ROLE OF BRANHAMELLA CATARRHALIS IN LOWER RESPIRATORY TRACT INFECTIONS

Thesis

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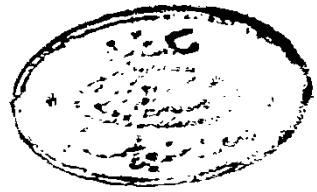

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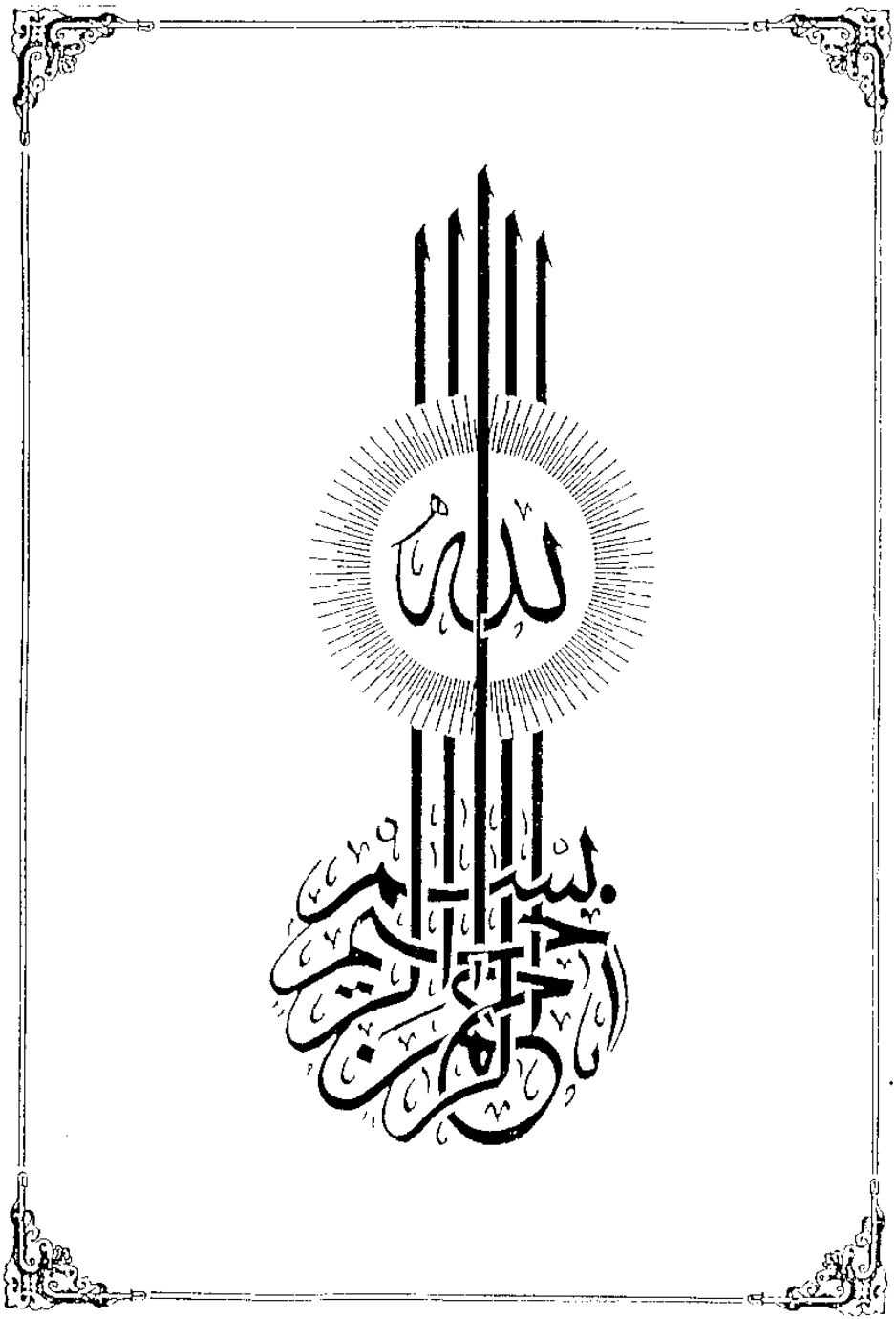

CONTENTS

INTRODUCTION
&
AIM OF THE WORK

INTRODUCTION

Branhamella catarrhalis previously known as Neisseria catarrhalis is an aerobic Gram-negative diplococcus. Although commonly assumed to be a commensal of the oropharynx, it is now recognised as an aetiologic agent of significant human diseases particularly in the immunodeficient host. It has been implicated in a wide variety of infections, including otitis media (Coffey et al. 1966), sinusitis (Brorson et al., 1976), meningitis (Goulon et al., 1973), septicaemia (Orsini 1954), and even endocarditis (Pollock and Holzman, 1976).

