organs (heart blood , liver , middle and internal ear and bone marrow) of ailing and recently dead rabbits. As well nasopharyngeal swabs were collected from apparently healthy and clinically diseased rabbits from different rabbitaries at various localities.

### Bacteriological examination :

All samples were subjected to bacteriological examination. The recovered bacteria were identified using the technique described by *Cowan and Steel (1966)*.

### Antibiotic sensitivity of the isolated bacteria :

The disc diffusion method of *Bauer et al.* (1966) was used and the results were interpreted by the criteria of *Difco Supplementary Literature* (1980).

### Serotyping of *Pasteurella multocida* isolates :

Capsular and somatic typing were conducted by the techniques of *Carter* (1955) and *Heddleston et al.* (1972).

### Vaccine preparation and evaluation :

A polyvalent vaccine was prepared from serovars A:B ; A:12 and D:11 of *P. multocida* which were the most common isolates among diseased rabbits. Anti - *Pasteurella multocida* antibodies were assayed in sera of vaccinates using the indirect haemagglutination test described by *Carter and Rappay* (1962). The immunity of vaccinated candidates were challenged by exposure to virulent *P. multocida* organisms used in vaccine preparation.

## RESULTS

Pneumonia was the prominent pathological lesion observed among 51 % of Zagazig province examined samples. Meanwhile 59 % of the examined dead rabbits from Giza province had gross pneumonic lesions ( Table 1 ). The grossly noticed lung lesions were either

consolidation ( 63 % ) atelectasis ( 32 % ) or abscesses with abundant yellowish white caseous pus filling most of the chest cavity ( 5 % ). Pneumonia was most predominant among the examined adult females ( 84 % ) than adult males ( 24 % ). Juvenile males had the lowest prevalence ( 8 % ) ( Table 2 ).

Out of the 129 lung samples examined bacteriologically bacterial species could be isolated from 119 ( 92.2 % ) of the examined samples , while the remaining samples were culturally negative. Seven bacterial species could be isolated as causative agents of single infection. They were tabulated in an order of frequency in table ( 3 ). Regarding the incidence of mixed bacterial infection recovered from pneumonic lungs of dead rabbits *Bordetella bronchiseptica* was the most frequently isolated organisms ( 33.61 % ) followed by *P. multocida* ( 27.7 % ). The incidence of the other bacterial species were illustrated in tables ( 3 and 4 ).

As shown in table ( 5 ) capsular and somatic serotyping of the isolated *P. multocida* strains revealed that ( 93 % ) of the isolates were capsular type A and only ( 7 % ) were of capsular type D somatic typing revealed the presence of seven O groups ( 1 , 3 , 4 , 7 , 9 , 11 and 16 ). Combination of both typing systems indicated that the most predominant serovars were A:3 ( 21.4 % ) ; A:12 ( 37.1 % ) and D:11 ( 11.4 % ).

Table ( 6 ) illustrated the antibiotic sensitivity of the differently isolated bacterial species.

Results tabulated in Tables ( 7 and 8 ) showed that comparable titers to type A and D P. multocida were noted in both groups of rabbits after either primary or secondary vaccination with the experimentally prepared polyvalent vaccine. Challenge test results were given in Table ( 9 ).

### DISCUSSION

In Egypt respiratory diseases occur frequently in domesticated rabbits both in colonies maintained for research purpose or human consumption ( *Gergis et al. , 1992* ). The results of this investigation revealed that pneumonia regularly caused clinical manifestations and deaths with an incidence ranging from 51-59 %. These findings were consistent with those of *Flatt and Dungworth (1971)*.

The higher prevalence of pneumonia in older rabbit females (78 %) may be due to the higher female to male ratio in rabbitaries and the result of longer duration of exposure to the organism. This sex associated differences was not observed in Juvenile rabbits.

Regarding the causative bacterial agents of pneumonia P. multocida was the most frequently isolated bacteria ( 82.3 % ) in

single bacterial infection while *Bord. bronchiseptica* was more common in mixed infection. These data agreed with the previous observations of *Hagan (1958)* , *Mushia and Schoenbaum (1980)*. Okerman (1988) stated that practically all rabbits are infected with *B. bronchiseptica* and they usually experience very few ill effects from it , although histological lesions are presented in the trachea and bronchi. He also noted that pneumonia caused by *B. bronchiseptica* is very rare and the infection can by no means be considered of great economical importance. Our trials to reproduce pneumonia in experimental rabbits by *B. bronchiseptica* were unsuccessful.

Serotyping results of the isolated *P. multocida* organisms reported in this study ( Table 5 ) were concomitant with those of *Brogden (1980)* and *Hippe and Schliesser (1981)*.

Titration of anti - *Pasteurella multocida* antibodies at different intervals after vaccination with the prepared polyvalent inactivated vaccine and challenge test results given in Tables ( 7 , 8 and 9 ) revealed that rabbits could be effectively immunized with this vaccine. Similar findings were also reported by *Cameron and Gertruida Smit (1970)*.

Pneumonic pasteurellosis could be effectively controlled in affected rabbitaries by eliminating of diseased animals ,

controlling the climate by ensuring minimal temperature fluctuations , good ventilation and no draughts. Our experimented studies have also shown that the infection taken hold more easily and produce a more serious illness when the ammonia concentration is high.



