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Inter and Intraserotype Variations of Streptococcus Pneumoniae Pathogenicity in Mice and Rabbits

AL-Janabi,AA J [a], Shnawa IMS [b,c*] and AL-Sadi AM [d]

[a] Department of Biology, College of Science, University of Kufa, Iraq.
[b] Department of Medical Biotechnology, College of Biotechnology, Al-Qasim Green University, Qasim, Babylon, Iraq.
[c] College of Nursing, University of Hilla, Babylon, Iraq.
[d] Islamic University, College of Health Department Technicians, of Medical Analysis, Najaf Branch, Najaf, Iraq.

Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

The small mammalian labratory animals, the mice and the rabbits were tempted to be pathogenicity models for *S. pneumoniae* serotypes 1 and 6, the infecting doses were 0.3ml for mice and 0.5 ml for rabbits. These infectious doses were rated to a count of 1X 10 to 7 CFU/ml. The infection routes were intrapreritoneal for mice and intranasal for rabbits. The gold standard criteria for pathogenicity were the death end point DEP.DEP was up to 24hr for mice and ranged from four to 14 days in rabbits. Mice was more susciptable than rabbits for *S. pneumoniae* serotypes infections. Autopsy cultures of blood, lungs, liver and kideys were producing same infecting serotype both from mice and rabbits. This isolation profile was indicating bacterimic state induced by infection. Different

**Corresponding author: E-mail: ibrahimshnawa3@gmail.com;*

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serotypes have shown different death end points duration time. There were inter and intraserotype variations in DEP. Moderate to severe respiratory disease conditions RDC was noted in mice while mild RDC in rabbits. Both of the tested models were similar lung histology to man. The histological features of this experimental infections have shown that the infected mice and rabbits were of bronchopneumonia type. It appears that both mice and rabbits were standing as valid laboratory animal models for these local clinical isolates of *S. pneumoniae* serotypes.

Keywords: Capsule; death end point; pneumococcal disease; bacterial virulence; respiratory disease; serotype variations.

1. INTRODUCTION

Bacterial eco-pathotype was showing variations in pathogenicity to laboratory animal models within the same pathogen species and /or serotypes. A theme proved right now both in man and small mammalian laboratory animals.Host health state, age, nutrition, genetic makeup, and other environmental insults do affect variations in the pathogenic potentials of human pathogenic bacteria. T hese infleunces are smoothly true for the pathogenicity of S.penumoniae in man and laboratory animals. Streptococcus pneumoniae pathogenicity in man and laboratory animals are mediated by either one or more of the following; Pneumolysin and IgA splitting enzyme, bacterial subunit structures like capsules, bacterial-host body immune reactions, serotype phase variations and virulence genes [1-10]. The objective of the present work was to report on an inter and intraserotype variations in pathogenicity of *S.pneumoniae* in mice and rabbits.

2. MATERIALS AND METHODS

2.1 S. pneumoniae

Pneumotropic clinical isolates of S. pneumoniae serotypes were constituting the first author collection. They were revived from the storage media, in liquid encriched media and recharacterized as gram positive cocci mainly in pairs, alpha hemoliysin producers, blie solubility positive, optochin positive, inulin fermentation. Positive results in KBOOB HI Strep Kit India and sertyped by DENKA SEIKEN CO.LTD [9,10,11].

2.2 Mouse Pathogenicity Test

A volume of 0.3 ml of a broth culture from *S. pneumoniae* containing 1x 10 to 7 CFU/ml. was injected intraperitoneally into mice. The injected mice were observed for the onset of respiratory signs over six hours. dead mice were eviscerated [11]. Blood, lung, liver and kiney were collected both for reisolation studies onto Gentomycin blood gar, growth were identified as in [12], and for histologic sections. Tissue blocks were processed for histology tissue sectioning and staining [13].

2.3 Rabbits Pathogenicity

A 24 hours Trypton Soy 5ml fresh broth culture was centriuged at 2500 rpm for three minutes. Pellte cells were washed with one ml saline. Cell counts were ratified to 1 x 10 to 7 CFU/ml. Then ,0.5 ml doses were inoculated intranasal in rabbits in an upright position for 5 minutes. Inoculated rabbits were obsereved for the onset of respratory signs over 24 hrs. post to the infection [14]. The diagnostic infection criteria for pneumonia were; body temp, movement ,mouth foam, and breath rate. Hisologic section as in [13], reisolation studies [15].

