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Review

Treatment of infectious disease: Beyond antibiotics

Anshul Nigam[a,*,1], Divya Gupta[b,1], Ashwani Sharma[c]

[a] IPLS Building, School of Life Science, Pondicherry University, Puducherry 605014, India
[b] Department of Biotechnology, Mangalayatan University, Beswan, Aligarh, Uttar Pradesh 202145, India
[c] Computer-Chemie-Centrum, Universität Erlangen-Nürnberg, Nägelsbachstr. 25, 91052 Erlangen, Germany

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ABSTRACT

Several antibiotics have been discovered following the discovery of penicillin. These antibiotics had been helpful in treatment of infectious diseases considered dread for centuries. The advent of multiple drug resistance in microbes has posed new challenge to researchers. The scientists are now evaluating alternatives for combating infectious diseases. This review focuses on major alternatives to antibiotics on which preliminary work had been carried out. These promising anti-microbial include: phages, bacteriocins, killing factors, antibacterial activities of non-antibiotic drugs and quorum quenching.

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1. Introduction

Microbial ecosystem is highly vivacious system and each species tries to excel in competition, the intra species competition is also fierce. Microbes foray over each other through chemicals to overcome competition. Humans had exploited these chemicals to cure various kinds of infectious diseases. Following the discovery of penicillin in 1928 by Scottish scientist and Nobel laureate Alexander Fleming; antibiotics have come a long way to cure infectious disease (Bennett and Chung, 2001). Today more than 100 different kinds of antibiotics have been discovered. The antibiotics have been found to cure various kind of infectious disease caused by microbes, but the advent of drug resistance in them, also known as 'superbugs' has pose new challenges for researchers (Dong et al., 2007; Livermore, 2004a; Williams, 2002). The rise and spread of drug resistance is attributed to evolutionary selection against antibiotics and high human mobility across globe (Heinemann, 2000; Levy and Marshall, 2004). Few prominent examples of acquired drug resistance include methicillin resistant

* Corresponding author. Tel.: +91 413 2654520; mobile: +91 9486374847.
E-mail address: anshulnigam2006@gmail.com (A. Nigam).
[1] Equally contributing authors.

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Staphylococcus aureus, fluoroquinolone-resistant *S. aureus* (Kaatz, 2005), erythromycin resistant *Streptococcus pyogenes* and *S. pneumoniae* (Frimodt-Møller et al., 2001) and vancomycin resistant enterocci (Kayser, 2003). Microbial resistance against antibiotics is a serious global health issue and has been recognized by number of reviewers (Berger, 2002; Dancer, 2001; Levy, 2001; Livermore, 2004b). The levels of resistance had continue to rise ever since it was discovered in year 2000, the World Health Organization alerted that infectious diseases may become non curable owing to high levels of multiple drug resistant pathogens (World Health Organization; Press Release WHO/41. http://www.who.int, 2000).

The mechanism of antibiotic action may be owing to inhibition of protein synthesis, DNA damage and cell wall biosynthesis (Walsh, 2000). While antibiotic resistance is conferred in microbes through variety of mechanisms, it may arise through the selection of pre-existing types, species and variants (Livermore, 2003). The resistance may also arise through mutation or DNA transfer. Mutation(s) can confer resistance to the microbes against antibiotics through variety of mechanisms. It may alter the antibiotic target or reduce its permeability or increase its efflux (Van Bambeke et al., 2003) or might up regulate an antibiotic-inactivating enzyme or bypass an enzymatic pathway. Gene transfer through plasmids and transposons, can spread resistance horizontally. The gene bla_{TEM}, which encodes TEM-1 β-lactamase, is the most common ampicillin-resistance determinant and has spread widely through this mechanism (Livermore, 2004a). Few species incorporate DNA released from dead cells of related species, resulting in modification of their own genes, Penicillin resistance in *pneumococci* has mainly spread through this mechanism (Spratt, 1994).

