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Therapeutic and Biotechnological Importance of Bacterial Vesicles and their Inhibitors

Jagannadham MV and Chattopadhyay MK*

Centre for Cellular and Molecular Biology (CSIR), Hyderabad, India

Introduction

Bacteria are unicellular organisms. But instead of living independently, each bacterial cell lives as a member of a society. They work together to bring benefits to the community. For this purpose they need to communicate with each other. Bacterial cells communicate among themselves using various tools. Outer Membrane Vesicles (OMVs) are one of the means of communication. They are small (diameter: 20 nm to 300 nm) bag-like structures released predominantly by gram-negative bacteria in the outer environment. Besides cell-to cell communication OMVs serve a number of other important purposes.

Historical Background

Though release of vesicles by the colon bacterium *Escherichia coli* was observed for the first time by D. G. Bishop and Elizabeth Work at Twyford Laboratories, London in 1965, it was two Indian scientists *viz*, S.N Chatterjee and J.Das, who published more concrete evidence of vesicle formation for the first time in 1966 and 1967. In course of their electron microscopic studies on the cholera pathogen *Vibrio cholerae* at the School of Tropical Medicine in Kolkata, they observed some blebs on the outer membrane of the organism [1]. In the face of scepticism expressed by other scientists on their observation, they continued their studies and established beyond any doubt that vesicle formation occurs in the logarithmic phase of the bacterial growth. They also proved that it is an active process and not the result of cell lysis or any structural deformity of the cells. Subsequently, vesicle formation was demonstrated in some other bacteria, even in tissues infected by pathogenic bacteria. A few years later, vesicle formation was demonstrated also in some gram-positive bacteria.

Functional Importance

Extensive investigations during the past few decades have revealed that vesicle secretion is an integral part of survival of gram-negative bacteria. So far, it has not been possible to isolate a variant of any gram-negative bacterium that does not produce the OMVs. The OMVs play a significant role in cell-to cell communication. The vesicles help the producer organism to acquire nutrients from the surroundings. This is how pathogenic bacteria obtain iron in an iron-deficient environment encountered inside the human body. Vesicles play a major role in secretion. They help bacteria in secreting more than one protein at a time. Besides proteins, they also transport RNA, secreted by the organism [2]. They foster bacterial virulence by carrying virulence factors (like enzymes) the target which might be located far away from the site of infection. Vesicles transport hydrophobic proteins. Transport of these molecules in the aqueous environment of human body might be problematic otherwise. Vesicles protect the producer from the attack of bacteriophages. When bacteria are challenged with some unfavorable conditions (e.g. high salt, oxidative stress) some proteins are denatured within the cells. Vesicles package these junks and transport them to the outer environment thus relieving the cell of the toxic burden. Vesicles also protect bacteria from antibiotics in various ways. In some cases, they carry antibiotic-inactivating enzyme. They are also found to carry the gene that encodes antibiotic-inactivating enzyme. Following incubation with such vesicles, recently antibiotic-sensitive cells of a bacterium were found to turn antibiotic-resistant by a group of researchers working at a laboratory in Spain. In some cases they appear to package the antibiotic and remove it from the cell. In some other cases they enclose the antibiotic-inactivating enzyme and shield it from inactivation by anti-enzyme antibody, produced in our body. Vesicles produced by an organism were found to bind two membrane-active antibiotics at our laboratory. The drugs could not reach the target cells as a consequence.

Therapeutic Potential of Inhibitors of Vesicle Formation

It is evident from the foregoing discussion that the vesicles have a potential role in antibiotic-

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Correspondence:
Jagannadham MV, Centre for Cellular and Molecular Biology (CSIR), Hyderabad, 500 007, India; E-mail: jagan@ccmb.res.in

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resistance of bacteria. They also promote pathogenesis. Hence inhibitors of vesicle formation are likely to be helpful in clinical management of bacterial infections. A couple of years back, a team of researchers led by Professor Nobuhiko Nomura at the University of Tsukuba (Japan) reported the repressive effect of indole and some of its derivatives on vesicle production by *Pseudomonas aeruginosa*. However, potential of these compounds as therapeutic agents needs to be established through extensive investigations.

Use of the vesicles as vehicle for drug delivery

Vesicles could be used as a vehicle for drug delivery. Amino glycoside antibiotics (gentamicin, kanamycin, neomycin) are broad-spectrum antibacterial agents. But they are ineffective against some pathogens *viz*, *Shigella* spp., *Listeria* spp., and *Salmonella* spp. during their intracellular growth phase since mammalian cell membrane is impermeable to these drugs. It has been possible to deliver gentamicin in to cultured human cells through vesicles produced by *sighella flexineri*. Scientists are also looking into the possibility of delivering therapeutic proteins into the target cells using vesicles.

The Kinesis Spindle Protein (KSP) plays a crucial role in cell division. Hence inhibitors of KSP can arrest mitosis. Various KSP-inhibitors have been tested so far as potential anticancer agents. RNA-interference is a process that involves inhibition of gene expression using small double-stranded RNA. They are called small interfering RNA (si-RNA). Regression of tumour could be achieved in an animal model using some engineered vesicles with reduced toxicity to deliver si-RNA, specifically targeted to KSP, in a collaborative study involving 3 laboratories in Korea.

