Biology and Medicine

Suarez, Biol Med (Aligarh) 2016, 8:6
DOI: 10.4172/0974-8369.1000321

Short Commentary OMICS International

Optimal Conditions for *Streptococcus pneumoniae* Culture and for Polysaccharide Production for Vaccines

Norma Suarez*

Department of Biotechnology, Institute of Hygiene, Faculty of Medicine, University of the Republic, Uruguay

***Corresponding author:** Norma Suarez, Department of Biotechnology, Institute of Hygiene, Faculty of Medicine, University of the Republic, Uruguay, Tel: 598 2 487 12 88; E-mail: nsuarez@higiene.edu.uy

Received date: June 30, 2016; **Accepted date:** September 09, 2016; **Published date:** September 16, 2016

Introduction

Bacterial pathogens are responsible for the most severe forms of pneumonia, bronchitis and other respiratory tract infections (RTI), which have high morbidity and mortality rates especially among the elderly and children under 5 years [1]. *Streptococcus pneumoniae* accounts for the majority of community-acquired bacterial pneumonias and is also capable of causing meningitis, otitis media, and other infectious diseases [2].

Characteristics of Pneumococcal Capsular Polysaccharides

The capsular polysaccharides of pneumococci have been shown to be essential for their virulence [3]. Vaccines that include a number of the most prevalent capsular serotypes have been developed in recent decades [4]. The polysaccharide capsule is about 200-400 nm thick, and it has anti-phagocytic protective properties. The highly charged nature of the polysaccharides at physiological pH inhibits interactions between pneumococci and phagocytes [5-7]. The capsule is resistant to the activation of the alternative pathway of complement on its surface, thus avoiding opsonization.

For use as vaccine antigens, capsular polysaccharides must fulfill requirements of purity, uniformity and an accurate molecular weight. Chemical evidence of composition and structure is critical and has been reported for many *S. pneumoniae* serotypes [7-9]. However, vaccines based on pneumococcal capsular polysaccharides (CPS) are not effective in children below the age of 5 years [10].

Bacterial Polysaccharide-Protein Conjugates (TD Antigens)

Recently there has been growing interest in the preparation and purification of bacterial polysaccharide-protein conjugates because of the need for more effective vaccines against several different pathogens [11]. The conjugation of bacterial polysaccharides to carrier proteins converts capsular polysaccharides (CPS) into thymus-dependent antigens (TD antigens) with an improved vaccine potential. Examples of conjugated vaccines include the seven-valent PCV7 and PCV13 [12,13]. These vaccines have been successfully used for the control of a few pathogens, but the number and diversity of pneumococcal antigens involved make the control of RTI very difficult. Moreover, although the development of promising technologies for polysaccharide-protein conjugation may lead to the adoption of these improved vaccines, several issues remain to be solved. Further empirical studies are needed to determine the overall yield, purification procedures and characterization of these products.

Many bacteria are becoming resistant to antibiotics, forcing exploration of alternative approaches to control the burden of infectious diseases [14,15].

Polyvalent Bacterial Lysates (PBL) Oral Administration

Oral administration of polyvalent bacterial lysates (PBL) has been observed to have a beneficial effect on recurrent RTI in childhood (by decreasing frequency and severity of episodes), as well as on adult chronic obstructive pulmonary disease [16,17]. PBL are mechanically fractioned particulates or chemically degraded lyophilized cells derived from pathogenic bacterial strains responsible for RTI, which may contain extracts derived from various bacterial species including *Streptococcus pneumoniae* [18].

Polysaccharide Production and Economical Analysis

To achieve highly purified capsular polysaccharides for polysaccharide and conjugate vaccines, the selection of the culture medium is crucial [19,20]. The culture medium is also critical for the preparation of PBL. Although complex media containing biological materials, such as blood or soy extracts, are not yet banned for the production of vaccines and other biotechnology products, the use of defined media is encouraged for health and safety reasons and because they facilitate harvesting and quality control of polysaccharide antigens [21].

S. pneumoniae culture conditions are critical for the determination of time profiles of bacterial cell growth and polysaccharide production, which dictate optimal harvest time. Uniformity in the molecular weight distribution of the polysaccharides is desirable for consistency in the manufacture of conjugate vaccines [22,23].

Many authors have reported the influence of nutrient medium composition (e.g vitamin content, carbohydrates) on the synthesis of high molecular weight capsule polysaccharide of *S. pneumoniae* [24], highlighting the association between culture media and polysaccharide production. Bacterial cell cultivation requires special attention to media composition to obtain the desired yield and product quality. Scaled up fermentations are generally performed with inexpensive complex carbon and nitrogen containing supplements such as yeast extract, or other sources of vegetable origin. While these media are inexpensive, they give rise to undesirable variability in the production process, ultimately associated with higher costs [25]. Chemically defined media become more attractive and competitive in relation to traditional media when the simplicity and economy of downstream operations (separation, purification, characterization and so on) are taken into account. The costs and benefits of the growth media selected must be carefully balanced. Recently, Marthos et al. [24] reported a

rapid and economical procedure for identifying the best cultivation conditions using peptones. Their method, using fractional factorial experimental design, could be used to screen for the most important constituents in chemically defined media.

This short commentary outlines recent research work contributing to the field of pneumococcal production for vaccine use [20,26]. Improvement of culture conditions using fully defined culture media is indicated for achieving reliable, reproducible, controlled growth cultures for all serotypes of *S. pneumoniae*. Production of CPS for the purpose of synthesizing effective vaccines requires an approach to the design of fermentation and downstream separations that treats them as a single integrated process.