Branhamella has become increasingly recognised as an aetiologic agent of lower respiratory tract infections (Ninane et al., 1978, Johnson et al., 1981) particularly in patients with compromised pulmonary functions. Many reports have described the association of B.catarrhalis with acute bronchopulmonary infections (McNeely et al., 1976, Srinivasan et al., 1981, Louie et al., 1983, and Mcleod et al., 1983).

Several investigators have recently reported an increased rate of isolation of Beta-lactamase producing B. catarrhalis strains (Sweeny et al., 1985, Christensen et al., 1986).

AIM OF THE WORK

The aim of the study is to isolate B.catarrhalis from bronchopulmonary infections and to evaluate its potential role as an aetiologic agent in these lower respiratory tract infections. In addition the study is undertaken to determine the in vitro susceptibility of the organism to selected antimicrobical agents as well as the effect of beta-lactamase production on different antibiograms.

REVIEW OF LITERATURE

Neisseriaceae

The family Neisseriaceae contains Gram-negative aerobic rods and cocci. It includes four genera: Neisseria, Moraxella, Kingella and Acinetobacter (Howard 1987). Branhamella, initially considered a species of Neisseria and subsequently established as a separate genus, is now a subgenus of Moraxella (Koneman et al., 1988).

Classification:

Comparison of homology, both of composition and of sequence of deoxyribonucleic acid (DNA), ribonucleic acid (RNA) and protcins allows examination of organisms at the genetic level.

Analysis by both chemical and physiochemical methods shows that base composition is constant and characteristic for each organism. It is generally expressed in terms of the mole fraction of guanine and cytosine, that is (G + C/ G + C + A + T) expressed as a percentage. Values of percent G + C for different organisms vary from 25 to 75 (Johnson 1987).

If the DNAs of two organisms differ by several mole % G+C, that is good evidence that the two organisms do not belong to the same species (Johnson, 1987).

A] Gram negative cocci:

Study of the base composition of the DNA of the Gram-negative cocci and their genetic transformation showed that they could be divided into two groups. One possessing a G+C content of about 50 moles percent and included: N. gonorrhoeae, N. meningitidis, N. flavescens, N. subflava, N. sicca, and N. mucosa. The other comprising N. cotarrhalis, N. ovis and N. caviae having a G + C content of 40 to 44 moles percent (Catlin and Cunningham 1961).

The organisms in this second group were then transferred to a new genus Branchamella by Buchanon and Gibbons (1970). Later on, it was reclassified as a subgenus of Moraxella. This is based on their similar base content of DNA which is 40-45 moles percent (Bovre 1984).

Genus Neisseria:

Members of the genus Neisseria have a G + C content of about 50 moles percent. Their classification depends on fermentative ability, supplemented by antigenic structure and pigment formation (Wilson and Wilkinson 1984).

In addition to the two commonly recognised pathogens (N. gonorrhoeae and N. meningitidis), there are 11 species in the genus Neisseria which are: N. sicca, N. subflava, N. flavescens, N. mucosa, N. cinerea, N. denitrificans, N. elongata, N. canis and N. lactamica. N. subflava includes the organisms previously designated as N. subflava, N.

perflava, and N. flava. They are now considered collectively as one species because of their close genetic, morphologic and biochemical relationship (Vedros 1985).

B] Gram negative rods:

1. Moraxella:

Moraxella, a Gram-negative rod is one of the family Neisseriaceae and possesses G + C content of DNA which is 40-45 moles percent. The genus Moraxella includes 5 species which are: M. lacunata, M. nonliquefaciens, M. osloensis, M. phenyl pyruvica and M. bovis (Wilson and Wilkinson 1984).

Branhamella is a subgenus of Moraxella and comprises B. catarrhalis which is the only definitive member. B. caviae, B. cuniculi and B. ovis may be also included (Howard 1987).

2. Acinetobacter:

They are short Gram negative rods which possess a G+C content of 39-47 moles percent. There is only one single species in the genus which is A. calcoaceticus (Joklik et al., 1984).

3. Kingella:

They are gram-negative rods. The genus contains 3 species of which K. kingae is the most common one (Fallon and Young; 1989).