3. RESULTS

The onset of respiratory signs RS and the death end poinits DEP in mice and rabbits were indicating that mice were more vunerable to infection with *S. pneumoniae* than rabbits, Table 1 .DEP for mice were up to 24 hours while that of rabbits were ranging from 4 to 14 days. There were an interserotype, Table 2 and intraserotype, Table 3 variations in pathogenicity of *S. pneumniae* serotypes both in mice and rabbits. Mice have shown moderate to severe while rabbits have shown mild forms of respiratory disease. Applying Koch's postulates on infected mice and rabbits, findings in, Table 4 were confirming the pathogenicity of S. pneumoniae to man. Both of the gross, histology, Table 5, Fig. 1 and Fig. 2. The sections showed suppurative neutrophilic cell infiltration in lung paranchyma, alveoli, and intrabronchal lumen. These tissue reations is consitant with brochopneumoniae rather than pneumonia. Reisolation studies of the infections in mice and rabbits were indicating that these animals reaching bacterimic state of infection since the bacteria have been resisolated from blood, lungs, and liver. Hence they are valid models simulating brochopnemonia in man.

Table 1. Interserotype variations in *S. pneumoniae* in mice and rabbits

Serotype	Mice/IP/0.3ml. 1x10 to 7 DEP in hours	Rabbits/05ml./1x10 to 7 DEP in days
1	12	4
2,6,8	18-20	6-8
7	24	14

Table 2. Intraserotype variations in pathogenicity of *S. pneumoniae* serotype 6 in mice and rabbit

Number of isolates	Mice/IP/0.3ml 1x10 to 7DEP in hours	Rabbit/IN/0.5ml 1x10 to 7,DEP in days
1	18	6
3	20	8
3	24	10
1	24	14

Table 3. Pathogenicity spectrum of *S. pneumoniae* serotypes in mice and rabbits

Serotype	Disese in mice	Disease in rabbits
6	Moderate respiratory disease condition	Mild respiratory disease condition
1,2,8	Moderate to severe respiratory disease condition	Mild respiratory disease condition
3,4,5,7	Mild to moderate respiratory disease condition	Mild respratory disease condition

Table 4. An autopsy reisolation culture studies for mice and rabbits infected with Serotype 1 and 6

Infected animal	Blood	Lung	Liver	Kidney
Mice; 1 ,6	+	+	+	+
Rabbit; 1, 6	+	+	+	+

Table 5. Histologic features of mice and rabbit lungs infected with *S.pneumoniae* serotype 1

serotype	Animal	Dose	DEPduration time	Histologic features	Conclusion
1	Mice	0.3 ml, IP, 1x10 to 7 CFU/ml.	12 hours	Neutrophilic infiltration in lung paranchyma, aveoli and intrabronchial inflammatory cells infiltration as compard to normal tissue contour in control	Bronchopneumonia
1	Rabbit	0.5 ml, intranasal, 1x10 to 7 CFU/ml.	4 days	Neutrophilic infiltration in the lung paranchyma, alveoli and, intrabronchial inflammatory cell infiltration and debries. Aa compared to normal tissue contour in controls.	Bronchpneumonia

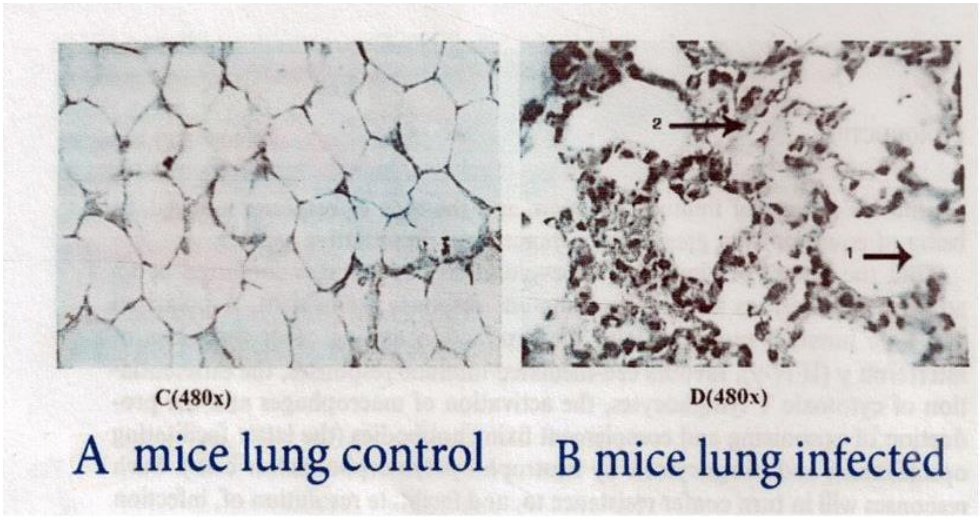

Fig. 1. A-normal mice lung tissue archticthure B-Infected mice lung tissue showing 1- Inflammatory cell infiltrate in lung tissue paranchyma ,2- Intrabronchial inflammatory cell infiltrate. 1 and 2. Consistent with brochpneumonia

