EXEPRIMENTAL PREPARATOIN OF *PASTEURELLA MULTOCIDA* AUTO-GENUS VACCINE AND RELATED IMMUNE POTENTIAL

By

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B.Sc. Agric. Sci. (Biotechnology), Fac. Agric., Cairo Univ., 2008

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APROVAL SHEET

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ABSTRACT

Pasteurella multocida, a common pathogen in rabbits, is usually associated with upper respiratory disease. So, the aim of the present study was to isolate *Pasteurella* from clinical specimens from rabbits and to select strains for the preparation of an inactivated vaccine for active immunization of rabbits in the same area. A total of 150 nasal swab samples was collected from diseased rabbits (Seforabbit farm) from Giza Government showing symptoms typical for pasteurellosis and from dead rabbits showing lesions of lung congestion and pneumonia. Blood agar (BA) and MacConkey's agar were used as primary culture media for isolation of organisms from the samples. The collected samples were inoculated into tryptic soy broth medium for 24 h at 37 °C and reinoculated onto solid plate media containing 7 % sheep blood agar medium or Das medium. The bacteriological investigation revealed eight isolates of *P. multocida* isolates, four isolates of *Staphylococcus* spp., two isolates of *E. coli*, one isolate *Streptococcus* spp. and one isolate of *Proteus* spp. A total of 36 Swiss mice was used for pathogenicity testing of the *P. multocida* isolates. Mice died between 24 h and 48 h after exposure. To confirm *P. multocida* isolates, heart blood smears from dead mice were stained with Leishman's stain and revealed the bipolar staining. The heart blood and lung samples were inoculated onto blood agar plates. The two virulent *Pasteurella multocida* isolates used in the preparation of the intended vaccine were identified by capsular PCR technique and they were found to be of capsular type A. Non immunized, challenged rabbits (n=3) became dyspneic, depressed, anorectic and feverish. None of the immunized rabbits (n=4) died or showed any clinical signs. However, the non-immunized rabbits died 4 days post challenge.

Key words: Auto-genus vaccine, immune *Pasteurella multocida*, upper respiratory disease, rabbits.

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INTRODUCTION

Pasteurella multocida, is a common pathogen in rabbits, isusually associated with upper respiratory disease. Rabbits can become infected with *Pasteurella multocida* immediately after birth and the prevalence of *Pasteurella multocida* colonization increases with age until about 5 months, most adult rabbits are believed to infect with *Pasteurella multocida* (Manning *et al.*, 1989).

Pasteurella multocida is an important pathogen of rabbits used in biomedical research. The organism is ubiquitous in conventionally raised rabbits and caused a wide variety of inflammatory lesions including rhinitis, pneumonia, otitis media,pyometra, orchitis, abscesses, oculoconjunctivitis and septicemia (Flatt, 1974). Epizootics and enzootics of acute fatal pneumonia in rabbits have been reported (Flatt and Dungworth, 1979).

P. multocida is a bacterial pathogen of rabbits that generally accesses the body at mucosal surfaces; particularly the nasopharynx transmission is mainly by direct contact with nasal secretions from infected rabbits and may be the greatest when rhinitis induces sneezing and aerosolization secretions. Colonization and disease are influenced by factors related to both the host and the pathogen (Deeb *et al*., 1990).

Rabbits have a growing role as companion animals and are traditionally used as source of animal protein (Maria *et al*., 2012).

Respiratory disease due to *P. multocida* has been infected as a major cause of financial loss to the rabbit industry. More than 50 % of adult rabbits either dies or is culled to *P. multocida* infection. Rabbit colonies free of *P. multocida* infection have been established by