References

1. Nazzari E, Torretta S, Pignataro L, Marchisio P, Esposito S (2015) Role of biofilm in children with recurrent upper respiratory tract infections. Eur J Clin Microbiol Infect Dis 34: 421-429.
2. World Health Organization (2007) Wkly Epidemiol Rec 82: 93-104.
3. De Velasco A, Verheul AFM, Verhoef J, Snippe H (1995) Microbiological Reviews 59: 591-603.
4. Ogunniyil LA, Grabowicz M, Briles DE, Cook J, Paton JC (2007) Development of a Vaccine against Invasive Pneumococcal Disease Based on Combinations of Virulence Proteins of Streptococcus pneumoniae. Infect Immun 75: 350-357.
5. Kadiouglu A, Weiser JN, Paton JC, Andrew PW (2008) The role of Streptococcus pneumoniae virulence factors in host respiratory colonization and disease. Nat Rev Microbiol 6: 288-301.
6. Li Y, Weinberger DM, Thompson CM, Trzcinski K, Lipsitch M (2013) Surface Charge of Streptococcus pneumoniae Predicts Serotype Distribution. Infection and Immunity 81: 4519-4524.
7. Weinberger DM, Trzcinski K, Lu YJ, Bogaert D, Brandes A, et al. (2009) Pneumococcal Capsular Polysaccharide Structure Predicts Serotype Prevalence PLoS Pathog 5: 1-9.
8. Suárez N, Franco-Fraguas L, Texeira E, Massaldi H, Batista-Viera F, et al. (2001) Production of capsular polysaccharide of Streptococcus pneumoniae type 14 and its purification by affinity chromatography. Appl Environ Microbiol 67: 969-971.
9. Leal MM, Pereira DSG, Jessouroun E, Couto MAPG, Pereira N (2011) Investigation of cultivation conditions for capsular polysaccharide production by Streptococcus pneumoniae serotype 14. Electron J 14: 1-7.
10. Daniels CC, Rogers PD, Shelton CM (2016) A Review of Pneumococcal Vaccines: Current Polysaccharide Vaccine Recommendations and Future Protein Antigens. J Pediatr Pharmacol Ther 21: 27-35.
11. Suárez N, Franco-Fraguas L, Ferreira F, Massaldi M (2008) Improved conjugation and purification strategies for the preparation of protein-polisaccharide conjugates. Journal of Chromatography A 1213: 169-175.
12. Black SB, Shinefield HR, Hansen J, Elvin L, Laufer D, et al. (2001) Postlicensure evaluation of the effectiveness of seven valent pneumococcal conjugate vaccine. Ped Infect Dis 20: 1105-1107.
13. Van Deursen AMM, Van Mens PS, Vlaminickx BJM, De Greeff SC, DeMelker HE, et al. (2012) Invasive pneumococcal disease and 7-valent pneumococcal conjugate vaccine, the Netherlands. Emerg Infect Dis 18: 1729-1737.
14. Fowler T, Walker D, Davies SC (2014) The risk/benefit of predicting a post-antibiotic era: is the alarm working? Ann N Y Acad Sci 1323: 1-10.
15. Cullen L, McClean S (2015) Bacterial Adaptation during Chronic Respiratory Infections. Pathogens 4: 66-89.
16. Cazzola M, Anapurapu S, Page CP (2012) Polyvalent mechanical bacterial lysate for the prevention of recurrent respiratory infections: a meta-analysis. Pulm Pharmacol Ther 25: 62-68.
17. Pasquali C (2014) Enhanced Mucosal Antibody Production and Protection against Respiratory Infections Following an Orally Administered Bacterial Extract. Frontiers in Medicine 1: 41-50.
18. Rial A, Lens D, Betancor L, Benkiel H, Silva JS, et al. (2004) Intranasal Immunization with a Colloid-Formulated Bacterial Extract Induces an Acute Inflammatory Response in the Lungs and Elicits Specific Immune Responses. Infect Immun 72: 2679-2688.
19. Zhang J, Greasham R (1999) Chemically defined media for commercial fermentations. Appl Microbiol Biotechnol 51: 407-421.
20. Texeira EJ, Checa J, Rial A, Chabalgoity JA, Suárez N (2015) A new chemically defined medium for cultivation of Streptococcus 6: 54-62.
21. World Health Organization Technical Report Series (2009) 977.
22. Massaldi H, Bessio M, Suárez N, Texeira E, Rossi S, et al. (2010) Features of Bacterial Growth and Polysaccharide Production of Streptococcus pneumoniae serotype 14. Biotechnology and Applied Biochemistry 55: 37-43.
23. Gonçalves VM, Zangirolami TC, Giordano RLC, Raw I, Tanizaki MM, et al. (2002) Optimization of medium and cultivation conditions for capsular polysaccharide production by Streptococcus pneumoniae serotype 23F. Appl Microbiol Biotechnol 59: 713-717.
24. Marthos BV, Ferril ALS, Figueirdo DB, Zangirolami TC & Gonçalves VM (2015) Capsular polysaccharide production by Streptococcus pneumoniae serotype 1: from strain selection to fed-batch cultivation. Appl Microbiol Biotechnol 99: 10447-10456.
25. Restrepo A, Salazar B, Agudelo M, Rodriguez C, Zuluaga A and Vesga O (2005) Optimization of culture conditions to obtain maximal growth of penicillin-resistant Streptococcus pneumoniae. BMC Microbiology 5: 34.
26. Morais V, Surárez N (2016) Economic Evaluation of Streptococcus pneumoniae Culture Media. American Journal of Biochemistry and Biotechnology 12: 133-138.