This review focuses on the alternatives to the antibiotics on which scientific community has been looking forward for years to overcome the problem of drug resistance. Following are major classes of alternatives:

a. Phages
b. Bacteriocins
c. Killing factors in microbes
d. Antibacterial activities of non-antibiotic drugs
e. Quorum quenching

2. Phage therapy

Phage's represent distinguish set of viruses that infect bacteria. The earliest mention of phages dates back to 1896, Ernest Hankin, a British bacteriologist, reported that an unidentified substance that passes through bacterial filters possessed antibacterial activity. The observations of Hankin were further investigated by others microbiologist. It was Felix d'Herelle who coined the term bacteriophage and demonstrated its clinical utility in treating infection.

Among all the alternatives to antibiotics mentioned in the review, phages not only went to clinical trials but also were produced at large scale during 1940s. The phages were administered to humans (i) orally, (ii) rectally, (iii) locally, (iv) as aerosols or intrapleural injections, and (v) intravenously (Sulakvelidze et al., 2001).

Although presently phage therapy is out of fashion from all over the world but it still continues in Georgia (former Soviet Republic). This drop in phage therapy is majorly attributed to the fact that phage's were applied for therapeutic purpose even before being fully understood. The application of phages as antimicrobials was pushed to brink with the advent of antibiotics (Kutter et al., 2010)

2.1. Mode of action

Bacteriophages replicate follow two distinguish modules:

(A) *Lytic module*: It comprises of following steps:
  (B) Attachment
  (C) Injecting phage DNA into the bacterial cell
  (D) Synthesis of bacterial components terminates
  (E) Replication of phage DNA, and production of new capsids
  (F) Phage components are assembled and released (lysis) (Fig. 1).

(G) *Lysogenic module*:Steps I, II, IV and V are similar to those of lytic phase (i.e., attachment, injection and release). The III step involves integration of DNA into the host chromosome (lysogenization) which replicates along with host DNA for several generations (prophage). The prophage after several generations may break free from bacterial genome to induce cell lysis producing new phage particles (Fig. 1). Due to the long infection cycle, lysogenic phages are unsuitable candidates for phage therapy (Lorch, 1999). Phages impart their resistance to bacterial restriction enzyme through genome modification (Andriashvili et al., 1986).

2.2. Advantages and disadvantages

The phages had been produced at commercial level for few years for therapeutic purpose (Summers, 1999) but their efficacy had always being questioned (Eaton and Bayne-Jones, 1934; Krueger and Scribner, 1941). This may be owing to the fact that scientist involved in discovery of phages were over enthusiastic about its application as bactericidal agent had overlooked clinical data (Kutter et al., 2010). The most interesting phenomenon associated with phages is that of auto dosing. It happens because phages are self replicating inside the bacterium host (Abedon and Cameron, 2010).

High specificity is a major advantage as well as disadvantage associated with phages. Although it ensures minimal damage to health friendly micro flora (Skurnik et al., 2007; Gupta and Prasad, 2011) but at the same time it is necessary to identify disease causing bacterium, limiting their usage for presumptive treatment (Loc-Carrillo and Abedon, 2011). Similar constraints are not associated with antibiotics. Efforts are on to identify phages acting against broad spectrum of bacteria (Jensen et al., 1998; Melo et al., 2014) or to genetically modify them to enhance their spectrum of action (O'Flaherty et al., 2005; Merril et al., 2007). The major side effect associated with phage therapy is considered due to the release of endotoxins from bacteria lysed *in vivo* by the phages (Lorch, 1999).

Even though the phage therapy is applied in few geographical locations over the decades and lot of research had been carried out with clinical perspective. However, bacterial immunity in this scenario had never been explored. The bacterial immune response may be innate or adaptive. The former response is either mediated through restriction modification that aids in differentiating between self and foreign DNA on basis of methylation pattern or lack of machinery required for phage replication (Abedon, 2012). Albeit innate immunity in bacteria was discovered decades ago however discovery of adaptive immunity began in late 80s when CRISPR (clustered regularly interspaced short palindromic repeats) sequences were identified in *E. coli* (Ishino et al., 1987; Nakata et al., 1989). The linkage of CRISPR sequences with adaptive immunity, nonetheless, was established only in previous decade. It is interesting to note that CRISPR mediated immune response involves gene silencing mechanism and is also inheritable (Barrangou et al., 2007; Brouns et al., 2008; Hale et al., 2009). We emphasize that clinical manifestation of bacterial immunity over phages must be evaluated prior to their application as antibacterial agent.