## SUMMARY

Out of 119 rabbit carcasses with pneumonics examined bacteriologically 7 bacterial species could be isolated as causative agents of single infection. *Pasteurella multocida* was isolated among other bacterial species in 27.7% of pneumonic lungs. Seventy isolates of *P. multocida* were serotyped. Capouler typing revealed that 93% were type A and 7% belonged to type D. Somatic typing revealed the presence of seven Ogroups. Combination of both typing systems revealed that A:3, A:12 and D:11 were the most frequent isolates. A polyvalent adjuvanted vaccine prepared from these isolates afforded protection in vaccinated rabbits against septicaemic pasteurellosis.

Table (1): Incidence of pneumonia in dead rabbits.

Origine of rabbits	Number examined	% affected with pneumonia
Giza	49	59 %
Zagazig	70	51 %

Table (2): Prevalence of pneumonia among rabbits according to age and sex.

Juvenile rabbits		Adult rabbits	
Males	females	males	females
8%	9%	42%	84%

Table (3): Incidence of aerobic bacteria in pneumonic lungs of dead rabbits.

Microorganisms	Incidence (%)		
	Pure culture	Mixed culture	Total
Past. multocida	54.6 %	27.7 %	82.3 %
Bord. bronchiseptica	41.17%	33.61%	74.78%
Kleb. pneumoniae	9.2 %	15.9 %	25.1 %
Staph. aureus	8.4 %	13.44%	21.84%
E. coli	6.72%	12.06%	19.32%
Salmonella	4.20%	10.92%	15.12%
Strept. pyogens	3.36%	10.08	13.44%
Citrobacter	0.0 %	9.2 %	9.2 %
Moraxella	0.0 %	8.4 %	8.4 %
Bseud. aerogenosa	0.0 %	7.8 %	7.8 %
Neisseria species	0.0 %	5.04%	5.04 %

Table (4): Incidence of the most common organisms isolated in mixed infection .

Organism	Incidence
Past.mültocida + Staph. aureus	27.7%
Bord. bronchiseptica + Strept.pyogens	22.11%
: : + Niessleria sp.	11.50%
E. coli + Citrobacter	9.2 %

Table (5): In vitro sensitivity of the most common bacteria isolated from pneumonic lungs of dead rabbits.

Antibiotic	Conc. per disc	Past mitocida	Bord. bronchiseptica	Kleb. pneumoniae	Staph. aureus	E. coli	Salmonella
Penicillin G	10 units	91.83%	59.15	63.33	7.6	56.52	44.49
Streptomycine	10 ug	29.54%	43.66	70.00	38.46	78.26	72.22
Erythromycine	15 ug	100%	35.21	50.00	50.00	34.73	38.88
Lincomycine	2 ug	51.02%	40.04	23.33	57.69	25.08	33.33
Oxytetracycline	30 ug	95.91%	100.00	86.66	73.07	47.82	50.00
Chloramphenicol	30 ug	100.00%	67.60	80.00	92.30	100.00	100.00
Halidinic acid	300 ug	96.93%	71.83	60.00	65.38	100.00	88.88
Ampicillin	10 ug	52.04%	100.00	73.33	34.61	65.21	50.00
Gentamycine	10 ug	100%	100.00	100.00	100.00	82.60	72.22
Cephaloxin	30 ug	100%	92.95	100.00	100.00	78.26	55.55
Coltrimoxazole	300 ug	45.91%	63.38	76.66	100.00	73.91	61.11

Table (6): The number and percentage of different Pasteurella multocida serovars isolated from rabbits at Kaluobia Governorat.

isolated serovars	No. of isolates	%*
A:1	6	8.5%
A:3	15	21.4%
A:4	6	8.5%
A:7	8	11.4%
A:9	2	2.8%
A:12	26	37.1%
A:16	2	2.8%
D:1	1	1.4%
D:3	2	2.8%
D:4	1	1.4%
D:14	1	1.4%

\*percentage was calculated according to the total No. of isolates (70 isolates).

Table 7 - Geometric mean titers of anti-Past.multocida type A antibodies at different intervals after vaccination of rabbits with polyvalent pasteuruella vaccine.

Type of adjuvant added to vaccine	Pre vaccination	* GMT of antibodies to type A Past. multocida			
		Days after vaccination			
		boostering			
		15	30	45	60
Alum	5	86	343	755	1576
Oil	5	80	320	788	1689

Table(8) : Immune response of rabbits vaccinated with polyvalent Past.multocida vaccine as measured by IHA test using type D Past.multocida antigen

Type of adjuvant added to vaccine	Pre vaccination	* GMT of antibodies to type A Past. multocida			
		Days after vaccination			
		boostering			
		15	30	45	60
Alum	5	53	226	640	1280
Oil	5	57	211	680	1372

Table(9) : Indirect haemagglutination titers and protection values from immunized rabbits challenged with Past.multocida strains.

Type of vaccine	Challenge strains	IHA titers at time of challenge	Percent protection
Alum ppt. vaccine	Type 3:A	1576	80
	Type 12:A	1576	100
	Type 11:D	1280	80
Oil adjuvant vaccine	Type 3:A	1689	80
	Type 11:A	1689	100
	Type 11:D	1372	90
Non vaccinated controls	Type 3:A	0	0
	Type 12:A	0	10
	Type 11:D	0	0

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