Vesicle-based vaccines

Bacterial OMVs carry and deliver proteins. This property can be exploited in the development of new vaccines. The OMVs contain immune stimulators like LPS, proteins and DNA. Hence they are good adjuvants. Vesicles derived from pathogens have been used for a long time in the development of vaccines against the respective organisms. Vesicle-derived vaccine was found to improve production of antibody against infection caused by *Burkholderia mallei*, the causative organism of glanders [3]. Investigations involving *Fransicella*, a zoonotic pathogen, revealed that the organism produced tube-shaped vesicles besides spherical vesicles. In contrast to the spherical vesicles, the tube-shaped vesicles are derived only from the outer membrane. Production of the spherical vesicles and tubular vesicles was found to be co-coordinately regulated in *F. novicid* and dependent upon the growth phase and composition of the medium [4]. Both types of vesicles were found to be effective as vaccine adjuvant [5]. Engineered vesicles displaying antigen-specific tumour on the surface, were found to be effective in immunotherapy of cancer in animal model [6]. Interferon-mediated effect of bacterial vesicle was found to eradicate tumour without any notable side-effect in animal model. The effect was found to be durable [7]. In general, the OMV- based vaccines developed from gram-negative bacteria use detergent extraction to minimize the amount of Lipopolysaccharide (LPS), which is toxic to host cells. Several other strategies are being developed to produce OMVs from mutant strains containing detoxified LPS. Various methods are used for the production of engineered vesicles and the efficacy of the preparation in generating the specific antibody is observed. A vesicle-based vaccine against the causative organism of meningitis was approved for clinical use in Europe some time back. The prospect of the use of wild type OMV vaccines against Men B had been studied since the 1970s. Pertussis (whooping cough) is a contagious bacterial infection characterized by severe coughing fits. It affects 16,000 people worldwide every year and claimed 61,000 lives in 2013. Earlier, killed cells of the causative organism (*Bordetella pertussis*) were used as a component of DTP (Diptheria-Tetanus-Pertussis) vaccine. It was associated with some undesirable side- effects. In 1980, an acellular vaccine was developed. However, resurgence of pertussis infection in the twenty first century called for further development. A vaccine, based on OMVs prepared by a collaborative work between two groups in Argentina, was found to be safe and also induce immunity against pertussis in mouse model. The efficacy of a vesicle-based vaccine against glanders has been recently demonstrated in mouse model [3]. Recently, vesicle-based vaccine has also been found to be effective in protecting mice from acute lungs infection caused by *P. aeruginosa* [8].

Other uses of the vesicles

The efficacy of a multi-enzyme catalyzed process is dependent on the assembly of the enzymes. In an investigation conducted at the University of Delaware (USA), it has been possible to achieve a 23-fold increase in glucose production by hydrolysis of cellulose using three enzymes assembled on bacterial vesicles, compared to the production that could be achieved using free enzymes. Hence, it is obvious that bacterial vesicles have immense potential to be used for various therapeutic and biotechnological purposes. Large scale investigations appear to be the need of the hour to exploit these potentials.

References

1. Chatterjee D and Das J. Electronmicroscopic observations on the excretion of cell-wall material by vibrio cholerea. J Gen Microbiol. 1967;49:1-11.
2. Dauros-Singorenko P, Blenkiron C, Phillips A, Swift S. The functional RNA cargo of bacterial membrane vesicles. FEMS Microbiol Lett. 2018;365(5).
3. Norris MH, Khan MSR, Chirakul S, Schweizer HP, Tuanyok A. Outer membrane vesicle vaccines from biosafe surrogates prevent acute lethal glanders in mice. Vaccines (Basel). 2018;6(1).
4. Sampath V, McCaig WD, Thanassi DG. Amino acid deprivation and central carbon metabolism regulate the production of outer membrane vesicles and tubes by Francisella. Mol Microbiol. 2018; 107(4):523-541.
5. McCaig WD, Koller A, Thanassia DG. Production of outer membrane vesicles and outer membrane tubes by Francisella novicida. J Bacteriol. 2013;195(6):1120-32.
6. Grandi A, Tomasi M, Zanella I, Ganfini L, Caproni E, Fantappiè L, et al. Engineered synergistic protective activity of tumor-specific epitopes in bacterial outer membrane vesicles. Front Oncol. 2017;7:253.
7. Kim OY, Park HT, Dinh NTH, Choi SJ, Lee J, Kim JH, et al. Bacterial outer membrane vesicles suppress tumor by interferon-γ-mediated antitumor response. Nat Commun. 2017;8:626.
8. Zhang X, Yang F, Zou J, Wu W, Jing H, Gou Q, et al. Immunization with Pseudomonas aeruginosa outer membrane vesicles stimulates protective immunity in mice. Vaccine. 2018;36(8):1047-54.