Neisseria

The genus Neisseria comprises two important human pathogens N. gonorrhoeae and N. meningitidis. In addition to these commonly recognised pathogens, a variety of saprophytic species are now classified in the Neisseria genus. Some species are normal inhabitants of the human respiratory tract and are usually considered non pathogenic (Doern 1988).

Branhamella was formerly classified in the genus Neisseria. It is now considered a subgenus of Moraxella and is referred to as Moraxella (Branhamella) catarrhalis (Bovre 1984). Although this designation may be taxonomically correct, yet it has not received widespread use and the terminology B. catarrhalis will be used. Branhamella is usually discussed with Neisseria species because its morphology and biochemical identification parameters most closely resemble those of Neisseria (Finegold and Baron 1986).

History:

The first member to be discovered was the gonococcus. It was observed by Albert Neisser in 1879 in the pus cells of patients with gonorrhoeae.

Weilchselbaum isolated the meningococcus from cerebro spinal meningitis in the year 1887. Pfeiffer described the

Micrococcus catarrhalis in 1896. He found it in the bronchioles and alveoli of children with bronchopneumonia. In 1902 Von Lingelsbreim described a number of Gram-negative cocci in the nasopharynx of healthy and diseased persons. These included the Micrococcus pharyngis siccus, the M. pharyngis cinereus, the Diplococcus mucosus, and the M. pharyngis flavus. Sarah Branham (1930) in the united states described a new member N. flavescens responsible for occasional cases of meningitis. Another one N. lactamica was described by Hollis et al., (1969).

General Characters:

Morphology:

Neisseria are aerobic Gram-negative cocci approximately 0.8 um in diameter. The members of the group differ somewhat in their morphology and arrangement.

Neisseria and Branhamella are recognised in a clinical specimen by the appearance of oval Gram-negative diplococci with flattened or concave opposing edges and by parallel long axis. They are either lying free in the specimen or inside polymorphonuclear leucocytes (Fallon and Young 1989).

Most members of the group are arranged in pairs, tetrads or small groups. Individual cocci are kidney shaped: when the organisms occur in pairs, the flat or concave sides are adjacent. One characteristic difference

which distinguishes Gram-negative from Gram-positive diplococci is the way in which the main axis of the oval is directed. The axis of the Gram-negative diplococci is always at right angle to the axis joining the two cocci, while in Gram-positive diplococci it is coincident with it. Gram-negative cocci divide at right angle to the axis joining them, so that the formation of tetrads is commoner than that with Gram-positive cocci (Wilson and Wilkinson 1984).

Morphology of the same organism may vary according to environmental conditions. In the body the meningococcus and the gonococcus present almost typical arrangement in the form of diplococci with flattened or slightly concave adjacent sides. In culture they appear as oval or spherical cocci with the typical diplococcus arrangement. In Gram stain most Neisseria are decolourized without difficulty but some tend to retain the violet stain. This occurs particularly when the organisms are arranged in groups or dense clumps (Wilson and Wilkinson 1984).

Capsules are demonstrable in some freshly isolated stains of meningocccci (Clapp et al., 1935), gonococci (Richardson and Sadoff 1977), in N. mucosa and occasionally in N. subflava.

Pili are present in gonococci, N. subflava and in small numbers on primary culture of meningococcus (Wistreich and Backer 1971).

The ultrastructure of the cytoplasm and the cell wall of the Neisseriae are similar to other Gram-negative organisms. The cell envelope is composed of three major elements: the cytoplasmic membrane, the rigid peptidoglycan layer, and the outer membrane which contains lipopolysaccharides, phospholipids, and proteins which are immunologically significant (Joklik et al., 1984).

Growth requirements:

Neisseria species other than N. meningitidis and N. gonorrhoeae grow on nutrient agar devoid of blood. N. meningitidis and N. gonorrhoeae require serum or blood or addition of other accessory growth factors as milk, animal fluids and certain vegetable extracts. Free iron is required for growth and starch, cholesterol or albumin should be added to media to neutralize the inhibitory effects of fatty acids (JZoklik et al., 1984).

The media mainly used is made up of chocolate agar or proteose peptone base to which various supplements such as haemoglobin, yeast extract, starch, glucose, cystine, trypticase or serum have been added (Thayer and Martin 1964, Brookes and Heden 1967).

A selective medium should be used for isolating the pathogenic Neisseria from sites that may be contaminated with bacterial flora as nasopharyngeal specimens. Selective media include modified Thayer Martin (MTM) agar, New York