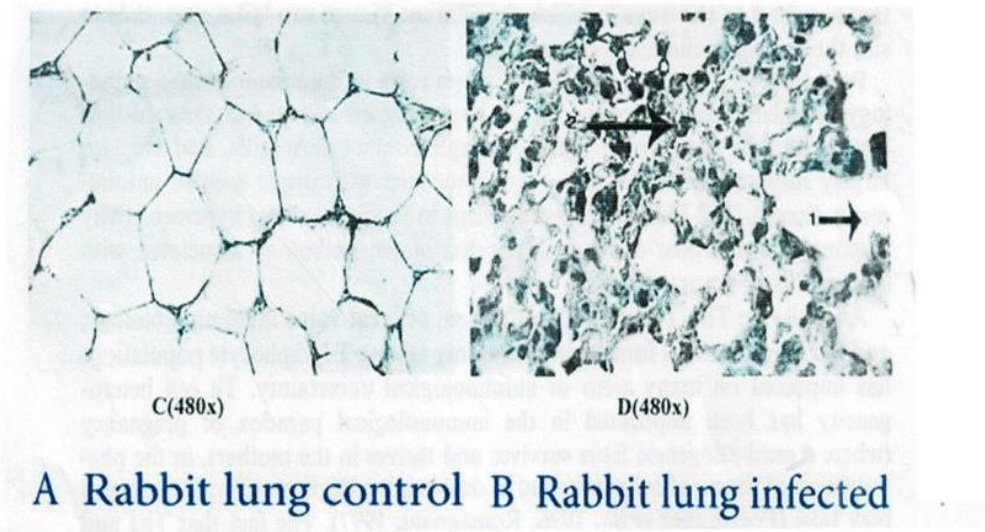

Fig. 2. A Normal rabbit lung tissue archcticture, B-infected rabbit lung tissueshowing 1 – Inflammatory cell infiltrate in lung tissue paranchyma,2-Intrabroncheolar inflammatory cell infiltrate. 1 and 2 consistent with bronchpneumonia

4. DISCUSSION

Digging through the current literature allover the world have shown that *S.pneumoniae* does have multiple pathoserotypes [1-9]. There were serotype dependent pathogenic potentials [3,4,5]. The present communication was tempting to uncover an inter and intraserotype variations in pathogenicity to mice and rabbits Tables 1-3. Mice have shown pneumonic disease similar to that of man with moderate to severe disease forms.The DEP of mice were 12 to24 hours.While that of rabbits were ranging from 4 to 14 days.Such pathogenicity spectrum is rather similar to that of man [14,16]. Serotype -1 was reported to be the most virulent, Table 5 , than other serotypes this was inline with results of other workers.But serotype 6 was of low virulence [17-21]. Infection of mice and rabbits with *S.pneumoniae* were either producing lober pneumonia or bronchopneumonia [20,21,22], results in Table 5 Fig. 1 was indicating bronchpneumonia.Results in this experimental settings are being novel in this area.

5. CONCLUSIONS

Different *S.pneumoniae* serotypes have shown different pathogenicity spectrum.There were an inter and intraserotype variations in pathogenicity to mice and rabbits as indicated by DEP duration time.Disease picture rather simulating that of man.Mice and rabbits were proved as valid laboratory animals for pathogenicity of this pathogen in human.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Authors hereby declare that No genrative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to –image generators have been used during writing or editing of this manuscript.

CONSENT

It's not applicable.

ETHICAL APPROVAL

1-The care, housing, handling and intervensions on both rabbits and mice were don following the international acts regulating the care, housing, handlings, and intervenstions.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Wang HE, Touromousis P, Webster S et al.Naturally occuring serotype 3 of *Streptococcus pneumoniae* strains that lack functional pneumolysins and autolysins hav attnuated virulence but induces localized protective immune responses. Plos ONE 2023;13(3): e0282884. DOI.10.1371/journal.pone.0282843.
2. Marquat ME. Pathogenicity of Streptococcus penumoniae: Cutting to chase on protease. Virulence. 2023;12 (1):766-787.
3. Xie Y-P, Li W, Fu Y, et al. Properties of mucoid serotype 3 *Streptococcus pneumoniae* from children in China. Front. Infect. Microbial.2021;11. DOI.10.3389/fcimb.2021..648040.
4. Jacques LC , Panagoituo S , Balt azar M , et al.Increased pathogenecity of penumococcal serotype 1 is driven by rapid autolysis and release of pneumolysins .Nat.Commun. 2020; 11: 1892 .
5. Minhas V. Pathogenesis of *Streptococcus pneumoniae* ;A molecular characterization. Ph.D Thesis.Scool of biological Dciences, University of Adelaide, Australia; 2020.
6. AL Najjar S , Sitthicaroenchai P , Gallup J et al.*Streptococcus pneumoniae* seotype 22F infection in respiratory syncytial virus infected neonatal lambs chances morbidity. bioRxiv; 2020. DOI.10.1101/2020.6.9.142034 preprint.
7. Nyongesa WM. Streptococcus pneumoniae, Prevalence, Antibiotic and Associated Risk factors among chlidren attendinggertrudes childrens hospital In Nairobi City Country Kenya. Ph.D Thesis in Medical Immunology in the School of Medicine,Kenytta University; 2015.
8. Brooks LRK, Mias GI. Streptococcus pneumonias virulence and host immunity: Ageing, diagnostics and prevention. Front. Immunol. 2018;9. DOI.10.3389/fimmu.2018.0.1366.
9. Levenson W, Chin-Hong P, Joyce EA, Nussbaum J, Schwartz B. Review of medical microbiology and immunology 15th ed. McGraw-Hill Lange, New York. 2018; 119-121.
10. Carroll KC, Hobden JA, Miller S, Morse SA, Mietzner TA, Detrick B, Mitchell TG McKerrow JH Sakanari JA. Jawetz, Melnick Adelbergs Medical Microbiology 27th ed. McGraw-Hill Lange, NewYork. 2016;222-224.
11. Tarja K, Esa R, Pentti U et al. Survival of Streptococcus pneumoniae, Hemophilus infleunzae and Moraxella chatrrhalis frozen in Skim milk-Trypton glucose-Glycerol medium. J. Clin. Microbiol. 2004; 42(1):412-414.
12. Sottile MI, Rytel WM. Application of counterimmunoelectrophoriesis in the identification of *Streptococcus pneumoniae* in clinical isolates. J. Cli. Microbiol. 1975; 2(3):173-177.
13. Bancroft JD, Stevens A. Theory And Practice of Histological Techniques. Churchill-Livingstone, New York.1982;117,
14. Nishmoto AT, Rosch JW, Tuomanen EI. Pneumolysin: Pathogenesis and therapeutic targets.Front.Microbiol. 2020; 11:1543. DOI.10.3389/fmicb.2020.01543.
15. Dennesen PJ, Vander SN, Kessels AG. Resolution of Infectious parameters after

antimicrobila therapy in patients with ventricular-associated penumonia. Am. J. Resp. 2001;163:1371-1375.
16. Hausdorff WF Feikin DR , Kingman K. Epidemiological difference among pneumococcal serotypes. Lancet. Infect. Dis. 2005;5:83-93.
17. Al Annee SRJ, McGee L, Jackson D et al. Association of serotypes of *Streptococcus pneumoniae* with disease severity and outcom in Adults: An international Study. Clin. Infect. Dis.2007 45:46-51.
18. Garua J, Calbo E. Capsular types predicting patients outcomes in pneumococcal bacterimia. Clin. Infect. Dis. 2007;45:52-54.
19. Alonso DE , Velasco AFM Verheul AMP et al.Epitope specificity of rabbit immunoglobulin G IgG elicited by pneumococcal type 23F synthetic oligosaccharide and native polysaccharide 23F Ig.Infect.Immun.1994;623): 799-808.
20. Tarini NMA, Astawa NM, Sukramma DM et al. The histological features and bacterial counts after exposure to *Streptococcus pneumoniae* serotypes 2,3,4, and 19F in lung of Balb/c mice.Bali.Med.J. 2019;8 (3):662-666.
21. Chiavolini D, Pozzi G, Ricci S.Animal models for *Streptococcus pneumoniae* diseases. Clin.Microbiol. Rev. 2008; 21(4):666-685.
22. Borsa N, Pasquale MG, Restrepo MI. Animal models for pneumococcal pneumonia. Int. J. Mol. Sci. 2019;20(17): 4220. DOI.10.3390/ihms.2017.4220.

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